

PATHOLOGICAL AND PHYSIOLOGICAL RELATIONSHIPS OF
PARASITIC DISEASE IN A SELECT COTTONTAIL
RABBIT POPULATION

by

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INTRODUCTION

In introducing this work, the question might rightfully be asked; why study parasitic disease of cottontail rabbits? Andrews (1969), in part, answers this when he stated, "scant data available on cottontail rabbit parasites in the southeastern United States emphasize the need for critical study in this section of the country. It seems quite possible that the decline in rabbit populations may be closely associated with parasitism". Further justification is given when the same author stated, "the literature is quite extensive on parasites and special disease entities of this host species. Unfortunately, most investigators have primarily confined their research to describing new parasites and elucidating life cycles, rather than investigating the pathogenicity of these parasites".

Other authors have discussed the need for disease research in wildlife populations. One of the earliest was Leopold (1939;339) when he stated, "there is a great need of interpreting the role of the disease factor in determining population density and its fluctuations . . . Disease not only kills game, but also its predators and buffers. Disease may affect distribution, density, sex ratio and fertility". McGinnes (1958) stated, "In spite of optimum environmental conditions, a wildlife population may be unable to increase or even maintain itself in the presence of disease". Herman (1969) expressed the need for intensive studies of relationships between disease and physiology, nutrition, population density, and other host factors.

Dorney (1963) called for integration of disease research with studies of animal behavior, home range, population dynamics and habitat preferences.

The major emphasis of this study was on the investigation of parasitic disease in a select cottontail rabbit population. In order to carry out this investigation, it was also necessary to develop techniques suitable for the control of parasites under laboratory and field conditions. The objectives of this study were as follows:

1. To determine the degree of parasitism present in a confined cottontail rabbit, *Sylvilagus floridanus*, population and to compare this with parasitism of wild populations.
2. To determine the effectiveness of antihelminth drugs and insecticide-generating collars in reducing parasitism of a confined cottontail rabbit population.
3. To determine pathological and physiological changes related to the reduction of parasitic burdens in a confined cottontail rabbit population.

LITERATURE REVIEW

Due to the extensive literature available on parasites of the cottontail rabbit, only the literature on parasite control measures and that which is pertinent to physiological and pathological effects of parasites on the cottontail rabbit is reviewed. For a more complete listing of literature involving host parasite records for the cottontail, see Andrews (1969).

Parasite Control Techniques

A few authors have investigated the use of drug treatments to control internal parasites of rabbits. Vail and McKenny (1943) suggested treatment of domestic rabbits with tetrachlorethylene to control stomach worms, *Obeliscoides cuniculi*, and oil of chenopodium followed by castor oil dosage for the pinworm, *Passalurus ambiguus*. Lucus (1969) found the enzyme-inhibiting drug Nitrorynol effective in eliminating *Fasciola hepatica* from European rabbits. By treatment with this drug, 12 out of 12 rabbits were cured of fluke infestation. Yuill (1964) treated cottontails with a number of drugs to reduce parasite burdens. He found the broad spectrum antihelminth drug, Rulene, to be unsatisfactory because it appeared to have toxic effects on rabbits. However, through the use of sulfamethazine and an experimental broad spectrum antihelminth drug (8304, American Scientific Laboratories), he was able to reduce coccidia and roundworms by 46 percent and 51 percent, respectively. He also used Nemural tablets and another experimental drug (9084, American Scientific Laboratories)

as anticestodal treatment. Through the use of the experimental drug, he was completely successful in eliminating tapeworms of the genus *Cittotaenia*. After treatment with these drugs, he found significant differences in cottontail reproduction, with 48 percent more young being produced by drug treated animals.

Lampio (1952) was able to control helminth parasites of rabbits in Finland by destroying winter fecal accumulations with quicklime. Dorney (1963) suggested habitat manipulation to encourage segregation of cottontails into distinct summer and winter ranges to reduce parasitic infections.

Several investigators have tried to reduce ectoparasite loads of wild animals. McGinnes (1958) sprayed a 100 x 100 foot rabbit pen with three applications of Toxaphene and Malathion. One pound of each insecticide was used in each application. He also dusted rabbits individually with Malathion and Rotenone. He was unsuccessful in recovery of treated rabbits. However, two mortalities were observed which he felt were due to Malathion poisoning. Yuill (1964) obtained unsuccessful results in his attempt at reducing *Wohlfahrtia vigil* myiasis in cottontails by dusting with the insecticide Co-Ral. Howe (1965) was also unsuccessful when he tried controlling winter ticks in elk by a ground emulsion or water application of insecticides and by the use of systemics. Miles and Wildcomb (1953) achieved reduction of fleas, but were unable to control the sticktight flea, *Echidnophaga gallinacea*, on small rodents with DDT dusting of 5 pounds per acre. The effectiveness of this dusting was for a 2 week period. Ryckman et al. (1953) were unable to eliminate more than 98

percent of the fleas of the California ground squirrel with burrow entrance application of the insecticides Heptachlor, Dieldrin, and Aldrin. Dieldrin, however, had undesirable side effects since dead squirrels and rabbits were found after its application. In a later work (Ryckman et al. 1954), these authors found these insecticides to be effective for a 9 month period. Barnes and Kartman (1960) used 10 percent DDT dust in insecticide bait-box stations to control fleas of diurnal rodents. When these were baited daily for twelve days, they reduced fleas strikingly in a 24 hour period, but achieved little residual control. However, a second DDT application, in conjunction with twice weekly baiting for 28 days, gave residual control effective after a 42 day period. Using this same technique, Kartman (1960) achieved close to complete control of fleas on *Microtis californicus* when bait boxes were dusted with 5 percent DDT powder over a 6 month period. Miller et al. (1970) tested two methods of application and five insecticides in attempting to control fleas of wild rodents and rabbits. They found significant reductions of flea indices with Sevin and Bay 29493, but Malathion, Dursban, and Diazinon did not significantly reduce fleas. The reductions these authors observed were only temporary and did not result in flea control. Clark et al. (1971) used the systemic insecticides Fenthion, Mirex, and Diazinon on laboratory caged cottontail rabbits, kangaroo rats, and cotton rats. Complete control of fleas could be obtained in rabbits by introducing the insecticides Fenthion and Mirex in feed. These animals had the choice of selecting treated or nontreated feed.

Diazion was only 64 percent effective when rabbits had a choice of food selection, and higher concentrations had to be given in what the authors referred to as a "no choice test" to achieve complete control.

Parasite - Host Relationships

Parasites with which this investigation concerns itself can be divided into three general categories. These are the arthropods, the helminths, and the protozoans.

Arthropods

Of the arthropod parasites, the class Acarina has been the most studied with respect to pathologic effects on host species. Linduska and Lindquist (1952) evaluate the relationship of ticks to wildlife as follows:

That while comparatively little is known of the relationship of ticks and wildlife, the widespread distribution, abundance of species, and immensity of numbers of ticks has led to some suspicions that populations of one group may be regulated by the other. Ticks hold special significance for wildlife in their capacity for disease transmission. Exclusive of this role, however, the mechanical effects of their feeding can be an important factor in predisposing animals to other pests and disease. Through sheer numbers they frequently reduce the vitality of their host to the point of occasionally being the direct cause of death.

There appears to be much evidence in support of this statement. Smith and Cheatum (1944) concluded that a dramatic decline of rabbits on Fisher Island, New York, was apparently due to the infestation of rabbits by *Ixodes dentatus* and, to a lesser degree, *Haemaphysalis leporis-palustris*. The pathological conditions associated with these infestations were described as being "anemic conditions of the lungs and kidneys [sic], pale and watery-appearing blood, and many small

pus pockets at the site of tick attachment". Bell and Chalgren (1943) found massive subcutaneous infection of a cottontail by the tick, *Ixodes dentatus*. They concluded the infection was apparently due to inflammatory swelling of the host around the hypostome of the tick, causing the parasite to be engulfed. Cheatum (1941), Bell and Chalgren (1943), Llewellyn and Handley (1945), and Linduska and Lindquist (1952) reported finding pyogenic disease (subcutaneous abscesses) in cottontails. This disease is caused by one or more species of bacteria of the genus *Staphylococcus* and is associated with ticks (Linduska and Lindquist 1952). Linduska and Lindquist (1952) reported on rabbit papillomatosis (rabbit horns), a virus disease frequently fatal for rabbits and spread mechanically by ticks. Reilly (1971) reviewed the pathology of tularemia in cottontail rabbits and the role of the tick as a vector of this disease. Mullmann and Woronecki () summarized the importance of ticks as vectors. They stated that ticks are known to transmit five groups of disease; rickettsial, bacterial, spirochetal, viral, and protozoal. Ticks can also produce a toxic paralysis.

Siphonaptera, like ticks, are also important in that they too are disease vectors. To date, there is little evidence that fleas have a significant pathological or physiological effect on their host. Linduska and Lindquist (1952) stated fleas are important to wildlife mainly as an annoyance.

Two other arthropod parasites which have been investigated in relationship to observed effects on their host are the myiasis causing flies, *Wohlfahrtia vigil* and *Cuterebra* sp. Yuill and Eschle (1963)

found that for the Great Lakes area, *Wohlfahrtia* myiasis caused up to 22 percent mortality in penned cottontails. Geis (1957) found three Michigan cottontails infected with *Cuterebra*. Two of these were so weak that they were caught by hand, and the third was dead, apparently as a direct result of *Cuterebra* infection. The same author found a highly significant increase in white blood cell count and also lower average body weights in rabbits infected with *Cuterebra*. Pelton (1968) found significant reductions in bone marrow fat content of cottontails infected with *Cuterebra*. Rabbits with *Cuterebra* larvae had a mean bone marrow fat content of 23.2 percent, those with emergence scars had a mean of 36.4 percent, and those without larvae or scars had a mean of 49.1 percent.

Helminths

Despite extensive literature on helminth host records and incidence of infestation in the cottontail rabbit, relatively few authors have reported on effects of helminths on their host. Clancy et al. (1940) represent one exception. They reported that 50 percent of the rabbits they found to be malnourished were also infected with three or more species of parasites.

Of the helminth parasites for the cottontail rabbit, the largest number of species is found in the class Nematoda. Andrews (1969) investigated the pathological effects of ten species of nematodes which he found infecting cottontails. With the single exception of *Gongylonema pulchrum*, he was unable to report significant pathological

effects from these worms. In *G. pulchrum* infections, he found a limited amount of damage associated with the migrations of these worms in the esophagus of the host. However, he concluded that because this species occurs in rabbits only occasionally it poses little threat to their populations.

The most frequently reported nematode parasite of the cottontail is *Obeliscoides cuciculi*. Only Herman and Jankiewicz (1943) and Morgan and Waller (1940) reported pathological effects due to this parasite. These authors reported a mild catarrhal gastritis associated with heavy infestations.

One other nematode parasite reported as a pathogen in the cottontail is *Trichostrongylus calcaratus*. Morgan and Waller (1940) reported catarrhal duodenitis associated with heavy infestations.

Andrews (1969) reported on infestation of rabbits by the intestinal fluke, *Hasstilesia* sp. Although he observed heavy fluke infestation, up to, and exceeding 10,000 worms, without gross pathological signs, he concluded:

It is probable that large numbers of flukes would have deleterious effects on the host. It seems likely that in severe cases, ulceration and hemorrhage might occur at attachment sites. These sites, similarly, should provide portals of entry for numerous pathogens with resultant microabscess formations. Large numbers of flukes imbedded among the villi undoubtedly would reduce the surface area of the villi, thereby interfering with the normal digestive process. Prolonged infections would likely result in thickening of the mucosa, which could interfere with absorption as well as the normal peristaltic action of the gut. The expected clinical signs should include bloody feces and general emaciation in severe cases, especially during stress conditions.

Of cestodes reported for the cottontail, only the cysticerci of *Taenia pisiformis* and tapeworms of the genus *Cittotaenia* were found to have been connected with pathological effects on their host. Whitlock (1939) doubted whether even very heavy infections of *T. pisiformis* would be lethal to its rabbit host. He felt that any harm done was probably that of weakening the rabbit and making it more susceptible to other diseases or predators. However, Andrews (1969) found this parasite to cause extensive liver damage from migrating larvae. He also suggested that, "cysticerci may cause extensive and perhaps irreparable damage to the host", including interference with vital body processes and reproduction. Clancy et al. (1940) reported on the parasite *Cittotaenia variabilis*. These authors found rabbits with heavy infestations of this parasite to be in extremely emaciated condition.

Protozoans

The most commonly reported protozoan parasite of the cottontail is the intestinal coccidia of the genus *Eimeria*. Waller and Morgan (1941, cited by Andrews, 1969) found lesions in the vermiform appendix and illeocecal valve caused by the parasite. McGinnes (1958) reported one rabbit so heavily infested by coccidia that blockage of the intestine resulted. Andrews (1969) found no evidence that coccidia were associated with debilitating or morbid processes.

Erickson (1946) reported on the protozoan parasite *Sarcocystis*. Cysts were found which composed up to half the muscle mass on some of the 79 cottontails which he investigated. Andrews (1969), however, found in his investigation that this parasite appeared to be of little significance to the general health of the host.

MATERIALS AND METHODS

A total of 35 male and 12 female cottontail rabbits was used in a 2 x 2 factorial experiment. The experiment was designed to test the effects of a broad spectrum antihelminth drug, 1-tetramisole hydrochloride (Tramisol, American Cyanamid), treatment and insecticide-generating collar treatment on internal and external parasite loads and select physiological measurements of a confined cottontail rabbit population. Insecticide-generating collars contained 2, 2-dichlorovinyl, dimethyl phosphate (Sergeant's Sentry Dog Collar).

All animals were trapped on the Radford Army Ammunition Plant, Montgomery County, Virginia, between September 15, 1972, and October 14, 1972. Prior to treatment, rabbits were weighed, sexed, tagged with metal ear tags, examined for ectoparasites, and a heparinized capillary tube of blood for packed cell volume determination was collected by puncture of a marginal ear vein. Rabbits were randomly assigned to one of four treatment groups in replicates, i.e. one in each treatment group before a second set was begun. One-half of the animals of each sex received the drug Tramisol, administered by stomach tube and syringe at a dosage of 8 mg. per kg. of body weight in distilled water solution. The remaining rabbits received a sham treatment, consisting of stomach tube administration of distilled water. Half of each of the above groups received insecticide-generating collars. The remaining half received a sham collar treatment. Insecticide collars were fastened with the

manufacturer's buckle, or by a steel rivet which was applied with a rivet gun ("Pop" Rivetool, USM Corporation). Sham collars were made from TV cable. These were similar in size and shape to the insecticide collars.

Immediately after treatment, each rabbit was placed in one of four one-quarter acre pens which had been randomly assigned to receive a particular treatment group. Commercial rabbit pellets and water were given ad libitum at several feeder stations placed within the pens.

Ninety days after introduction of the first animal, all animals remaining were recovered by trapping. In addition to the penned rabbits, samples of five wild rabbits were collected from two locations. One of these samples was collected on the Radford Army Ammunition Plant and the other was taken from the woodlot in which the pens were located.

Rabbits and their ectoparasites were killed in a chloroform atmosphere in a large white enamel bucket. Prior to death, blood slides were made for differential cell counts and capillary tubes of blood were collected for packed cell volume determination. This blood was collected by a puncture of a marginal ear vein. Additional blood to be used in serum protein and corticoid analysis was collected prior to death by cardiac puncture. Ectoparasites were recovered by brushing the fur of the rabbit with a stiff toothbrush over a white enamel tray and by careful visual examination of the skin and fur. Forceps were used to transfer recovered ectoparasites to individually labeled vials of 80 percent ethyl alcohol and 5 percent

glycerin solution.

Animals were next weighed and then necropsied. Endoparasites free in the body cavity or attached to the mesenteries were recovered and placed in 10 percent formalin. The amount of kidney, loin, and mesentery fat was noted by describing fat as being heavy, medium, light, or none. Adrenals, kidneys, spleen, liver, and eyes were collected and placed in 10 percent formalin. The gastrointestinal tract was then removed for dissection.

The stomach, small intestine, caecum, and large intestine were then separated and examined for helminths by use of the gravity flotation technique of Clancy et al. (1940). Due to time limitations, only those helminths visible to the naked eye were recovered. Helminths were first fixed in hot alcohol-formol-acetic fixative (AFA) and then placed in AFA in vials.

Packed cell volume was obtained by use of an Adams Readocrit centrifuge. The serum protein analysis was conducted by Biuret assay, as described by Bausch and Lomb Incorporated (1965). Serum corticoid levels were measured using the protein binding technique of Murphy (1967). Eye lens were processed and weighed according to techniques described by Rongstad (1966). Analysis of variance testing was conducted through the use of a computerized program. The program was a least-squares analysis of data with unequal subclass numbers by Walter R. Harney, Biometrical Services, Agricultural Research Service, U.S.D.A., Plant Industry Station, Beltsville, Maryland, as modified for this investigation by Charles W. Smart,

Computer Programmer, Division of Forestry and Wildlife Resources, Virginia Polytechnic Institute and State University. Simple correlation analysis was conducted using a computerized statistical analysis system developed by Anthony J. Barr and James H. Goodnight, Department of Statistics, North Carolina State University. In addition, some of the data were analyzed by a Duncan's multiple range test (Steel and Torrie 1960:107-109).

RESULTS AND DISCUSSION

Between December 5 and December 20, 1973, 31 of the original 47 rabbits were recovered from the enclosures. In addition, 11 rabbits were known to have died during the treatment period. Two of these died from unknown causes, eight were known or suspected avian predator kills, and one rabbit was suspected of being a mammalian predator kill. Five rabbits could not be accounted for.

Unless otherwise noted, the results presented here are from those rabbits which were collected alive at the termination of this experiment. Data collected on remaining rabbits can be found in Appendix Tables 1-6. These tables provide a complete listing of all data collected during the course of this experiment.

General Parasite Observations

Table 1 lists those species of parasites which were identified during this investigation. In order to meet the objectives of this investigation, it was not necessary to identify all parasites to genus or species. Because of this, and a limited amount of time in which to collect data, most parasites were identified only to class and grouped according to the location from which they were recovered on their host.

Ectoparasites and Insecticide Treatment

Data on ectoparasites recovered, or noted, on enclosure rabbits are shown in Table 2-4. Since a deliberate attempt was made to not disturb these parasites during the initial handling and treatment of

Table 1. Parasites identified in this investigation

Parasite	Identifying Authority
Diptera	
<i>Cuterebra buccata</i>	E. P. Catts, Univ. of Delaware
Siphonaptera	
<i>Cediopsylla simplex</i>	Author
<i>Odontopsyllus multispinosus</i>	Author
Acarina	
<i>Ixodes dentatus</i>	Author
<i>Haemaphysalis leporispalustris</i>	Author
Trombiculidae	Author
Trematoda	
<i>Hastelesia tricolor</i>	Author
Cestoda	
<i>Taenia pisiformis</i> (cysticerci)	Author
<i>Cittotaenia</i> sp.	Author
Nematoda	
<i>Trichuris leporis</i>	Author
<i>Obeliscoides</i> sp.	S. Pursglove Univ. of Georgia
<i>Trichostrongylus</i> sp.	S. Pursglove Univ. of Georgia
<i>Dermatoxys</i> sp.	S. Pursglove Univ. of Georgia
Protozoa	
<i>Eimeria</i> sp.	Author

Table 2. Pretreatment ectoparasite infection of 47 rabbits

Treatment	Number of rabbits	Fleas : percent : infected	Ticks : percent : infected	<i>Cuterebra</i> - : percent : infected	mean no. : of larvae
Collar + drug	12	25	75	25	0.38
Collar + no drug	12	17	75	20	0.20
No collar + drug	12	25	33	50	0.80
No collar + no drug	11	45	36	38	0.38

Table 3. Post treatment ectoparasite infection of 31 rabbits

Treatment	Number of rabbits	Fleas -		Ticks -		Total -	
		percent infected	mean : number	percent infected	mean : number	percent infected	mean : number
Collar + drug	8	0	0.00	0	0.00	0	0.00
Collar + no drug	5	0	0.00	0	0.00	0	0.00
No collar + drug	10	20	0.20	30	0.50	50	0.70
No collar + no drug	8	50	0.63	12	0.13	62	0.70

Table 4. Mean square table for ectoparasites recovered from penned rabbits

Source	Degrees of Freedom	Mean Squares for Number of Parasites Recovered -		
		Fleas	Ticks	Total
Collar	1	1.427*	0.706	4.140**
Drug	1	0.452	0.218	0.042
Collar x Drug	1	0.452	0.218	0.042
Error	27	0.201	0.374	0.438

*p<.05

**p<.01

rabbits, only the presence or absence of fleas and ticks could be recorded at that time. The presence of maggots of *Cuterebra* sp. was observed only at the time of pretreatment examination. Both the number of larvae present and recent emergence pores were used in calculating means and infection incidence for this species. Pages 90 and 91 of the appendix contain additional observations on *Cuterebra*.

Insecticide-generating collars were completely effective in eliminating ectoparasites from the treated groups (Table 3). As can be seen from Table 4, this reduction was highly significant ($p < .01$).

Table 5 lists the ectoparasite data collected from the two wild samples with the data collected from the control group (the group which did not receive either the drug or insecticide-generating collars). Although higher numbers of ticks and fleas were collected from the wild rabbits, the only significant difference ($p < .05$) was between the number of ticks collected on the Radford sample and the number of ticks collected from the control group (Tables 5 and 6).

It seems possible that the lower number of ectoparasites recovered from the control group may have been due to one of two factors. One of these was the environment of the penned rabbits, and the other was the presence of rabbits which had been treated with insecticide-generating collars in an adjacent pen.

Support for the postulate that an environmental effect may have been operative on ectoparasite populations is provided by the fact that the treatment enclosures were located completely within a secondary

Table 5. Ectoparasite infection of wild rabbits compared to ectoparasite infection of control rabbits

Group	Number of rabbits	Fleas -		Ticks -		Chiggers
		percent infected	mean : number	percent infected	mean : number	percent infected
Control	8	50	0.63	12	0.13a	0
Woodlot	5	80	1.00	80	8.20a,b	0
Radford	5	80	1.60	80	31.60b	100

a, b Groups bearing different superscripts are significantly different from each other ($p < .05$)

Table 6. Mean square table for ectoparasites recovered from wild rabbits and control

Source	Degrees of freedom	Fleas	Mean squares - : Ticks :	Total
Group	2	1.46	1394	1485
Error	15	1.40	548	593

hardwood forest stand. This might be expected to have adverse effects on those species of parasites dependent on other habitat types for their development. Since the habitat within the Radford Army Ammunition Plant was old field succession and mixed conifer cover, ectoparasites introduced with their hosts into the enclosures might have had lower survival rates than in their previous habitat. The finding that wild rabbits collected from the same woodlot in which the enclosures were located, had higher ectoparasite densities might also be expected. These rabbits had access to forest edge, agricultural crop land, pasture, and overgrown clearings in addition to the woodlot.

It is equally feasible, however, that insecticide treatment may have reduced parasite loads of those penned rabbits which were not treated with insecticide-generating collars. One reason to suspect this is that the density of rabbits within the enclosure was quite high. This density would be expected to increase parasite numbers, since chances for host-parasite contact would be greatly increased. Other authors (Mohr 1971; Mohr and Adams 1963; and Forcum et al. 1969) have shown that positive relationships between rabbit densities and ectoparasite densities exist. A second reason for suspecting insecticide-induced reduction of ectoparasites on noninsecticide treated rabbits was that individual treatment groups were penned in adjacent enclosures. Since all that separated these groups was wire fencing, it was possible for insecticide and noninsecticide treated rabbits to come into relatively close contact with each other.

Another difference between the control and wild populations was the presence of Trombiculidae on the Radford Army Ammunition Plant sample. Because of their minute size and the large numbers of these parasites found (each rabbit had well over 100 chiggers attached within its ear canals), quantification of chigger numbers was not attempted. It is not surprising that these parasites were not recovered from the other wild sample or from the penned animals because, according to Sweatman (1971:17 and 48), these parasites are often seasonal in occurrence and are also more closely associated with a particular soil type or ecological situation than with a particular host species.

Helminths and Drug Treatment

Upon analysis, no significant differences were found in the number of cestode parasites recovered from individual treatment groups (Table 7 and 8). Because of its small size, no attempt was made to collect the intestinal fluke, *Hasstilesia* sp. However, 100 percent of the rabbits harbored this parasite. Additional observations on *Hasstilesia* can be found in an appendix to this manuscript.

Data collected on the nematode parasites recovered from penned rabbits are given in Tables 9 and 10. No significant differences were evident between treatment groups for nematodes recovered from the small intestine, large intestine, or caecum.

Drug treatment had a significant ($p < .05$) effect in reducing stomach nematodes and had a highly significant ($p < .01$) effect in reducing the total number of nematodes recovered from treated groups (Table 8). Since only those nematodes which were visible to the naked

Table 7. Cestode infections of 31 penned rabbits

Treatment	Number of rabbits	Small intestine cestodes -		Body cavity cysticerci -	
		percent infected	mean number	percent infected	mean number
Collar + drug	8	87.5	3.0	87.5	11.0
Collar + no drug	5	100.0	3.0	100.0	11.6
No collar + drug	10	100.0	5.4	80.0	20.3
No collar + no drug	8	87.5	4.1	87.5	10.1

Table 8. Mean square table for cestodes recovered from penned rabbits

Source	Degrees of freedom	Small intestine cestodes	Body cavity cysticerci
Collar	1	22.3	111.3
Drug	1	3.0	166.7
Collar x drug	1	3.0	211.1
Error	27	19.9	140.8

Table 9. Nematode infections of 31 penned rabbits

Treatment	Number of rabbits	Stomach nematodes -		Small int. nematodes -		Large int. nematodes -		Caecum nematodes -		Total nematodes -	
		: percent : infect.:	: mean : no.:	: percent : infect.:	: mean : no.:	: percent : infect.:	: mean : no.:	: percent : infect.:	: mean : no.:	: percent : infect.:	: mean : no.:
Collar + drug	8	50	0.9	0	0.0	0	0.0	37	0.4	75	1.2
Collar + no drug	5	100	5.0	40	1.0	40	0.6	40	1.0	100	7.6
No collar + drug	10	40	1.8	0	0.0	20	0.6	50	1.3	90	3.7
No collar + no drug	8	75	4.5	0	0.0	12	0.1	50	0.7	87	5.4

Table 10. Mean square table for nematodes recovered from penned rabbits

Source	Mean Squares for Number of Parasites Recovered -				
	Stomach	Small Intestine	Large Intestine	Caecum	Total
Collar ^a	0.33	1.82	0.03	0.83	0.09
Drug ^a	8.47*	1.82	0.03	0.01	117.09**
Collar x Drug ^a	3.69	1.82	2.10	2.51	39.74
Error ^b	1.10	0.44	0.68	2.20	14.62

^a1 degree of freedom

^b27 degrees of freedom

* p<.05

** p<.01

eye were recovered, the effect of drug treatment may have reduced nematodes more than the data indicate.

Data on helminth parasites collected from the wild samples and compared to the control group are given in Tables 11 - 14. Higher nematode numbers were found in the large intestine and the caecum of the Radford sample. This difference was significant ($p < .05$). Since the Radford sample and the sample collected from the woodlot were from two different populations, it is not surprising that these differed in the number of nematodes recovered. However, since both the Radford sample and the penned rabbits were collected from the same wild population, differences between these two groups were unexpected. This difference may have been due to the age structure of the control group and that of the Radford sample. Support for this postulate comes from the fact that the control group was composed of older individuals, as determined by significantly ($p < .05$) heavier eye lens weights (Table 19). This may indicate that some acquired immunity to large intestine and caecum nematodes occurs in the cottontail rabbit. It could be expected that older rabbits might build up immunity to these parasites as the result of prolonged infection. Smyth (1961:27) and Chandler and Read (1961:27) document acquired immunity to helminth infections.

Protozoan Parasites

The protozoan parasite *Eimeria* sp. was noted in histological sections of the small intestine of one rabbit. However, because of the time that would have been involved, no attempt was made to quantify

Table 11. Cestode infection of wild rabbits compared to cestode infection of control rabbits

Group	Number of rabbits	Small intestine cestodes -		Body cavity cysticerci -	
		percent infected	mean : number	percent infected	mean : number
Control	8	100.0	3.0	87.5	10.1
Woodlot	5	80.0	1.2	80.0	12.2
Radford	5	80.0	1.8	100.0	8.6

Table 12. Mean square table for cestodes recovered from wild rabbits and control

Source	Degrees of freedom	Small intestine cestodes	Body cavity cysticerci
Group	2	15.8	16.4
Error	15	13.4	74.6

Table 13. Nematode infection of wild rabbits compared to nematode infection of control rabbits

Group	Number of rabbits	Stomach nematodes -		Small int. nematodes -		Large int. nematodes -		Caecum nematodes -		Total nematodes -	
		percent infect.	mean no.	percent infect.	mean no.	percent infect.	mean no.	percent infect.	mean no.	percent infect.	mean no.
Control	8	75	4.5	0	0.0	12	0.1 ^a	50	0.7 ^a	87	5.4 ^{a,b}
Woodlot	5	80	2.8	0	0.0	0	0.0 ^a	40	1.6 ^a	80	4.4 ^a
Radford	5	80	1.8	20	1.6	80	1.8 ^b	100	9.0 ^b	100	14.2 ^b

^{a,b} Groups bearing different superscripts are significantly different from each other (p .05).

Table 14. Mean square table for nematode numbers recovered from wild rabbits and control

Source	Mean Squares for Number of Parasites Recovered -				
	Stomach	Small Intestine	Large Intestine	Caecum	Total
Group ^a	12.00	4.62	5.38*	114.46*	154.28
Error ^b	11.97	3.41	0.65	16.58	49.86

^a2 degrees of freedom.

^b15 degrees of freedom.

* $p < .05$

protozoan parasites or to record their incidence.

Host Physiological Changes and Parasite Infection

No significant differences in eye lens weights, body weights, or organ weights were found between treatment groups (Tables 15 and 16). Means of blood measurements recorded are given in Tables 17 and 18. Packed cell volume, corticoid levels, and basophil percentages were not significantly different between treatment groups. Drug treatment significantly ($p < .05$) decreased lymphocyte and monocyte percentages, serum globulin fractions, total serum protein, and significantly ($p < .05$) increased neutrophil percentages (Table 18).

Previous studies with the drug Tramisol have shown that it is metabolized and eliminated from the mammalian body at a rapid rate and that drug residues are nondetectable after a three day period (American Cyanamid Company, 1971?:54). Because of this, it is believed that differences observed are related directly to reductions of nematodes in the drug treated groups.

Schalm (1965:459) presented evidence for the postulate that nematode reduction can affect levels of circulating lymphocytes. He associated the lymphocyte and its close relative the plasma cell with the function of host immunological response. These cells are thought to shed their cytoplasm into the lymph. The cytoplasm contains gamma globulin and, in the immunized animal, antibody. Since most nematodes must penetrate host tissue in some phase of their life cycle (Chandler and Read, 1961:27), it seems probable that a relationship between host immunological reaction and nematode presence would

Table 15. Mean eye lens weights, body weights, and organ weights of 31 penned cottontail rabbits

Treatment	Number of rabbits	Eye lenses (mgs)	Body - initial (gms)	Body final (gms)	Adrenals (mgs)	Kidneys (gms)	Spleen (mgs)	Liver (gms)
Collar + drug	8	238	1223	1152	242	9.0	968	32.3
Collar + no drug	5	221	1158	1154	250	9.2	756	35.3
No collar + drug	10	229	1207	1213	253	9.0	939	34.5
No collar + no drug	8	260	1317	1218	270	9.1	909	35.2

Table 16. Mean squares table for eye lens weight, body weights, and organ weights of penned rabbits

Source	Degrees of freedom	Eye lenses	Body initial	Body final	Adrenals	Kidneys	Spleen	Liver
Collar	1	1607	3720	37063	1683	0.02	27843	8.3
Drug	1	354	3755	183	1171	0.15	106700	24.7
Collar x drug	1	4135	55395	435	146	0.01	59862	9.3
Error	27	1592	24742	21589	2401	1.53	330699	33.2

Table 17. Mean blood measurements from 31 penned cottontail rabbits

Treatment	Number of rabbits	Packed cell volume -		Serum corticoid (ng/100 mls)	Serum protein - (gms/100 mls)			Differential cell count - (percent)			
		initial	final		tot.	alb.	glob.	neut.	lymp.	mono.	baso.
Collar + drug	8	44.3	44.4	66.1 ^a	6.5 ^a	4.0 ^a	2.5 ^a	57.9	31.3	9.2	2.0
Collar + no drug	5	44.2	44.8	102.5	8.3	4.9	3.4	37.5	48.6	12.3	1.7
No collar + drug	10	43.1	45.8	69.0	5.4	3.4	2.0	54.0	34.7	8.4	3.4
No collar + no drug	8	44.5	46.2 ^a	53.9 ^a	7.8 ^a	4.0 ^a	3.8 ^a	41.2 ^a	45.1 ^a	12.6 ^a	1.8 ^a

^aOne blood sample was lost during processing, mean is calculated from measurements collected from 7 rabbits.

Table 18. Mean square table for blood measurements from penned rabbits

Source	Degrees of freedom	Packed cell volume -		Serum corticoid	Serum protein -			Differential cell count -			
		initial	final		tot.	alb.	glob.	neut.	lymp.	mono.	baso.
Collar	1	7.6	11.6	2396	4.3	3.8	0.0	50	32	3	17
Drug	1	31.2	0.8	286	31.2*	4.3	12.4*	7424*	5152*	402*	26
Collar x Drug	1	0.4	0.0	4520	0.7	0.2	1.6	378	297	11	11
Error	27	21.8	22.1 ^a	3252 ^b	7.1 ^b	3.2 ^b	2.4 ^b	1064 ^a	1024 ^a	53 ^a	20 ^a

^aOne less degree of freedom than indicated.

^bTwo less degrees of freedom than indicated.

* p<.05.

exist. This may also account for differences in serum protein which were recorded. Payne et al. (1965) also recorded significant changes in plasma proteins in response to parasitic infection. These authors found deer mice which were infected with *Cuterebra* larvae had significantly increased globulin fractions concurrent with significantly decreased albumin fractions. They postulated that parasite ingestion of albumin may have been connected with the differences observed. Since albumin fractions were not significantly different in the various test groups in this investigation, the differences in serum protein were probably due to host reaction to the parasites or to parasite by-products.

It is equally feasible that nematode reduction would also effect monocyte numbers. Schalm (1965:453) stated that, "monocytosis is evidence of chronicity, reflecting the need for destruction of pathogens too difficult for the neutrophils to handle and/or the need for removal of tissue debris in the chronic inflammatory process". The presence of nematodes might be expected to induce an elevated monocytic condition and this could account for larger numbers of these cells in rabbits which did not receive drug treatment.

The significant increase of neutrophil percentages in drug treated rabbits may be a response to changes that took place in numbers of other leucocyte cell types. If, in fact, host response to nematode reduction was that of shifting lymphocytes and monocytes out of peripheral circulation, then an increase in neutrophil percentages would be expected.

Data on eye lens weights, body weights, and organ weights collected from the two wild samples and compared to the penned control group are presented in Tables 19 and 20. Spleen weights for the woodlot sample were significantly heavier ($p < .05$) than those of the control and the Radford sample. Significant ($p < .05$) differences also existed between the eye lens weights of the control group and the two wild samples. The control population was therefore apparently composed of older individuals.

Blood measurements of the control and wild samples are shown in Tables 21 and 22. Packed cell volume of the woodlot sample was significantly ($p < .05$) higher than those of the control group or Radford sample.

Reasons for these differences are not clear. The differences in eye lens weights may be due to a seasonal difference in trap response between older and younger rabbits. Since the control group was originally collected 3 months earlier than the wild samples there is some support for this postulate. No reason can be given, however, as to why a seasonal difference in trap response between older and younger rabbits might occur.

Differences observed in spleen weight and packed cell volume are equally difficult to explain. It is possible that genetic differences may have been involved, since the woodlot sample was from a different population than the other two groups. Different stressors operating on the three groups may also have contributed to the differences between packed cell volumes and spleen weights.

Table 19. Mean eye lens weights, body weights, and organ weights of wild rabbits compared to control rabbits

Group	Number of rabbits	Eye lenses (mgs)	Body (gms)	Adrenals (mgs)	Kidneys (gms)	Spleen (mgs)	Liver (gms)
Control	8	260*	1218	270	9.1	909	35.2
Woodlot	5	206	1204	262	8.0	2242*	36.8
Radford	5	199	1188	227	7.6	1139	33.7

* Significantly different ($p < .05$) from the other two groups

Table 20. Mean square table for eye lens weights, body weights, and organ weights of wild rabbits compared to control rabbits

Source	Degrees of freedom	Eye lenses	Body	Adrenals	Kidneys	Spleen	Liver
Group	2	7390*	1354	2993	4.2	2876577*	11.5
Error	15	1426	26357	1266	1.7	450464	29.4

*
p<.05

Table 21. Mean blood measurements of wild rabbits compared to control rabbits

Group	Number of rabbits	Packed cell volume (percent)	Serum protein -			Differential cell count -				
			corticoid (ngs/100 mls)	tot. (gms/100 mls)	alb. (gms/100 mls)	glob.	neut.	lymp.	mono.	baso.
Control	7	46.2 ^a	54	7.8	4.0	3.8	41.2	45.1	12.6 ^a	1.8
Woodlot	5	52.9 ^b	86	9.1	5.3	3.7	53.0	38.8	6.4 ^b	1.8
Radford	5	46.9 ^a	72	6.5	3.7	2.8	55.7	30.5	8.7 ^{a,b}	3.8

^{a,b} Groups bearing different superscripts are significantly different from each other (p<.05).

Table 22. Mean square table for blood measurements of wild rabbits compared to the control rabbits

Source	Degrees of freedom	Packed cell volume	Serum corticoid	Serum protein - tot. : alb. : glob. :			Differential cell count - neut. : lymp. : mono. : baso.			
Group	2	73.0	2057	8.3	3.8	1.6	1457	1251	234	28.5
Error	14	10.5	2138	9.8	4.8	3.0	622	680	72	17.6

Peterson (1960) postulated that a decreased spleen weight was a sensitive indicator of stress in cottontail rabbits. It is possible that the woodlot sample may have been under less stress than the other two groups. Confinement at high density could be considered a stressor operating on the control group that was not operative on the woodlot sample. Significantly higher numbers of large intestine nematodes and caecum nematodes, along with chigger infestation and higher ectoparasite densities, were additional stressors operating on the Radford sample to which the woodlot sample was not exposed.

Relations of Fat Condition to Confinement and Parasites

No significant differences were found between fat indices recorded for individual treatment groups, or between fat indices recorded for penned and wild rabbits (Table 23). Although not significantly different, the penned rabbits did have lower fat indices than wild rabbits. This may have been due to the stress of confinement for the penned rabbits, or it could have been related to differences in diet between the confined and wildlife populations.

Differential Cell Count: A Comparison With Previous Studies

Since much of the significance reported in this investigation involves differential cell count, it appears appropriate to compare results obtained here with those obtained by other investigators. This comparison is made in Table 24.

As can be seen by Table 24, eosinophils were not recorded in this investigation. Vande Vusse (1964) described this cell for the

Table 23. Body fat condition of 31 penned and 10 wild cottontail rabbits

Group	Number of rabbits	Mean fat indices - ^a			
		kidney	loin	mesentery	total
Collar + drug	8	1.12	1.00	1.63	3.75
Collar + no drug	5	2.00	2.00	2.60	6.60
No collar + drug	10	1.60	1.70	2.20	5.50
No collar + no drug	8	1.37	1.25	1.62	4.25
Wild woodlot	5	2.40	2.20	2.20	6.80
Wild Radford	5	2.40	2.40	2.80	7.60

^aIndices were calculated by assigning a value of 3 to fat described as heavy, a value of 2 to fat described as medium, a value of 1 to fat described as light, and a value of 0 when no fat was present.

Table 24. A comparison of differential cell counts recorded in this investigation to those reported by previous investigators

Number of rabbits	Differential cell count percentages -					Authority
	neut.	lymp.	mono.	baso.	eosi.	
2	47.2	44.6	5.9	0.8	1.5	Musacchia et. al. (1955)
19	40.0	54.0	4.0	1.0	2.0	Youatt et. al. (1961)
170	55.2	36.1	7.0	0.8	0.9	Vande Vusse (1964)
38	45.0	47.0	6.0	1.0	0.5	Hurlbut (1967)
40	50.6	37.6	9.3	2.4	---	Present study

cottontail rabbit. According to his criterion, the eosinophil lacked distinct granules in Wright's stain preparation and was more intense in color than the neutrophil. In Giemsa's stain, this cell was decidedly acidophilic while the neutrophil took on little acidophilic reaction. Although leukocytes were noted which may have met Vande Vusse's criterion for an eosinophil, they were included in the neutrophil count in this study because they could not be distinctly differentiated from the neutrophil. This may have been partly due to the stain used, a Wright-Giemsa combination (LaMar Dip Stain, LaMar Laboratories, Inc.). However, another factor which made this investigator reluctant to attempt to differentiate eosinophils from neutrophils was the large difference between the eosinophil described by Vande Vusse and that which has been described for the domestic rabbit. Schalm (1965:296) described the domestic rabbit eosinophil as being a larger size and having intensely acidophilic granules 3 to 4 times the size of those of the neutrophil.

Despite the fact that the eosinophils could not be differentiated into a separate class, it is felt that this does not overly bias the results obtained. Under normal circumstances, eosinophils could be expected to constitute only a small percent of the total neutrophil count recorded. Also, if eosinophil percentages were different between treatment groups, this could only have affected the neutrophil percentage data. Since eosinophilia has been associated with parasitic infection, it might be expected that including eosinophils with neutrophil counts could have increased the neutrophil percentages observed in those

rabbits which were not drug treated. If anything, this would mean that differences between neutrophil percentages of drug and non-drug-treated groups were actually greater than the results indicate.

Although the percent of neutrophils and lymphocytes reported in this study are within the range of those reported by other authors, I observed higher monocyte and basophil percentages. The higher monocyte percentages recorded for penned rabbits, in comparison to the wild samples (Table 21), indicate that the stress of confinement may have had an effect on monocyte numbers. Vande Vusse (1964) has shown there may be a seasonal variation in differential cell count percentages. This may also have contributed to differences between the differential cell count reported in this investigation and those reported by previous investigators.

Basophil percentages reported in this study are more than double those reported by other authors. This was true for both the wild samples and the penned rabbits. Schalm (1965:293) reported on a number of authors recording basophil percentages for the domestic rabbit. Values obtained range from a mean of 1.2 percent to that of 8.0 percent. Schalm (1965:294) also reported that, "the rabbit is unique among the common laboratory animals in that the basophil leukocyte is commonly present in considerable numbers in peripheral blood".

Parasite Correlation Analysis

A simple correlation analysis was conducted on all parasite and host physiological data obtained during this study (Tables 25 and 26). A highly significant ($p < .001$) correlation of 0.57 was found

Table 25. Simple correlations between parasites and host body weight, eye lens weight, liver weight, spleen weight, kidney weight, adrenal weight, and fat index of 41 cottontail rabbits

Parasites	Body wt.	Eye lens wt.	Liver wt.	Spleen wt.	Kidney wt.	Adrenal wt.	Fat index
Ectoparasites							
<i>Cuterebra</i>	-0.03 ^a	-0.57 ^{a**}	0.22 ^a	-0.01 ^a	0.06 ^a	-0.22 ^a	0.24 ^a
fleas	0.27	0.12	0.12	0.24	-0.03	0.05	0.23
ticks	0.20	-0.03	0.12	0.28	-0.13	0.05	0.22
Cestodes							
small int.	-0.08	0.08	0.10	-0.37*	0.17	0.06	-0.17
body cavity cysticerci	0.15	0.13	-0.09	0.00	-0.07	0.20	-0.12
Nematodes							
small int.	-0.04	-0.27	-0.12	-0.13	-0.15	-0.11	0.31
large int.	0.04	-0.25	0.18	-0.13	-0.04	-0.18	0.31*
caecum	0.06	-0.20	-0.14	0.11	-0.34*	-0.04	0.20
stomach	0.01	0.13	-0.13	-0.06	-0.05	-0.03	-0.03
Total^b	0.25	0.02	0.02	0.17	-0.19	0.14	0.15

^aOnly includes data from 31 rabbits.

^bTotal does not include *Cuterebra*.

* p<.05

** p<.01

Table 26. Simple correlations between parasites and host blood measurements collected from 40 cottontail rabbits

Parasites	Serum protein -			Serum corticoid	Packed cell volume	Differential cell count -			
	tot.	alb.	glob.			neut.	lymp.	mono.	baso.
Ectoparasites									
<i>Cuterebra</i>	-0.03 ^a	-0.03 ^a	0.06 ^a	-0.05 ^a	0.25 ^b	-0.12 ^c	0.12 ^c	0.16 ^c	-0.18 ^c
fleas	0.11 ^d	-0.02 ^d	0.24 ^d	0.27 ^d	0.16 ^e	0.06	0.14	0.02	0.40*
ticks	0.16 ^d	-0.04 ^d	0.24 ^d	0.33 ^{d*}	0.12 ^e	0.12	-0.24	0.09	0.45**
Cestodes									
small int.	-0.15 ^d	-0.01 ^d	-0.18 ^d	0.02 ^d	0.02 ^e	-0.31	-0.26	-0.10	-0.14
body cavity cysticerci	0.02 ^d	-0.02 ^d	0.10 ^d	-0.06 ^d	-0.05 ^e	-0.02	-0.04	0.02	0.34*
Nematodes									
small int.	-0.11 ^d	0.01 ^d	-0.16 ^d	0.10 ^d	0.05 ^e	0.03	-0.07	0.19	-0.09
large int.	-0.05 ^d	0.01 ^d	-0.05 ^d	-0.08 ^d	0.17 ^e	-0.13	0.13	0.04	-0.08
caecum	-0.07 ^d	-0.03 ^d	-0.05 ^d	0.10 ^d	0.01 ^e	0.12	-0.18	-0.04	0.22
stomach	0.14 ^d	0.13 ^d	0.05 ^d	0.03 ^d	-0.16 ^e	-0.21	0.22	-0.08	0.03
Total ^f	0.12 ^d	0.03 ^d	0.21 ^d	0.28 ^d	0.07 ^e	0.13	-0.27	0.06	0.57***

^{a,b,c,d,e}Data from 29,31,30,39, and 41 rabbits, respectively.

^f*Cuterebra* not included in total.

* p<.05.

** p<.01

*** p<.001

between basophil percent and total parasite loads. Basophil percentage was also found to have a 0.45 ($p < .01$) correlation with ticks and 0.40 and 0.34 ($p < .05$) correlation with fleas and body cavity cestodes, respectively.

Although documentation of basophilic response to parasitic disease in wild animal species could not be found, it is believed that basophilia may be of use in diagnosing parasitic disease in the cottontail rabbit. Evidence for this is that eosinophilia has been found to be a common response to parasitic disease (Schalm 1965:455; Chandler and Read 1961:250) and that basophils are thought to respond to those same factors which cause changes in numbers of circulating eosinophils (Ham and Leeson 1961:185).

Because of the highly significant correlation which was found between basophils and parasite burdens, further investigations into this relationship may prove productive. The use of basophil percentages may have many applications in cottontail rabbit research and management. Examples of their possible use are to diagnose the general health of wild rabbit populations, to screen rabbits for parasitic disease prior to their use in research projects, or, since parasite densities appear to be related to host densities, to determine a relative index to rabbit population density. Since game census techniques are often difficult and time consuming, a density index of this nature might be particularly valuable in cottontail rabbit management. Other authors have proposed the use of parasite counts as a game census technique (Prestwood et al. 1972).

A highly significant ($p < .01$) correlation of -0.57 was found between *Cuterebra* parasitism and eye lens weights (Table 25). Bellig (1962) summarized the observations of five other researchers who recorded this parasite in Virginia cottontail rabbits. These investigators' findings support those of the present study, since they report *Cuterebra* larvae more prominent in juvenile rabbits. These findings suggest that the cottontail may develop immunity, or resistance, to this parasite. However, attempts by other investigators to induce *Cuterebra* immunity through repeated infection or large doses of larvae, in some host species, have failed (Capelle 1971:297).

A significant ($p < .05$) correlation of 0.33 was found between serum corticoid levels and numbers of ticks (Table 26). Although not significant, a correlation of 0.27 was also found between corticoid levels and numbers of fleas. These findings may indicate that ectoparasites can operate as significant stress agents on their hosts. The relationship between increased corticoid levels and various stressors appears well documented (Christian 1963, Schalm 1965:440, and Ham and Leeson 1961:772). Further support for a relationship between ectoparasites and stress was found in separate correlation analyses which were conducted on the penned rabbits and the wild rabbits. The penned rabbits which had very low ectoparasite numbers (the maximum number found on any one rabbit was 3), had non-significant correlations of -0.06 and -0.05 between corticoid levels, and ticks and fleas, respectively. However, wild rabbits, which had much higher numbers of ectoparasites (see Table 5, p. 22), had significant ($p < .05$)

correlations of 0.68 and 0.67 between corticoid levels, and ticks and fleas, respectively. Although the results presented here are by no means conclusive, they do suggest that further studies with corticoid-parasite relationships may be profitable in elucidating roles of parasitism in population regulation.

Four additional significant ($p < .05$) correlations were found between parasite presence and host physiological measurements. Correlations of -0.37 and 0.31 were found between small intestine cestode numbers and spleen weights, and cestode numbers and neutrophil ratio, respectively (Table 25). A correlation of -0.34 was found between caecum nematode numbers and kidney weight, and a correlation of 0.31 was found between large intestine nematodes and fat index (Table 25). No explanation can be given by this investigator for these correlations.

CONCLUSIONS

Pesticides have long been used for the improvement of man's own health and that of his domestic animals. Pesticide use has also been extended, on occasion, to wild animal species because of the role of wild animals as reservoir hosts for diseases and pests. However, no reports have been found that pesticides have been used to improve the health or well-being of wild animal populations.

This is not to say that the pesticides or techniques which were used in this study have implications for management of disease in wild animal populations. However, it is felt that these may have value in studying the effects of parasitic disease on wildlife populations. Once the effects of parasites and disease are known, it may then be practical to proceed to the next logical step; disease management in wildlife populations.

Although a number of authors have attempted to use pesticides in controlling ectoparasites of wild animals, it is this investigator's opinion that the techniques they have used are of limited value in determining the effects of ectoparasites on game species and other esthetically important wildlife populations. The reasons for this are many. Either they are too costly, in terms of time or money involved, they have undesirable environmental side effects, or they can only be used in conducting controlled or seminatural laboratory experiments. The use of the insecticide-generating collar circumvents all of these problems. These collars are relatively inexpensive, are

easily applied, and can be used under controlled, seminatural, or natural conditions.

In the literature review for this investigation, it was found that only one other author, (Yuill, 1964), used drugs in an experiment to determine the effects of parasites on a physiological aspect of the cottontail rabbit's biology. It is felt that the effectiveness of this technique, which was shown by both Yuill's study and the current investigation, could do much to further knowledge on the effects of parasitism in wild animal populations. Since stomach tube administration of drugs is a relatively simple task (see Appendix for a description of a portable restraining device used to administer drugs and collect blood samples), this technique also can be used equally well in controlled, seminatural, or natural conditions. When used in conjunction with the insecticide-generating collars, use of these drugs enables the investigator to selectively manage the particular parasitic disease conditions to be studied.

Many investigators have reported on parasites and parasitic disease in wild animal populations. However, relatively few of these have addressed the question which most needs to be answered. What effect does parasitic disease have on wild animal populations?

It is felt that the results obtained here indicate that parasitic disease may, under some conditions, operate as a limiting factor to cottontail rabbit populations. Changes in the generalized blood picture may significantly alter host defense mechanisms to other physical or social stress mechanisms and, as a result, increase

mortality. In addition, the presence of parasitic disease alone may constitute a significant enough challenge to induce mortality. The study by Yuill (1964) and the finding of a correlation between corticoid levels and parasitism in this investigation, suggests that parasites may also affect cottontail reproduction. Evidence that decreased reproduction can result from population stressors is reviewed by Christian (1963). These facts all point to a further need for more studies into the relationships of parasitic disease and wild animal physiology, particularly in the area of blood measurements.

SUMMARY

Forty-seven rabbits were used in a 2 x 2 factorial experiment to determine the effects of parasite reduction on host physiology. Insecticide-generating collars containing 2, 2-dichlorovinyl dimethyl phosphate and an antihelminth drug, 1-tetramisole hydrochloride, were used in reducing parasite loads. Thirty-one rabbits were recovered at the end of the treatment period. In addition to these rabbits, five rabbits were collected from each of two wild populations. All rabbits were examined for parasite burdens and various physiological measurements were recorded.

Insecticide-generating collars were found to be completely effective in eliminating ectoparasites from rabbits to which they were applied. Antihelminth drug treatment significantly reduced total nematode and stomach nematode numbers in rabbits which received drug treatment.

Analysis of parasites collected from wild rabbits showed that these rabbits had higher ectoparasite densities than the penned experimental rabbits which did not receive insecticide-generating collars. It was postulated that differences in ectoparasite loads may have been due to the environment of the penned rabbits or the presence of insecticide-generating collars on rabbits in pens adjacent to noninsecticide treated rabbits.

Reduction of nematode numbers in drug treated animals altered significantly the generalized blood picture. Drug treated rabbits had significant reductions in lymphocyte and monocyte percentages,

and significant increases in percentages of neutrophils. Drug treated rabbits also were found to have significant reduction in total serum protein levels and serum globulin fractions.

A correlation analysis was conducted on all parasite and physiological data collected during this investigation. A highly significant positive correlation was found between basophil leukocytes and total parasite numbers. It is suggested this measurement may be an important diagnostic tool for wildlife management and research. A highly significant negative correlation found between *Cuterebra* parasitism and eye lens weights indicates rabbits may acquire immunity to this parasite. A significant positive correlation was found between ectoparasite numbers and serum corticoid levels. Because of this it is felt that corticoid-parasite studies may be a fertile area for research into the effects of parasites on wild animal population regulation.

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APPENDICES

Appendix Table 1. Collection dates, sex, eye lens weights, and body weights of 57 cottontail rabbits

Rabbit number	Sex	Eye Lens (mg)	Collection Date		Body Weight	
			Initial	Final	Initial (gm)	Final (gm)
COLLAR/DRUG-TREATMENT						
264	M	287.6	9/06/72	12/08/72	1241	1108
301	M	106.0	9/07/72	9/11/72 ^a	731	668
259	M	237.0	9/09/72	12/09/72	1196	896
269	M	_____	9/21/72	_____ ^b	978	_____
298	M	270.2	9/24/72	12/14/72	1295	1259
210	M	167.6	10/01/72	12/11/72	995	1134
266	M	264.2	10/02/72	12/10/72	1221	1038
277	F	170.4	10/03/72	12/06/72	1110	1157
232	M	271.4	10/08/72	10/11/72 ^a	1161	916
233	F	194.0	10/10/72	12/12/72 ^c	1286	_____
224	F	246.0	10/11/72	12/18/72	1338	1315
279	M	260.2	10/12/72	12/14/72	1386	1308
COLLAR/SHAM-DRUG TREATMENT						
244	M	181.0	9/04/72	12/07/72	892	1042
242	M	241.0	9/09/72	12/14/72	1189	1085
256	M	_____	9/14/72	_____ ^b	1058	_____
300	M	_____	9/21/72	_____ ^b	823	_____
295	M	_____	9/23/72	12/01/72 ^d	1387	_____

a died of unknown cause

b unrecovered

c suspected mammal kill

d suspected avian kill

Appendix Table 1. continued

Rabbit number	Sex	Eye Lens (mg)	Collection Date		Body Weight	
			Initial	Final	Initial (gm)	Final (gm)
205	M	255.8	9/25/72	12/12/72	1266	1078
212	M	_____	10/05/72	_____ ^e	1252	_____
283	F	88.6	10/02/72	10/08/72 ^a	923	625
218	M	_____	10/05/72	10/20/72 ^a	1284	_____
241	F	_____	10/10/72	10/12/72 ^e	920	_____
215	M	183.6	10/12/72	12/08/72	1129	1200
271	F	243.4	10/14/72	12/06/72	1315	1368
SHAM COLLAR/DRUG TREATMENT						
206	M	262.2	9/05/72	12/20/72	1300	1312
222	M	184.4	9/07/72	9/15/72 ^d	996	922
292	M	172.0	9/11/72	12/07/72	885	1119
239	M	_____	9/21/72	12/20/72 ^e	1324	_____
204	M	246.0	9/23/72	12/15/72	1267	1285
253	M	260.0	9/25/72	12/14/72	1493	1505
282	F	161.2	10/03/72	12/06/72	931	1172
270	M	225.6	10/04/72	12/09/72	1243	1207
228	F	258.4	10/10/72	12/17/72	1420	1389
252	M	249.2	10/10/72	12/18/72	1308	1035
219	F	164.0	10/11/72	12/11/72	1008	1078

a died of unknown cause

d suspected avian kill

e avian kill

Appendix Table 1. continued

Rabbit number	Sex	Eye Lens (mg)	Collection Date		Initial (gm)	Final (gm)
			Initial	Final		
293	M	290.0	10/11/72	12/09/72	1215	1208
SHAM COLLAR/SHAM DRUG TREATMENT						
238	M	265.4	9/06/72	12/19/72	1330	1323
258	M	268.2	9/07/72	12/13/72	1213	1196
289	M	_____	9/18/72	_____ ^b	937	_____
268	M	254.8	9/23/72	12/17/72	1473	1389
208	M	_____	9/23/72	_____ ^b	1360	_____
278	M	260.8	9/27/72	12/08/72	1261	996
229	F	294.0	10/02/72	12/06/72	1375	1142
276	F	_____	10/04/72	_____ ^b	1481	_____
202	M	267.4	10/04/72	12/16/72	1275	1324
249	M	260.4	10/06/72	12/12/72	1435	1377
248	F	207.0	10/13/72	12/07/72	1174	999
WOODLOT SAMPLE						
C-1	F	190.0	_____	12/03/72	_____	1262
C-2	M	200.2	_____	12/03/72	_____	1198
C-3	M	264.8	_____	12/15/72	_____	1220
C-4	M	141.4	_____	12/22/72	_____	1049
C-5	M	234.2	_____	12/22/72	_____	1284

b unrecovered

Appendix Table 1. continued

Rabbit number	Sex	Eye Lens (mg)	Collection Date		Initial (gm)	Final (gm)
			Initial	Final		
RADFORD SAMPLE						
R-1	F	141.2	_____	12/22/72	_____	840
R-2	F	168.0	_____	12/24/72	_____	1131
R-3	M	252.0	_____	12/26/72	_____	1321
R-4	F	236.6	_____	12/28/72	_____	1322
R-5	M	195.2	_____	12/29/72	_____	1330

Appendix Table 2. Organ weights and fat indices for 41 cottontail rabbits

Rabbit number	Organ Weights				Fat Indices		
	Liver (gm)	Spleen (mg)	Adrenals (mg)	Kidneys (gm)	Kidney	Loin	Mesentary (category ^a)
COLLAR/DRUG-TREATMENT							
264	33.6	1670	258.2	10.8	0	0	1
259	18.8	340	259.9	6.4	0	0	1
298	39.4	1076	232.7	9.1	3	3	3
210	36.0	253	212.8	8.2	0	0	0
266	26.4	1011	261.9	9.5	0	0	1
277	36.7	419	205.0	9.4	3	2	3
224	31.7	379	227.2	9.4	0	0	1
279	35.7	2598	241.0	9.5	3	3	3
COLLAR/SHAM-DRUG TREATMENT							
244	36.9	702	242.2	9.7	3	3	3
242	32.9	919	286.3	9.5	3	3	3
205	29.5	794	248.7	8.0	0	0	1
215	35.4	774	275.5	9.2	2	2	3
271	41.6	593	243.4	9.7	2	2	3
SHAM-COLLAR/DRUG-TREATMENT							
206	32.5	478	331.2	11.5	2	2	2
292	26.5	1082	194.2	7.4	0	0	1
204	37.7	1318	202.0	9.0	3	3	3

a Fat index categories: none = 0; light = 1; medium = 2; and heavy = 3

Appendix Table 2. continued

Rabbit number	Organ Weights		Adrenals (mg)	Kidneys (gm)	Fat Indices		
	Liver (gm)	Spleen (mg)			Kidney	Loin	Mesentary (category ^a)
253	38.8	511	329.9	8.6	1	1	3
282	37.3	592	199.4	8.8	2	2	3
270	38.3	1838	318.7	10.0	3	3	3
228	42.1	347	211.6	9.2	2	3	2
252	32.8	670	268.6	8.7	0	0	1
219	32.2	1732	147.0	8.0	3	3	3
293	26.7	826	323.0	8.9	0	0	1
SHAM COLLAR/SHAM DRUG TREATMENT							
238	32.3	713	217.4	8.9	1	0	1
258	36.7	1639	285.2	8.3	2	2	3
268	40.0	694	299.6	11.9	3	3	3
278	25.0	517	317.6	7.3	0	0	0
229	28.4	1621	231.7	8.1	0	1	1
202	41.4	498	280.2	11.0	3	3	3
249	40.5	1048	300.5	8.5	2	1	2
248	37.3	542	225.7	9.0	0	0	0
WOODLOT SAMPLE							
C-1	32.3	1373	224.2	8.6	2	2	2
C-2	36.2	1918	248.6	7.0	2	1	1
C-3	35.8	2271	288.5	8.2	3	3	3

a Fat index categories: none = 0; light = 1; medium = 2; and heavy = 3

Appendix Table 2. continued

Rabbit number	Organ Weights		Adrenals (mg)	Kidneys (gm)	Fat Indices (category ^a)		
	Liver (gm)	Spleen (mg)			Kidney	Loin	Mesentary
C-4	35.3	3334	244.8	7.0	2	1	1
C-5	44.2	2312	302.6	7.3	3	3	3
RADFORD SAMPLE							
R-1	31.5	341	185.0	6.2	1	1	2
R-2	28.6	634	212.1	7.1	3	2	3
R-3	39.1	703	228.9	8.8	3	3	3
R-4	39.5	1504	272.7	8.9	2	3	3
R-5	29.9	2511	234.4	6.8	3	3	3

^a Fat index categories: none = 0; light = 1; medium = 2; and heavy = 3

Appendix Table 3. Packed cell volumes for 57 cottontail rabbits

Rabbit number	Initial (percent)	Final (percent)	Rabbit number	Initial (percent)	Final (percent)
COLLAR/DRUG-TREATMENT					
264	41.0	44.0	301	38.0	—
259	51.0	53.5	269	49.0	—
298	38.5	42.0	210	42.0	43.0
266	45.0	44.5	277	40.0	41.5
232	43.0	—	233	35.0	—
224	44.0	43.0	279	36.5	43.5
COLLAR/SHAM-DRUG TREATMENT					
244	42.0	42.7	242	41.5	39.0
256	39.5	—	300	53.5	—
295	43.0	—	205	45.0	45.0
212	50.0	—	283	56.0	—
218	52.0	—	241	48.5	—
215	47.5	51.0	271	45.0	46.5
SHAM-COLLAR/DRUG TREATMENT					
206	45.0	49.0	222	42.0	—
292	42.5	42.5	239	40.0	—
204	32.5	44.0	253	45.0	47.5
282	49.0	59.0	270	44.5	41.5
228	40.5	40.0	252	41.5	50.5
219	50.0	41.0	293	40.5	43.0

Appendix Table 3. continued

Rabbit number	Initial (percent)	Final (percent)	Rabbit number	Initial (percent)	Final (percent)
SHAM-COLLAR/SHAM-DRUG TREATMENT					
238	38.0	40.0	258	42.5	50.0
289	48.0	_____	268	49.0	49.0
208	46.0	_____	278	37.5	_____
229	44.0	44.5	276	42.5	_____
202	48.5	45.5	249	53.5	47.0
248	43.0	47.5			
WOODLOT SAMPLE					
C-1	_____	50.5	C-2	_____	52.0
C-3	_____	59.5	C-4	_____	48.5
C-5	_____	54.0			
RADFORD SAMPLE					
R-1	_____	49.0	R-2	_____	46.0
R-3	_____	47.5	R-4	_____	47.0
R-5	_____	45.0			

Appendix Table 4. Blood measurements from 41 cottontails

Rabbit number	Serum Protein			Serum Corticoid <u>ng per</u> <u>100 mls</u>	Differential Cell Count			
	Total (gms. per 100 mls)	Alb.	Glob.		Neut.	Lymph.	Mono.	Baso.
COLLAR/DRUG TREATMENT								
264	8.5	4.7	3.8	50.0	70.5	20.0	8.5	1.0
259	5.4	3.5	1.9	67.5	81.5	9.0	7.0	2.5
298	5.0	2.9	2.1	20.0	49.0	42.5	6.5	2.0
210	—	—	—	—	41.0	48.0	9.5	1.5
266	10.8	5.7	5.1	200.0+	73.0	9.0	15.0	3.0
277	3.8	2.5	1.3	30.0	51.5	39.0	8.5	1.0
224	6.4	4.2	2.2	80.0	43.5	44.0	11.0	4.5
279	5.3	4.4	0.9	15.0	53.0	39.0	7.5	0.5
COLLAR/SHAM-DRUG TREATMENT								
244	5.7	3.3	2.4	132.5	21.0	65.0	13.0	1.0
242	7.6	5.0	2.6	42.5	25.5	65.0	6.5	3.0
205	10.5	4.6	5.9	82.5	47.0	44.0	7.5	1.5
215	11.1	7.4	3.7	200.0+	54.5	25.0	20.0	0.5
271	6.5	4.4	2.1	55.0	39.5	43.5	14.5	2.5
SHAM-COLLAR/DRUG TREATMENT								
206	6.1	4.2	1.9	120.0	82.5	10.5	6.0	1.0
292	4.2	2.9	1.3	75.0	68.5	21.0	9.0	1.5
204	3.4	3.0	0.4	27.5	44.5	41.0	8.0	6.5
253	3.6	1.7	1.9	22.5	63.0	23.5	11.0	2.5

Appendix Table 4. continued

Rabbit number	Serum Protein			Serum Corticoid <u>ng per</u> <u>100 mls</u>	Differential Cell Count			
	Total (gms per 100 mls)	Alb. per 100 mls)	Glob.		Neut.	Lymph.	Mono.	Baso.
282	7.8	3.7	4.1	87.5	40.0	47.0	11.0	2.0
270	5.4	3.3	2.1	22.5	23.0	67.5	11.5	0.5
228	7.5	4.7	2.8	0.0	58.0	32.0	5.5	4.5
252	6.4	4.8	1.6	200.0+	82.5	13.0	4.5	0.0
219	3.7	2.8	0.9	27.5	42.0	49.0	6.5	2.5
293	5.4	2.9	2.5	107.5	36.0	42.0	10.5	11.5
SHAM-COLLAR/SHAM-DRUG TREATMENT								
258	11.0	2.9	8.1	35.0	34.0	43.5	21.0	1.5
268	13.4	9.0	4.4	82.5	31.0	55.5	11.5	2.0
229	5.4	1.9	3.5	52.5	44.0	44.0	9.0	3.0
202	10.5	7.4	3.1	82.5	33.5	54.5	9.0	3.0
249	3.7	1.8	1.9	40.0	33.0	55.0	15.5	1.5
248	2.8	1.1	1.7	0.0	64.5	23.0	11.5	1.0
WOODLOT SAMPLE								
C-1	8.4	4.7	3.7	115.0	46.0	48.5	4.0	1.5
C-2	6.0	3.7	2.3	25.0	38.0	56.0	6.0	0.0
C-3	9.7	5.1	4.6	72.5	59.0	35.0	3.0	3.0
C-4	12.3	7.4	4.9	132.5	51.0	32.5	14.0	2.5
C-5	9.0	5.7	3.3	77.5	71.0	22.0	5.0	2.0

Appendix Table 4. continued

Rabbit number	Serum Protein			Serum Corticoid	Neut.	Lymph. (percent)	Mono.	Baso.
	Total (gms per 100 mls)	Alb. per 100 mls)	Glob. per 100 mls)	ng per 100 mls				
RADFORD SAMPLE								
R-1	6.2	2.8	3.4	82.5	36.5	54.0	6.5	3.0
R-2	3.0	2.5	0.5	52.5	53.5	35.5	9.0	2.0
R-3	6.5	4.7	1.8	47.5	69.5	21.0	6.5	3.0
R-4	8.4	4.0	4.4	200.0+	54.0	20.5	15.5	10.0
R-5	8.4	4.4	4.0	50.0	65.0	21.5	6.0	1.0

Appendix Table 5. Ectoparasites noted or recovered from 57 cottontail rabbits

Rabbit number	Initial Examination			Final Examination		
	Fleas	Ticks	Cuterebra ^a	Fleas	Ticks	Chiggers
COLLAR/DRUG TREATMENT						
264	x		0	0	0	0
301	x	x	1	—	—	—
259		x	0	0	0	0
269			0	—	—	—
298		x	0	0	0	0
210		x	0	0	0	0
266		x	1	0	0	0
277		x	2	0	0	0
232		x	0	—	—	—
233			1	—	—	—
224		x	0	0	0	0
279	x	x	0	0	0	0
COLLAR/SHAM-DRUG TREATMENT						
244	x	x	0	0	0	0
242		x	0	0	0	0
256		x	0	—	—	—
300		x	0	—	—	—
295		x	0	—	—	—
205	x		0	0	0	0

a includes larvae and/or emergence pores

Appendix Table 5. continued

Rabbit number	Initial Examination			Final Examination		
	Fleas	Ticks	Cuterebra ^a	Fleas	Ticks	Chiggers
212			0	—	—	—
283		x	2	—	—	—
218		x	0	0	0	0
241			1	—	—	—
215		x	1	0	0	0
271		x	0	0	0	0
SHAM-COLLAR/DRUG TREATMENT						
206		x	0	1	0	0
222			0	—	—	—
292		x	1	0	1	0
239			0	—	—	—
204		x	0	1	0	0
253	x	x	0	0	0	0
282	x		3	0	3	0
270			2	0	1	0
228			1	0	0	0
252	x		0	0	0	0
219			1	0	0	0
293			0	0	0	0
SHAM-COLLAR/SHAM-DRUG TREATMENT						
238	x		0	1	0	0

a includes larvae and/or emergence pores

Appendix Table 5. continued

Rabbit number	Initial Examination			Final Examination		
	Fleas	Ticks	Cuterebra ^a	Fleas	Ticks	Chiggers
258		x	0	1	0	0
289	x	x	0	—	—	—
268		x	1	0	0	0
208	x		0	—	—	—
278			0	0	0	0
229	x	x	0	2	0	0
276	x		0	—	—	—
202			0	1	0	0
248			1	0	1	0
WOODLOT SAMPLE						
C-1	—	—	—	2	6	0
C-2	—	—	—	1	0	0
C-3	—	—	—	1	21	0
C-4	—	—	—	0	4	0
C-5	—	—	—	1	10	0
RADFORD SAMPLE						
R-1	—	—	—	0	0	100+
R-2	—	—	—	1	3	100+
R-3	—	—	—	1	1	100+
R-4	—	—	—	5	103	100+
R-6	—	—	—	1	43	100+

^a includes larvae and/or emergence pores

Appendix Table 6. Helminths recovered from 41 cottontail rabbits

Rabbit number	Cestodes		Nematodes		Caecum	Stomach
	Small Int.	Body Cavity	Small Int.	Large Int.		
COLLAR/DRUG TREATMENT						
264	1	17	0	0	1	2
259	8	18	0	0	0	2
298	6	6	0	0	1	0
210	1	3	0	0	0	2
266	1	8	0	0	1	0
277	3	0	0	0	0	1
224	4	18	0	0	0	0
279	0	18	0	0	0	0
COLLAR/SHAM-DRUG TREATMENT						
244	1	19	0	0	0	4
242	2	17	0	0	0	6
205	2	1	0	0	0	9
215	7	8	4	1	3	4
271	3	13	1	2	2	2
SHAM-COLLAR/DRUG TREATMENT						
206	5	13	0	0	2	1
292	2	21	0	0	0	1
204	2	3	0	0	2	12
253	1	39	0	0	0	4

Appendix Table 6. continued

Rabbit number	Cestodes		Nematodes		Caecum	Stomach
	Small Int.	Body Cavity	Small Int.	Large Int.		
282	2	31	0	3	0	0
270	7	11	0	3	0	0
228	13	40	0	0	1	0
252	17	0	0	0	1	0
219	2	0	0	0	0	0
293	3	45	0	0	7	0
SHAM-COLLAR/SHAM-DRUG TREATMENT						
238	3	17	0	0	1	9
258	1	9	0	0	0	1
268	7	11	0	0	0	1
278	1	10	0	0	1	7
229	0	25	0	0	0	8
202	2	8	0	1	3	10
249	3	1	0	0	1	0
248	16	0	0	0	0	0
WOODLOT SAMPLE						
C-1	2	6	0	0	0	8
C-2	1	0	0	0	0	0
C-3	1	8	0	0	0	1
C-4	0	26	0	0	2	3
C-5	2	21	0	0	6	2

Appendix Table 6. continued

Rabbit number	Cestodes		Nematodes		Caecum	Stomach
	Small Int.	Body Cavity	Small Int.	Large Int.		
RADFORD SAMPLE						
R-1	1	17	0	2	5	2
R-2	3	9	8	2	19	3
R-3	3	0	0	4	1	0
R-4	1	4	0	0	6	1
R-5	1	13	0	1	14	3

Seasonal Variation of Ectoparasite Loads in a Woodlot
Population of Cottontail Rabbits

Between January 28, 1973, and April 5, 1973, a follow-up experiment to the major investigation described in this manuscript was conducted. Its purpose was to determine if a select number of rabbits treated with insecticide-generating collars could reduce ectoparasite loads on nontreated rabbits within the population.

The design of this experiment called for treatment of two groups of rabbits. These two groups were part of the larger woodlot population of rabbits which had been sampled during the major investigation. The two groups were located at opposite ends of the woodlot. They were separated by more than a quarter mile distance. It was felt this distance would be adequate to insure the two groups remained separated. Half of one group received sham collar treatment and half of the other group received insecticide-generating collars. All rabbits were tagged with metal ear tags. These rabbits were to have been recaptured after a thirty-day period and ectoparasite loads of both groups quantified.

Although 21 rabbits were originally captured, treated, and released, only 4 of these were recovered. These 4 rabbits, with 11 others, were collected by shooting after retrapping efforts proved unsuccessful. No rabbits which had received

insecticide-generating collars were recovered. Because of the small number of rabbits recovered, and because it was doubtful that shooting sampled the original population that was tagged, the experiment was terminated.

Although this experiment was unsuccessful in terms of its original purpose, ectoparasite data collected provide insight into the seasonal nature of ectoparasite infection of cottontail rabbits. Ectoparasite numbers were quantified from 12 of the 15 rabbits collected. Since these rabbits were all collected from the enclosure woodlot, a comparison can be made with the ectoparasite data collected from the earlier woodlot sample of five rabbits. This comparison is made in appendix table 7.

As can be seen by appendix table 7, a dramatic increase in ectoparasite numbers occurred between winter and spring. Reports of seasonal changes in ectoparasite numbers for the cottontail rabbit have been made by Llewellyn and Handley (1945), McGinnes (1958), Stannard and Pietsch (1958), Mohr and Lord (1960), Forcum et. al. (1969), and McDonough ().

The findings presented here support those of previous investigators. In addition, they should be of value in describing seasonal changes in ectoparasite numbers of the cottontail rabbit within this region of Virginia.

Appendix Table 7. Winter and Spring ectoparasite loads of a woodlot cottontail rabbit population

Number of rabbits	Mean Number of Ectoparasites			Collection Dates
	Ticks	Fleas	Total	
5	8.2	1.6	9.2	December 3 - 22, 1972
12	80.6	13.6	94.2	March 20 - April 5, 1973

Cuterebra Host Records

Early in the course of this investigation, nine *Cuterebra* larvae were obtained from cottontail rabbits just after emergence from their host. Two of these were obtained from rabbits used in this study, and seven were obtained from rabbits which were being used in other research projects.

All larvae were collected during the months of August and September, 1972. Larvae were placed in a cup of moist soil, covered with woodchips, as described by Catts (1964), and allowed to pupate. They were then left undisturbed at room temperature.

On March 21, 1973, an adult fly emerged. The wings of this fly failed to unfold properly. It was felt this may have been due to a low humidity within the container holding the pupae. Because of this the soil containing the remaining pupae was remoistened. Two additional flies emerged with normal wings on March 27, 1973, and April 4, 1973, respectively. The three flies survived for periods of 6, 8, and 10, days, respectively.

The three flies, along with their puparia, were sent to Dr. E. P. Catts, Department of Entomology and Applied Ecology, University of Delaware, for identification. Dr. Catts identified these as being *Cuterebra buccata* (Fabricius) 1776. All were females.

According to Andrews (1969:20) the cottontail rabbit has been reported as a host species for four species of *Cuterebra*. These are *C. buccata*, *C. cuniculi*, *C. fontinella*, and *C. horripilum*.

Although *Cuterebra* parasitism has been reported for Virginia cottontails by a number of authors (McGinnes, 1958; Fortenberry, 1959; Krug, 1960; Wornecki, 1961; and Bellig, 1962), only McGinnes (1958) and Krug (1960) report on identifying these to species. Both of these authors also reported *Cuterebra buccata* as the species they found infecting Virginia cottontails.

Notes on the Intestinal Fluke (*Hasstilesia tricolor*)

Andrews (1969) reviewed the available literature on the rabbit intestinal fluke *Hasstilesia tricolor*. He reported that although it was probable this parasite could cause extensive host damage there was little evidence for this.

This parasite may have been a factor in the death of at least one rabbit observed during this investigation. This rabbit died six days after its original capture and introduction into an outdoor enclosure. During that period it dropped from its initial weight of 923 gms. to 625 gms. Necropsy revealed a severe enteritis of the small intestine along with heavy fluke infestation. Histological sections of the small intestine revealed extensive destruction of the villi associated with this parasite presence (figure 1).

The crowding of these flukes in the intestinal villi produced an unusual phenomena. Flukes were observed attached to one another by means of the oral and ventral suckers as seen in Figure 1.



Figure 1. Photomicrograph showing villi destruction by *Hasstilesia tricolor* and the "crowding" phenomenon.

A Rabbit Restraining Device

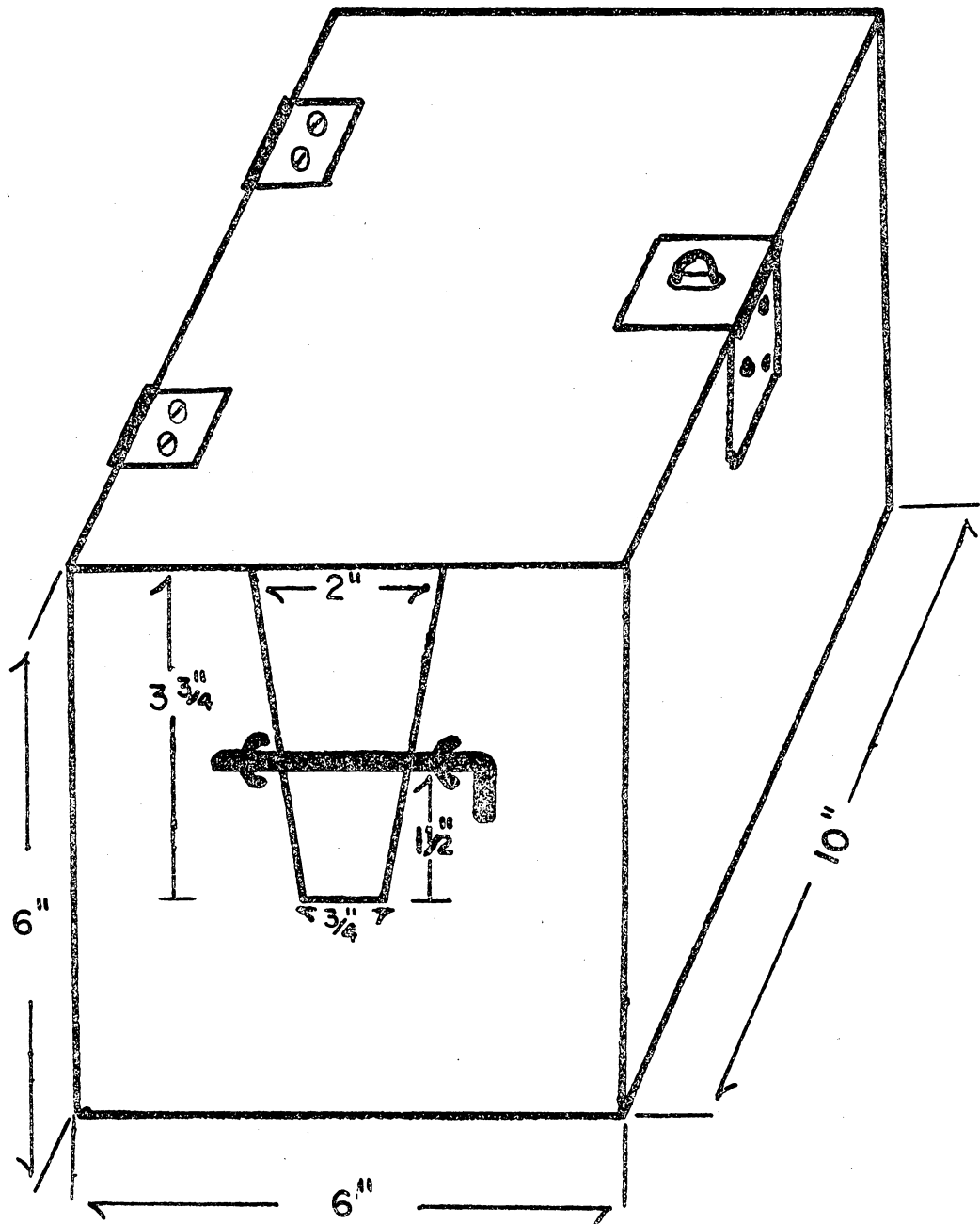
During the course of this investigation, it was necessary to collect cottontail rabbit blood samples from a marginal ear vein puncture and to administer drugs to rabbits through the use of a stomach tube. For one person to accomplish this task, it became necessary to design a restraining device which would secure rabbits and allow the investigator freedom of both hands.

A simple wooden box with a V-shaped opening and a hinged top was constructed to meet this purpose (Figure 2). Its inside dimensions were 10 x 6 x 6 inches. The V-shaped opening was cut in the front panel. The bottom of the V was flat and was $\frac{3}{4}$ of an inch across. The V was 3 and $\frac{3}{4}$ inches deep and was 2 inches across at its top.

The rabbit was placed in the box with its neck inside the V and its head outside the box. A sliding bar, made of heavy gauge wire and placed 1.5 inches from the bottom of the V, secured the rabbit's head in place. A latch on the side of the box was used to secure the top.

With the rabbit restraining in this manner, it was a simple task to collect blood samples from a marginal ear vein puncture, or to administer drugs through the use of a stomach tube.

Figure 2. A rabbit restraining device



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PATHOLOGICAL AND PHYSIOLOGICAL RELATIONSHIPS OF
PARASITIC DISEASE IN A SELECT COTTONTAIL
RABBIT POPULATION

by

Harry A. Jacobson

(ABSTRACT)

Forty-seven cottontail rabbits, *Sylvilagus floridanus*, were used in a 2 x 2 factorial experiment to determine the effects of parasite reduction on host physiology. Rabbits were treated with insecticide-generating collars (Sergeant's Sentry Dog Collar) and a broad spectrum antihelminth drug (Tramisol, American Cyanamid) to reduce parasite burdens. Rabbits were confined in 4 one-quarter acre outdoor enclosures during the treatment period. Thirty-one rabbits were recovered at the end of the treatment period. Data from these rabbits and 10 wild rabbits were analyzed. Treatment with insecticide-generating collars resulted in complete elimination of ectoparasites from rabbits which received this treatment. The two groups of rabbits which received treatment with the drug Tramisol had a significant reduction ($p < .05$) in the number of nematode parasites recovered. Reduction of nematode numbers was accompanied by significant changes in the generalized blood picture. Total serum protein, serum globulin fraction, lymphocyte percentage, and monocyte percentage were significantly ($p < .05$)

reduced and neutrophil percentage was significantly increased ($p < .05$) in drug treated groups. A simple correlation analysis was conducted between parasite burdens and select physiological measurements. A highly significant ($p < .001$) correlation of 0.57 was found between parasite burden and basophil percentage. A highly significant ($p < .01$) correlation of -0.57 was found between eye lens weights and the presence of *Cuterebra* maggots. A significant ($p < .05$) correlation of 0.33 was found between serum corticoid levels and numbers of ticks recovered. These results indicate that parasites may, under some conditions, operate as a limiting factor to cottontail rabbit populations.