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# METAZOAN PARASITES OF THE EASTERN COTTONTAIL RABBIT IN WESTERN KENTUCKY

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ABSTRACT: Forty-five eastern cottontail rabbits (Sylvilagus floridanus Allen), collected over a 15 mo period from two areas of western Kentucky, were examined for disease conditions, helminth and arthropod parasites. A single lymphosarcoma, two cutaneous papillomas and the following parasites were identified: Hasstilesia tricolor, Taenia pisiformis, Cittotaenia variabilis, Raillietina stilesiella, Obeliscoides cuniculi, Trichuris leporis, Longistriata noviberiae, Trichostrongylus calcaratus, Dermatoxys veligera, Odontopsyllus multispinosus, Cediopsylla simplex, Haemaphysalis leporispalustris, Ixodes dentatus, Amblyonma americanum, and Dermacentor variabilis. Simpson's Indices indicated that parasite communities were fairly diverse, with some degree of dominance exhibited by O. cuniculi, T. pisiformis and H. leporispalustris. The parasite faunas of rabbits from the two areas were similar. Hasstilesia tricolor was associated with the absence of kidney fat.

#### INTRODUCTION

Recent surveys of parasites infecting cottontail rabbits (*Sylvilagus* spp.) have indicated that particular species were capable of establishing some degree of dominance (Jacobson et al., 1978; Andrews et al., 1980). For the purpose of this investigation dominance refers to the most characteristic parasite of the host population, and not an action on the part of a particular parasite. If these endemic species, under suitable conditions, assume a pathogenic role, their effect on cottontail populations can be considerable (Sarles, 1932; Erickson, 1944; Smith and Cheatum, 1944; Jacobson et al., 1974).

Two geographically distinct populations of eastern cottontail rabbits (*S. floridanus* Allen) were sampled over a 15 mo period, beginning in the winter of 1979, in an effort to identify their metazoan fauna. Areas selected for this study, the Western Kentucky Wildlife Management Area (WKWMA) and Calloway County (CC), have shown a marked disparity in population trends during recent years. The cottontail population at WKWMA underwent a dramatic, sustained decline several years prior to this investigation. Total rabbit harvest decreased 87% and hunter return per man-day spent hunting decreased from 1.76 to 0.76. This decline could not be attributed to loss of suitable habitat. At the same time cottontails were abundant and populations stable in CC, as determined by land-owner survey and information supplied by the County Game Officer (Alexander, pers. comm.). Direct comparisons between population densities of the two areas were not possible because of differences in land use and management practices.

Incidental to the primary goal of this investigation, it was hoped that comparisons of the parasite communities from the two areas might yield some evidence of an active or relic pathogen, capable of supressing population levels of WKWMA cottontails. Jacobson et al. (1978) engaged a similar problem in a population of eastern cottontails at Fort Pickett, Virginia. Their results indicated that, in conjunction with an initial epizootic of tularemia, helminth parasites may have limited host growth rate and productivity. Unlike Fort Pickett, WKWMA does not have a previous history of tularemia epizootics.

#### MATERIALS AND METHODS

The study areas were located in the Jackson Purchase physiographic region. Calloway County is situated in southwestern Kentucky and is part of the Tennessee River Drainage Basin. Western Kentucky Wildlife Management Area, in northwestern Kentucky, is part of the Ohio River Drainage Basin. Soil composition of the two areas is similar, both have temperate climates with no distinct wet or dry season. Annual rainfall for CC and WKWMA are 1.22 m and 1.16 m, respectively (Humphrey et al., 1973; Humphrey, 1976).

Rabbits were collected along secondary roads during predawn and dusk with a shotgun or small caliber rifle and a spotlight. Immediately after shooting, rabbits were placed in the maw of muslin sacks and

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abdominal and pleural cavities were opened. Blood was drawn by cardiac puncture and aliquoted into untreated and heparinized vacutainer tubes. Adrenal glands were removed, the right gland was frozen for use in later studies and the left was stored in 10% buffered formalin for weight determination.

At necropsy rabbits were weighed and sexed. Eye lenses were removed and stored in 70% ETOH for age determination (Edwards, 1967). Samples of liver, small intestine and gross lesions were fixed in Bouin's, sectioned at 8  $\mu$ m, and stained with Harris' hematoxylin and eosin. One ml of heparinized blood was added to a tube containing 5 ml of 2% formalin, centrifuged and the sediment along with thick smears were examined for microfilariae. Two hematocrit tubes were filled with heparinized blood for determination of PCV. Kidneys and associated perirenal fat were removed and the amount of fat determined to the nearest 0.1 g (Riney, 1955).

Cestodes and trematodes were fixed in AFA and stained with Mayer's paracarmine. Nematodes were killed in warm glycerine alcohol and mounted in glycerine jelly. Arthropods were fixed in 70% ETOH. Fleas were cleared in 10% KOH and mounted. Sera were tested for antibodies to *Francisella tularensis* using a slide agglutination technique (Difco Laboratories, Detroit, Michigan 48232, USA).

Physiologic and parasitologic data were analyzed using SAS (SAS Institute, SAS Circle, Box 8000, Cary, North Carolina 27511, USA) packaged statistical tests. Variables were compared using Student's *t*-test and *F*-tests. Simple and multiple regression and correlation were used to evaluate associations between and among variables. Seasonal and site interactions were evaluated with Duncan's Multiple Range test. Associations were considered significant at P < 0.05.

Species diversity and assessment of dominance were determined with Simpson's Index (Holmes and Podesta, 1968). Similarities between the parasite fauna of CC and WKWMA were determined using percent similarity, also described by Holmes and Podesta (1968).

#### **RESULTS AND DISCUSSION**

A total of 45 cottontail rabbits, 27 from CC and 18 from WKWMA, were examined for disease conditions. The liver of one rabbit appeared mottled with white fibrotic tracts. Histologic examination suggested lymphosarcoma. Bell and Chalgreen (1943) reported a malignant lymphoblastoma in *Sylvilagus* sp. from Pennsylvania, involving the liver and kidney. Though a degree of hepatic dysfunction due to the extent of the sarcoma was evident, the rabbit appeared healthy.

Cutaneous papillomas of the forelimb were found on two occasions. Papilloma virus is readily transmitted by *Haemaphysalis leporispalustris* (Larson et al., 1936), which were found commonly on rabbits in this study. Papillomas are benign but are considered capable of interfering with normal movement (Andrews, 1969).

Caseating abscesses, most often found on the neck and chin, and believed to be the result of tick bite, were found on 76% and 33% of CC and WKWMA rabbits, respectively. Secondary bacterial infections were not evident.

All of the helminth and arthropod species recovered (Table 1) have been reported previously from *Sylvilagus* spp. in surrounding regions (Rozycki, 1941; Bell and Chalgreen, 1943; Moore and Moore, 1947; Ecke and Yeatter, 1956; McGinnes, 1964; Stringer et al., 1969; Jacobson et al., 1978). Collecting irregularities prohibited effective interpretation of seasonal trends and relationships of physiologic parameters to parasitemias.

Results of this investigation indicated that the metazoan fauna of rabbits in western Kentucky were diverse. Low Simpson's Indices (0.11) do not imply a concentraion of dominance in a single species. Moderate dominance was exhibited by Taenia pisiformis (0.44), Obeliscoides cuniculi (0.34), and Haemaphysalis leporispalustris (0.27) within their respective taxa. Parasite communities of the two areas showed a strong similarity (72%). The strongest similarity occured between arthropods (76%) while helminths were least similar (71%). All arthropods identified were recovered from both areas. The helminths Hasstilesia tricolor and Raillietina stilesiella were specific for WKWMA and CC, respectively. No significant differences in prevalences or intensities of infection with the remaining species were found.

Hasstilesia tricolor, recovered during the winter and spring of 1979, were associated with a complete absence of host perirenal fat. Uninfected rabbits from WKWMA and CC during the same period had an average of 0.3 g of kidney fat. Jacobson et al. (1974) observed a significant correlation between infection with H. tricolor and an emaciated condition in Virginia cottontails, noting that heavy infections are likely to occur during periods of environmental and physiological stress (winter and spring). Ledune (1933) reported that Ohio rabbits in poor condition were invariably infected with *H. tricolor* although trematodes also were found in healthy rabbits. It is not clear whether this trematode inhibits fat deposition or causes depletion of existing fat stores directly, by interfering with nutrient absorption, or indi-

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Parasite (location)	Prevalence	Mean intensity (SE)	Range
Trematoda			
Hasstilesia tricolor (SI) (77108) <sup> ,</sup>	16	2,405 (404.8)	884-3,536
Cestoda			
Taenia pisiformis (BC) (77107)	53	6 (1.4)	1-25
Cittotaenia variabilis (SI) (77109)	29	1 (0.1)	1-2
Raillietina stilesiella (SI) (77110)	11	21 (10.9)	3-63
Nematoda			
Obeliscoides cuniculi (ST) (77113)	44	13 (2.9)	1-36
Trichuris leporis (CA) (77115)	18	1 (0.1)	1-9
Longistriata noviberiae (LI) (77111)	11	42 (30.0)	3-132
Trichostrongylus calcaratus (SI) (77114)	9	12 (5.7)	3-28
Dermatoxys veligera (L1) (77112)	4	36 (27.0)	9-63
Siphonaptera			
Odontopsyllus multispinosus (SK) (77309)	22	7 (3.0)	1–27
Cediopsylla simplex (SK) (77310)	16	2 (0.8)	1–6
Acarina			
Haemaphysalis leporispalustris (SK) (77311)	76	20 (4.3)	1-78
Ixodes dentatus (SK) (77312)	27	6 (2.2)	1-24
Amblyomma americanum (SK) (77313)	25	6 (3.2)	1–34
Dermacentor variabilis (SK) (77314)	5	2 (0.5)	1-2

TABLE 1. Parasites recovered from 45 cottontail rabbits (Sylvilagus floridanus) from western Kentucky.

SI = small intestine, BC = body cavity, ST = stomach, LI = large intestine, CA = cecum, SK = skin.

<sup>b</sup> U.S. National Parasite Collection accession number (Beltsville, Maryland 20705, USA).

rectly by decreasing host resistance to other stresses.

Cysticerci of *T. pisiformis* were found primarily on the visceral mesenteries, areas surrounding the rectum, and occasionally the pleural cavity. Cysticerci were found year round. *Cittotaenia variabilis* and *R. stilesiella* were found only during the winter and spring months. Petechiae were associated with both cestodes, but their greatest impact may result from physical obstruction. Erickson (1944) found that a single adult *C. variabilis* was capable of occluding the small intestine. In equivalent numbers *R. stilesiella* are not likely to cause this problem, however infections with this cestode are often much greater, and in large numbers luminal obstruction is possible (Andrews, 1969). In this study no intestinal dysfunction was apparent in association with cestode infection.

Obeliscoides cuniculi characteristically embeds itself in the gastric mucosa and, depending on the severity of infection, has been associated with varying degrees of pathogenesis (Wiggins, 1977; Jacobson et al., 1978). Gastric hemorrhage was not seen in this study and infections were considered light in view of other reports (Harkema, 1936; Moore and Moore, 1947; Andrews, 1969). Petechiae were noted, however, in ceca infected with *Trichuris leporis*. *Longistriata noviberiae, Trichostrongylus calcaratus*, and *Dermatoxys veligera*, when found, were not associated with lesions.

Low prevalence and intensity of infestation

and the lack of any discernable lesions due to *Cediopsylla simplex* and *Odontopsyllus multispinosus* indicated these species pose no immediate threat to rabbits in western Kentucky; however their role in the transmission of viral and rickettsial diseases should not be overlooked (Kilham and Woke, 1953).

Direct effects of tick infestation, such as described by Rechav et al. (1980), could not be determined effectively. *Haemaphysalis leporispalustris* has been identified as the primary vector of tularemia in some areas and a potential vector of a number of other agents (Jellison and Parker, 1944; McCahan et al., 1962), antibodies to *Francisella tularensis* were not demonstrated in the present study.

In view of these results, and those discussed by Jacobson et al. (1978), it appears that subtle disturbances due to enteric parasites, i.e., *H. tricolor*, may have contributed to the decline in cottontail populations at WKWMA. Definite conclusions can not be made, however, until this host-parasite system is studied under experimental conditions.

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