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Source: Journal of Wildlife Diseases, 17(1) : 79-87

Published By: Wildlife Disease Association

URL: https://doi.org/10.7589/0090-3558-17.1.79

PREVALENCE OF *Cuterebra emasculator* IN SQUIRRELS IN MISSISSIPPI[1]

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Abstract: Between 1977 and 1979, 1,997 gray squirrels *(Sciurus carolinensis)* and 290 fox squirrels *(S. niger)* were examined for *Cuterebra emasculator* myiasis. Approximately 19% of the gray and 5% of the fox squirrels were infested with 1.9 and 2.5 larvae per host, respectively. Myiasis was seen between 14 August and 29 October. Peak infestations occurred in the second week of September. Adult and subadult squirrels had higher infestations than juveniles. Multiple infestations occurred in 51% of the hosts. Larval development sites were most prevalent in the axillary and back regions. The parasite was most prevalent in bottomland or flatland topography and hardwood habitat in east-central Mississippi. Fecundity of four virgin female flies averaged 771 eggs.

INTRODUCTION

Bot fly myiasis by *Cuterebra emasculator* has seriously affected squirrel *(Sciurus carolinensis, S. niger)* hunting in Mississippi. Infested squirrels are not eaten and myiasis results in substantial reduction of squirrel hunting.[20] Since squirrel hunting is the most popular form of hunting recreation in Mississippi, more information on this parasite's life history is important to squirrel management.[20]

This paper reports the prevalence of *Cuterebra* myiasis in squirrels in Mississippi. The seasonal occurrence of myiasis, habitats associated with *Cuterebra* infested squirrels and observations on parasite biology are provided.

MATERIALS AND METHODS

Dead squirrels were examined at hunter check stations in October, 1977-79. Squirrels were collected monthly on several state wildlife management areas (WMA) by shooting from July-December, 1977. During September, 1977, attempts were made to collect 10 squirrels in each of the 82 counties in Mississippi.

Collection date, species, sex, age, body weight and number of *Cuterebra* larvae or emergence scars were recorded. Squirrels were aged by the pelage technique as juvenile (< 6 months), subadult (6 to 16 months) or adult (> 16 months).[26] When possible, habitat description and topography at the collection site, adrenal weights, and locations of *Cuterebra* larvae or scars also were recorded.

Mature bot fly larvae that exited voluntarily or were excised from the host were placed in moist sand to pupate. Most puparia were stored at room temperature *(circa* 20 C) and humidity. Thirteen of 137 puparia collected during the survey were placed in an incubator at 6 C for 144 days and 39 puparia were stored at room temperature over 70% saturated NaCl to create a high humidity atmosphere.

In 1978, 22 of 40 unemerged pupae were injected with ecdysterone.[4] Ecdysterone solution was injected into the head cap-

[1] Supported by Federal Aid in Wildlife Restoration funds, Project W-48-26, Study VIII, Job VIII-4.

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sule at a dose of 0.35 μgms per gram of pupal weight.

Adult flies were stored at 6 C within four days after their emergence. Egg counts were obtained from four dead flies.

On 7 October 1978, 34 immature larvae obtained from hunter-killed squirrels were implanted subcutaneously in the dorsal region of 12 squirrels held as a lab colony at Mississippi State University. The animals were anesthetized with methoxyflurane[22] [4] and larvae were inserted as described by Bennett.[7] Ten squirrels received three larvae each and two squirrels received two larvae each.

Least squares analysis of variance was conducted on the SAS computer program.[6] The prevalence of bot fly parasitism was subjected to Chi-square tests.[15]

RESULTS

Myiasis of gray and fox squirrels. A total of 1,997 gray and 290 fox squirrels was collected. No significant ($p \leq 0.05$) differences were seen between infestation prevalence or number of larvae per infested host by host sex (Table 1). Significant ($p < 0.01$) differences were seen by host species. *Cuterebra* myiasis was found on 19% of gray squirrels with a mean of 1.95 larvae; whereas, 4.8% of the fox squirrels were infested with a mean of 2.5 larvae. Subadult gray squirrels had significantly ($p < 0.05$) higher infestation prevalence (24%) than adults (19%) and adults had significantly ($p < 0.01$) higher infestation prevalence than juveniles (3.8%). A single larva occurred on 49% of the infested hosts with the majority of hosts infested with 2 to 9 larvae.

Seasonal prevalence of *Cuterebra* myiasis between July-December, 1977 indicated that the highest monthly prevalence of parasites was in the second week of September (Figure 1). The earliest and latest observations of *Cuterebra* myiasis of squirrels were 14 August and 29 October 1977.

TABLE 1. Comparison of host species, age class and sex and *Cuterebra* infestation of squirrels.

	Gray[2]		Fox[2]	
Age class[1]	Male	Female	Male	Female
Adult				
Number	579	470	102	112
Percent infested	19.5	18.5	4.9	3.6
Mean no. larvae per infested	2.1	2.0	2.0	2.5
Subadult				
Number	379	313	33	34
Percent infested	23.2	25.9	12.1	2.9
Mean no. larvae per infested	1.9	1.9	3.3	3.0
Juvenile				
Number	144	112	9	0
Percent infested	4.2	3.5	0	—
Mean no. larvae per infested	1.3	1.0	—	—

[1]A significant ($p < 0.001$) difference exists between infestation prevalence and age class of gray squirrels ($x^2 = 51.2$ with 2df). All gray squirrel age classes are $p < 0.05$ ($x^2 \geq 4.9$ with 1df) different from each other in infestation prevalence.
[2]Species are significantly ($p < 0.01$) different in infestation prevalence ($x^2 = 35.6$ with 1df).

[4] Pittman-Moore, Inc., Washington's Crossing, New Jersey 08608, USA.

A high prevalence (≥ 60%) of the parasite in September, 1977 was confined to east-central Mississippi (Figure 2). Parasite prevalence in gray squirrels was significantly ($p < 0.05$) associated with hardwood forests, rather than mixed pine-hardwood or pine forests (Table 2). Similarly, squirrels collected in bottomland or flatland areas had significantly ($p < 0.05$) higher parasite prevalence than squirrels from upland and hilly areas.

The most prevalent larval development sites on the host's body were back (28%), axillary areas (27%), neck (14%) and sides (13%). The belly, forelegs and head accounted for 10%, 7% and 1% of the larval development sites. Myiasis had no significant ($p < 0.05$) effect on host adrenal weight, although there was a significant ($p < 0.05$) effect on body weights of gray squirrels (Table 3). There were significant interactions between age and infestation of both gray and fox squirrels. Adult and juvenile squirrels had heavier body weights in infested individuals than uninfested; whereas, subadult squirrels had heavier body weights for uninfested individuals than for infested.

Laboratory experiments. There was no difference between the emergence success of pupae maintained in high humidity and those which were maintained at room humidity. Eleven percent emerged from both groups. The median emergence date for nine flies reared at room temperature was 5 July (24 June-3 August). Pupal period was about 270 days (250-288). None of the 13 refrigerated pupae emerged. Four female flies emerged 7, 58, 58, and 59 days after receiving ecdysterone injection. Of a total of 13 flies, eight were males and five were females. Egg counts of four virgin flies were 247, 551, 830, and 1,437, with a mean of 771. Adult flies, puparia and larvae were placed in the U.S. National Museum, Washington, D.C. Flies were identified as *Cuterebra emasculator* Fitch by Dr. Curtis Sabrosky, U.S. National Museum.

All 10 squirrels that had three larvae implanted died within two days post-operation. Larvae dropped from the remaining two squirrels did not form

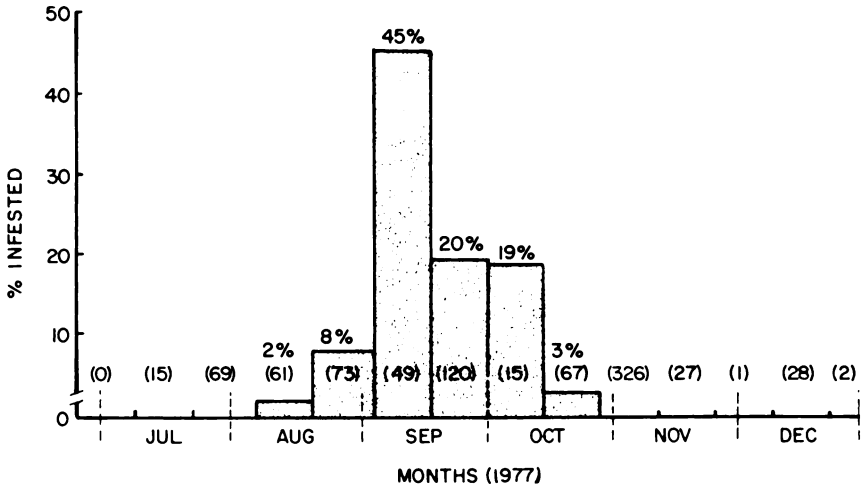

FIGURE 1. Biweekly prevalence of infestation in Mississippi squirrels by *Cuterebra emasculator*, July-December, 1977 (numbers in parenthesis represent sample size).

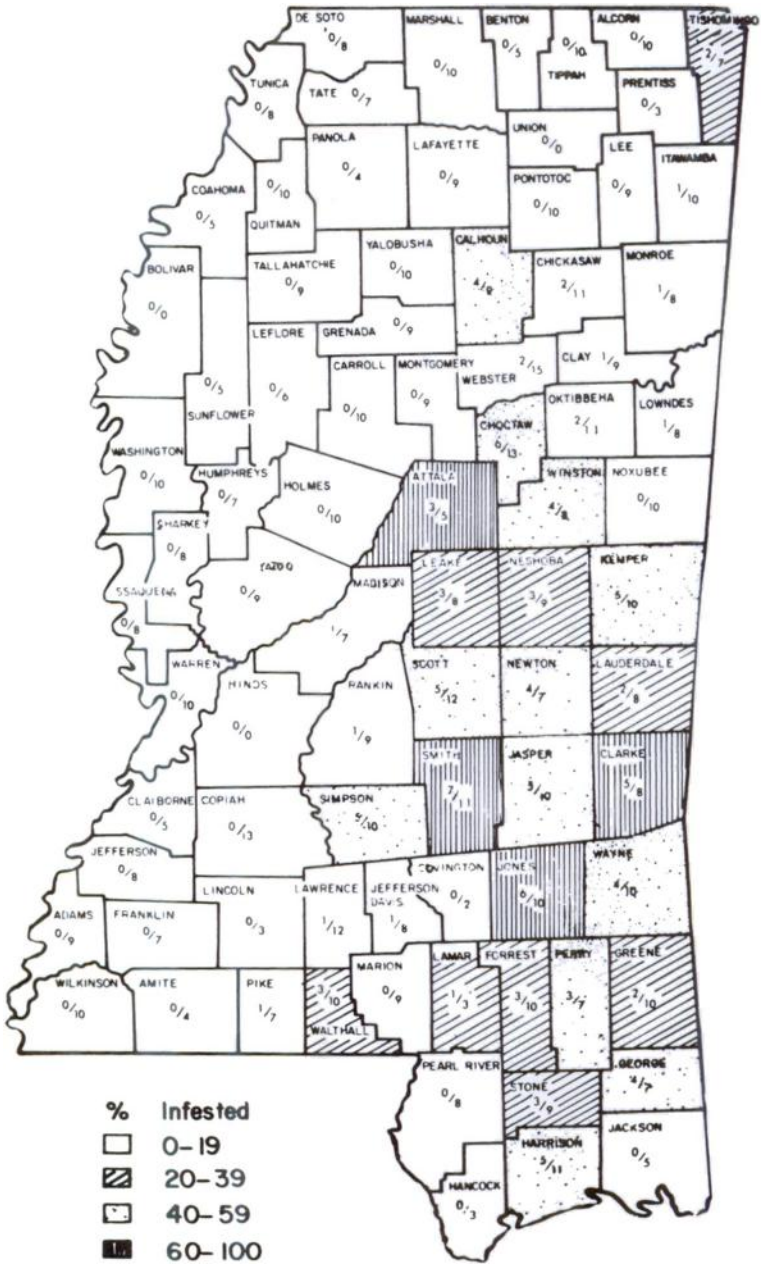

FIGURE 2. Prevalence of botfly infested squirrels in Mississippi, September, 1977 (number infested/number examined is indicated for each county).

TABLE 2. *Cuterebra* myiasis of gray squirrels in relationship to collection site habitat and topography.

Forest or terrain type	Number of squirrels examined	Percent[3] infested	Mean No. larvae per infested host
Forest Type[1]			
Hardwood	349	13.1[b]	2.1
Pine-hardwood	462	9.5[a]	2.0
Pine	14	7.1[a,b]	8.0
Topography[2]			
Bottomland	417	13.9[a]	2.0
Flatland	107	12.1[a]	2.5
Upland	94	7.4[b]	3.0
Hilly	218	6.8[b]	2.3

[1]Forest types were significantly different ($p<0.01$) in infestation prevalence ($x^2 = 15.5$ with 2df).
[2]Topography types were significantly ($p<0.05$) different in infestation prevalence ($x^2 = 8.6$ with 3df).
[3]Means without the same superscripts within forest or terrain type are significantly different at the $p<0.05$ level.

viable puparia. Gross examination of the dead squirrels showed varying degrees of larval migration and host induced trauma. One larva had migrated subcutaneously to the host's belly.

DISCUSSION

The lack of significant associations between host sex and infestation prevalence is similar to the findings of Dorney[18] and McKinney and Christian[21] of *C. emasculator* in chipmunks *(Tamias striatus)*. That the juvenile age class had a significantly smaller percentage of infested individuals than either the subadult or adult age groups is contrary to results of Dorney[18] and Bennett[7] who reported that *C. emasculator* infestation in chipmunks was greatest in the juveniles of both sexes and lowest in adult females. These data support Allison's[1] conjecture that oviposition site is not related to the squirrel's maternal area.

The significant interactions seen between adult, subadult and juvenile body weights in *Cuterebra* infestation are difficult to explain. McKinney and Christian[21] reported larger body size in infested chipmunks of all ages. Bennett[7] postulated that host activity accounted for increased infestation of juvenile and adult male chipmunks and McKinney and Christian[21] proposed that body size is related to activity and thus infestation. Perhaps host activity may account for larger body sizes of infested adult and juvenile squirrels, but smaller body size in infested subadult squirrels.

The finding that multiple infestation by *C. emasculator* in squirrels was more common than singular infestation contrasts with reports for this parasite in chipmunks.[7,21] Behavioral differences associated with host species, sex and age might be expected to influence the number of larvae acquired.

The most prevalent larval sites were the axillary region and the back, generally in the shoulder region, which concurs with other reports for gray squirrels.[1,2] No larvae appeared in the genital or groin area, which is the typical development site of *C. emasculator* in chipmunks.[7]

Parasite presence in squirrels between 14 August and 29 October with a peak in the second week of September is similar to other reports of *Cuterebra* parasitism

of squirrels.[1,2] This closely approximates seasonal prevalence of *C. emasculator* parasitism of chipmunks.[7,8,11]

Habitat associations and parasite prevalence differ from the report of Atkeson and Givens[2] who reported heaviest infestation in hilly localities with well drained sandy soils. We found heaviest infestations in bottomlands and flatlands. However, in the present study, and in an earlier investigation,[20] the parasites' lowest prevalence was in poorly drained bottomland hardwood forests of the Mississippi Delta. Whereas, eastern Mississippi counties have pine-hardwood or pine forests on better drained soils. Bennett[3] reported the highest infestation of *C. emasculator* in chipmunks occurred in secondary growth mixed forests and noted the lowest infestation in mature coniferous forests.

Success of adult emergence from puparia was similar to that reported by Allison[1] and was as expected for pupae reared from animals killed before most larvae could obtain full maturity. Ecdysterone injections appeared to increase successful emergence and decreased pupal periods.

Emergence of adult flies from undisturbed pupae was earlier than expected based on seasonal prevalence of host infestation. However, lab reared

TABLE 3. Comparison of mean body weights and adrenal weights of gray and fox squirrels by sex, age and *Cuterebra* infestation.

	Fox		Gray	
Age	Infested[1]	Uninfested	Infested[1]	Uninfested
	Male			
Adult				
Number weighed	1	48	40	132
Body weight (g)	800	680	433	430
Adrenal weight (g)	0.32	0.28	0.18	0.17
Subadult				
Number weighed	2	19	15	64
Body weight (g)	492	556	395	409
Adrenal weight (g)	0.19	0.24	0.12	0.15
*Juvenile**				
Number weighed			1	25
Body weight (g)			376	218
Adrenal weight (g)			0.08	0.07
	Female			
Adult				
Number weighed	3	55	23	123
Body weight (g)	777	665	433	434
Adrenal weight (g)	0.30	0.30	0.18	0.17
Subadult				
Number weighed	1	17	11	48
Body weight (g)	369	596	406	409
Adrenal weight (g)	0.13	0.25	0.16	0.15
*Juvenile**				
Number weighed			3	16
Body weight (g)			286	242
Adrenal weight (g)			0.07	0.07

[1]Infested gray squirrels were significantly different in weight from uninfested squirrels at $p \leq 0.05$ ($F = 4.1$) and there was a significant ($p < 0.05$) interaction between age and infestation for both gray and fox squirrels ($F = 3.7$ and 5.5, respectively).
*No juvenile fox squirrels were infested.

flies have been shown to emerge earlier than those under natural conditions.[9]

Egg counts obtained from *C. emasculator* were low compared to other *Cuterebra* species[3,4,11,19] which generally range from 1000-2000 eggs. This may suggest more recent adaptation by *C. emasculator* to *Sciurus* hosts. Lowered fecundity has been reported for *C. buccata* reared from laboratory rabbits when compared to flies reared from the normal cottontail rabbit host for *C. buccata*.[19]

In addition to low fecundity of *C. emasculator* reared from *Sciurus* hosts, other lines of evidence suggest recent evolution of the parasite and adaptation to *Sciurus* hosts. Seasonal occurrence and adult and larval morphology are not known to differ from the *C. emasculator* in chipmunks. However, host larval site location, prevalence of multiple infestation, habitat associations and host infestation prevalence are different between chipmunk and squirrel hosts. The use of larval transplants has been a successful means of obtaining mature larvae. Chipmunk hosts are suitable for such transplants[7,9] and successful use of the transplant technique has been similarly used with cottontail rabbits and *C. buccata* larvae (Jacobson unpubl.). Death of most squirrel transplant hosts and aberrant migratory behavior of transplanted larvae also may indicate more recent adaptation by *C. emasculator* to squirrel hosts. Low larval prevalence in fox squirrels may indicate this species is the least well adapted host for *C. emasculator*.

Cuterebra parasitism of *Tamias striatus* has been reported in Ontario,[7] Wisconsin,[18] New Hampshire,[21] New York,[23] Michigan,[10] Minnesota (Pers. comm. R.E. Lee, Itasca Biological Exp. Stat., Minnesota), and Pennsylvania.[21] *Cuterebra* parasitism of *Sciurus* hosts has been reported in southeastern but not northern Ohio,[13] Virginia,[16,24] West Virginia,[27] North Carolina,[1] Alabama,[2] Georgia,[17] Mississippi,[14,20] eastern Tennessee (Pers. comm. Mr. Joe Bruna, Kentucky Game & Fish Comm.), and South Carolina (Pers. comm. Mr. W.B. Conrad, South Carolina Wildlife and Marine Resources Department). Incidental infestation of red squirrels *(Tamiasciurus hudsonicus)* by *Cuterebra* spp. has been reported.[18] However, reports of *Sciurus* spp. infestation by *Cuterebra* are noticeably lacking in areas for which *Cuterebra* parasitism is prevalent in *T. striatus* and vice versa.

On the basis of the above, we hypothesize that distinct races of *C. emasculator* may exist. The fly infesting squirrels may be spatially and behaviorally isolated from the fly infesting chipmunks.

It is apparent that much remains unknown about the autoecology of *C. emasculator* and associated host parasite relationships. Of particular importance would be the elucidation of adult fly mating and oviposition behavior, and comparative studies of laboratory infestations of *Sciurus* and *Tamias* hosts.

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Received for publication 30 May 1980