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TULAREMIA IN THE WILDLIFE OF ARKANSAS

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INTRODUCTION

Tularemia is generally considered to be a disease of rabbits and hares from which, through handling, humans contract the infection. This consideration has been fostered by reports, such as by Francis (11), and by particular statistics, such as for Illinois (17), which implicate rabbits in well over 90% of human infections. Despite this general conception of its being a disease of rabbits and hares, tularemia is also known to affect many other animals and birds, at least 40 different species have been reported to be naturally infected (see 3, 12, 13). Also, despite the high incidence of human cases from contacts with infected rabbits in Illinois and elsewhere, in some areas, like Arkansas, human disease is more often contracted by bites of vectors like ticks (16), 63% being reported for the Ozark region of western Arkansas and Missouri (1,9). The contrasting sources of human infections in Illinois and Arkansas are of particular interest because of the high incidence of tularemia in these two states. In the period from 1924-49 Illinois led all states with 3051 human cases while Arkansas was second with 1613 cases (17). When the total population of the two states is compared, 7,897,241 for Illinois and 1,949,387 for Arkansas, according to the 1940 census (15), Arkansas emerges with the highest per capita human case rate of all the States. Tularemia in Arkansas has been the subject of several reports (1, 5, 6, 7, 9, 13) but none of these have involved a comprehensive survey of the disease in the wildlife of the state, although Calhoun **et al.** in 1956 reported findings for several animal species. An opportunity to make such a survey became possible along with a continuing taxonomic, distributional study of mammalian species in Arkansas being conducted by members of the Department of Zoology of the University of Arkansas (14). This report records the testing of tissues from more than 1500 animals with the isolation of **Pasteurella tularensis** from eight of them and the finding of a high antibody titer in the serum of a ninth. Tests on 24 birds and 28 snakes were negative.

MATERIALS AND METHODS

The methods used for the collection and preparation of the tissue samples were the same as reported previously (8).

The tissue samples were plated upon a meat infusion agar, after Francis (10), containing 5% blood, 1% glucose, 0.1% cystine (or

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cysteine hydrochloride) adjusted to pH 7.3, and autoclaved. In their report of this medium Brigham and Rettger (2) recommended drying of the surface of the agar before using, a procedure we followed. All plates were incubated at 37°C and were examined 2, 4, and 10 days after inoculation. Suspicious colonies were subcultured and if showing typical morphology upon staining by Gram's method, the organisms were checked by slide agglutination with specific antisera.

Standard tube agglutination tests were conducted on sera from animals using a commercially prepared antigen of **P. tularensis,** (Lederle).

RESULTS AND DISCUSSION

The results obtained on animal tissues are shown in Table I. It can be seen that in two instances, tularemia organisms were isolated from **Didelphis marsupialis,** the opossum; once each from **Glaucomys volans,** the flying squirrel; **Sciurus niger,** the fox squirrel; **Mus musculus,** the house mouse; and **Sigmodon hispidus,** the cotton rat; and twice from **Odocoileus virginianus,** the white-tailed deer. Of these six species, isolates from **G. volans,** and **S. niger,** the squirrels, are a new findings and not previously reported, although both of these species belong to the general groups of animals which are commonly infected. The finding of tularemia in deer is not surprising either, having been reported before (4), but also because they are subject to heavy tick infestation. In this regard, tick infection with **P. tularensis** may be relatively high in Arkansas judging from the comparatively high incidence of human infections resulting from tick bite (1, 9). In Table II it is seen that no **P. tularensis** organisms were isolated from 24 specimens of 11 different kinds of birds nor from 18 specimens of 6 kinds of snakes. In Table III it is seen that of the agglutination tests conducted on sera from 54 animals and 10 snakes, only one serum, that of **D. marsupialis,** showed a significant titer, 1:1280. Such a high titer suggests the oppossum had had a recent exposure to the organisms, such as might have been acquired from eating a dead infected rabbit. The low titer of 1:40 for **F. domestica,** a house cat, is probably not of much significance. The percentage of positive sera is somewhat lower than the 17.2% found by McKeever **et al.** (13) for 2004 mammals from Florida and Georgia they tested. This difference could be due to the relatively small sample herein tested. Of those McKeever **et al.** reported, the opossum was among the animals species most frequently found to have agglutinins. Others included the raccoon, the striped skunk, and the gray fox. Since only a few specimens of these animals were examined in the present study, there were apparently not enough to uncover a positive. However, Calhoun **et al.** (7) reported finding agglutinins in sera of racoons and opossums in their Arkansas material. Since these various animals are common in the state, they may also be important in the total picture of tularemia in Arkansas. It is interesting that tularemia was not detected in any of the more than 200 rabbits examined, but then, McKeever **et al.** (13) also found that only one of 188 rabbits tested for agglutinins was positive.

Table I

Pasteurella tularensis Isolated by Direct Culture of Blood and Tissue Samples

MAMMALS

	No. positive/No. tested
Didelphis marsupialis the opossum	(2)1/64
Blarina brevicauda short-tailed shrew	0/6
Cryptotis parva little short-tailed shrew	0/1
Lasiurus borealis red bat	0/5
Geomys bursarius pocket gopher	0/17
Glaucomys volans flying squirrel	(3)/13
Sciurus carolinensis southern gray squirrel	0/10
Sciurus niger fox squirrel	(2)/51
Mus musculus house mouse	(4)/170
Rattus norvegicus rat	0/14
Microtus pinetorum pine vole	0/24
Oryzomys palustris Texas rice rat	0/6
Peromyscus gossypinus Rhoads cotton rat	0/43
Peromyscus leucopus white footed mouse	0/213
Peromyscus maniculatus Ozark white footed mouse	0/8
Peromyscus nuttali southern golden mouse	0/26
Peromyscus sp.	0/16
Reithrodontomys fulvescens golden harvest mouse	0/232
Sigmodon hispidus Texas cotton rat	1/227
Sylvilagus aquaticus swamp rabbit	0/44
Sylvilagus floridanus cottontail rabbit	0/192
Sylvilagus sp.	0/2
Urocyon cinereoargenteus gray fox	0/14
Vulpes fulva red fox	0/9
Canis familiaries dog	0/2
Procyon lotor raccoon	0/25
Mephitis mephitis skunk	0/18
Felis domestica house cat	0/8
Odocoileus virginianus white-tailed deer	2/70

( ) indicate pools and number of individuals comprising each.

Table II
Pasteurella tularensis Isolated by Direct Culture of Blood and Tissue Samples
BIRDS AND REPTILES

	No. positive/No. tested
BIRDS:	
Buteo jamaicensis red-tailed hawk	0/1
Cathartes aura turkey vulture	0/5
Coccyzus americanus yellow-billed cuckoo	0/3
Cyanocitta cristata blue-jay	0/1
Dendroica pinus pine warbler	0/1
Falco sparverius sparrow hawk	0/1
Helmintheros vermivorus worm-eating warbler	0/1
Lanius ludovicianus loggerhead shrike	0/8
Quiscalus quiscula common grackle	0/1
Vireo olivaceous red-eyed vireo	0/1
Zonotrichia albicollis white-throated sparrow	0/1
REPTILES:	
Agkistrodon mokasen copperhead	0/2
Agkistrodon piscivorus water moccasin	0/8
Columber constrictor racer	0/2
Coluber flagellum coachwhip	0/2
Lampropeltis getulus king snake	0/2
Natrix sipedon water snake	0/2

Table III
Mammal and Reptile Sera Tested for **Pasteurella tularensis** Agglutinins

	No. positive/ No. tested	Titer
MAMMALS:		
Didelphis marsupialis the opossum	1/20	1:1280
Felis domestica house cat	1/3	1:40
Mephitis mephitis skunk	0/5	
Odocoileus virginianus white-tailed deer	0/1	
Procyon lotor raccoon	0/4	
Sylvilagus floridanus cottontail rabbit	0/4	
Sylvilagus aquaticus swamp rabbit	0/4	
Sylvilagus sp.	0/2	
Urocyon cinereoargenteus gray fox	0/1	
REPTILES:		
Agkistrodon mokasen copperhead	0/2	
Agkistrodon piscivorus water moccasin	0/3	
Coluber constrictor racer	0/2	
Coluber flagellum coachwhip	0/1	
Natrix rhombifera diamond-backed water snake	0/1	
Natrix sipedon water snake	0/1	

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