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BLOOD PARASITES OF
-
ILLINOIS WILD TURKEYS
(TITLE)

BY

Ulrike G. Guenthner

## **THESIS**

SUBMITTED IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF

MASTER OF SCIENCE

IN THE GRADUATE SCHOOL, EASTERN ILLINOIS UNIVERSITY CHARLESTON, ILLINOIS

1976 YEAR -

I HEREBY RECOMMEND THIS THESIS BE ACCEPTED AS FULFILLING THIS PART OF THE GRADUATE DEGREE CITED ABOVE

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Master's Degree Certificate for Comprehensive Examination

I certify that Ulrike G. Guenthner successfully passed a comprehensive examination. has

The examining committee consisted of:

Signatures of the Committee

14 october 1976

Adviser

The undersigned, appointed by the Chairman of the Department of Zoology, have examined a thesis entitled

### BLOOD PARASITES OF ILLINOIS WILD TURKEYS

Presented by

Ulrike G. Guenthner

a candidate for the degree of Master of Science and hereby certify that in their opinion it is acceptable.

# BLOOD PARASITES OF ILLINOIS WILD TURKEYS Ulrike G. Guenthner

Abstract: Blood smears, from 152 wild turkeys (Meleagris gallopavo silvestris) killed during hunting seasons in southern Illinois, were examined for blood parasites. Haemoproteus meleagridis infected 34% of the 83 adult and 19% of the 69 immature turkeys. Of the infected birds, 54% had one or two infected cells per 5000 erythrocytes and 78% had one to five. Infection rates were significantly higher in counties along the Mississippi River than in Pope County, located about 40 miles east of the river. Other parasites specifically searched for but not found were Leucocytozoon, Plasmodium, Trypanosoma and microfilaria.

The eastern wild turkey (Meleagris gallopavo silvestris) once ranged over all the forested regions of Illinois but due to habitat destruction and overkilling it disappeared completely in the 1900's (Felger, 1909). Reintroductions with pen-raised turkeys in the 1950's were unsuccessful (Hendricks, 1969), but releases of wild-trapped birds from West Virginia and Mississippi (Casey, 1957) have established populations in four southern Illinois counties. Limited spring hunting has been carried out on these birds since 1970. Although internal and blood parasites have been shown to cause mortality in turkey populations of other states (Eve et al., 1972, Forrester et al., 1974, Prestwood et al., 1971), no data are available for Illinois. This is a report of a survey

of blood parasites in Illinois wild turkeys which should be preliminary
to studies of the pathogenic effects these parasites might have on their
host.

This study was supported in part by the Illinois Department of Conservation Federal Aid in Wildlife Restoration Project W-63-R. Dr. R. Andrews and Dr. B. Ridgeway supervised the study and assisted with field and laboratory work. Dr. N. Levine, College of Veterinary Medicine, University of Illinois confirmed identification of the parasites found, and Dr. L. Hunt and Dr. W. Keppler edited this thesis.

#### **METHODS**

Turkeys have been hunted during April of each year since 1970 in three southern Illinois counties: Alexander, Jackson and Union. They have been hunted in Pope county since 1972. All birds shot have been examined at mandatory check stations maintained by the Illinois Department of Conservation in each county. At these stations the turkeys were weighed, aged (Taber, 1971), sexed, eviscerated and the location of kill recorded.

Thin smears were made from blood in 1971 and 1974 through 1976.

Blood was taken from the branchial or jugular vein exposed when the crop was removed, or from the heart after its removal, or from blood in the body cavity. Blood samples were labeled, airdried, fixed for two minutes in absolute methyl alcohol and airdried again. They were then stained with Giemsa's stain according to Wright's method (Humason, 1962). The entire slide was first examined for <a href="Leucocytozoon">Leucocytozoon</a> and microfilaria at 150x magnification. The slides were then searched at 1500x for Haemoproteus, Plasmodium and Trypanosoma until 5000 erythrocytes were

recorded and photographs were made. Data were analysed with the Chisquare test of the independence of variables (Naiman et al., 1972).

#### RESULTS

Haemoproteus meleagridis was the only blood parasite identified by examination of 152 blood smears from wild turkeys (Fig. 1). Gamonts were seen in 27% of the smears (Table 1). Levels of infection remained about the same for three of the four years but the percentage of infected birds was significantly lower in 1975. There were differences in infection rate in the four counties. The three counties along the Mississippi River, Alexander, Jackson and Union, ranged from 21% to 39% but were not significantly different. Pope county, approximately 40 miles east, however, had significantly fewer infected birds (8%). Adult turkeys had a significantly higher rate of infection than did juveniles (Table 2).

The level of parasitemia in each bird was low; 54% of the turkeys had only one or two infected cells per 5000 erythrocytes, 78% had one to five parasites. The remaining 22% had up to 20 infected cells (Table 3). Immature turkeys had a slightly lower level but this difference was not significant. Multiple infection of erythrocytes was observed in three adult turkeys; one erythrocyte in each bird contained two gamonts.

Unidentified organisms were seen in 29% of the blood smears (Table 4).

These organisms were easily distinguishable, elongate, cigar- or crescentshaped bodies averaging about 30 in length. Some had a flagellum-like,
others a knob-like process at one end (Fig. 2 and 3). These rods were
extracellular and often occurred in great numbers. They were most numerous

Table 1. Wild turkeys from four Illinois counties infected with Haemoproteus meleagridis.

	Alexander County Jackson County			County	Union	County	Pope (	County	Total	
Year	No. Examined	% Positive	No. Examined	% Positive	No. Examined	% Positive	No. Examined	% Positive	No. Examined	% Positive
1971	17	35	12	42	15	13			44	30
1974	10	30	13	46	17	24	2	50	42	33
1975	4		9	<b></b>	13	15	3		29	7
1976	_7	29	<u>10</u>	60	12	<u>33</u>	_8		<u>37</u>	32
Total	38	29	44	39	57	21	13	8	152	27

Table 2. Relationship between <u>Haemoproteus meleagridis</u> and age of turkey shot in southern Illinois.

	Adult	Juvenile	Total
Number examined	83	69	152
Number positive	28	13	41
Percent positive	34	19	27

Table 3. Level of parasitemia in adult and juvenile wild turkeys from southern Illinois.

No. of parasitized	Ad	ult	Juve	nile_	Tot	Total	
cells per 5000 erythrocytes	No.	%	No.	%	No.	%	
1-2	14	50	8	62	22	54	
3-5	8	25	3	23	11	24	
6-20	6	25	_2	<u>15</u>	_8_	_22	
Total	28	100	13	100	41	100	

Table 4. Wild turkeys from four southern Illinois counties infected with an unidentified organism.

Year	Alexander County		Jackson County		Union County		Pope County		Total	
	No. Examined	% Positive	No. Examined	% Positive	No. Examined	% Positive	No. Examined	% Positive	No.	% Positive
1971	17	35	12	67	15	-			44	32
1974	10	20	13	15	17	12	2	<del></del>	42	14
1975	4	75	9	56	13	39	3		29	45
1976	_7	43	<u>10</u>	<u>30</u>	12	<u>25</u>	_8	<u>25</u>	37	30
Total	38	37	44	41	57	18	13	15	152	29

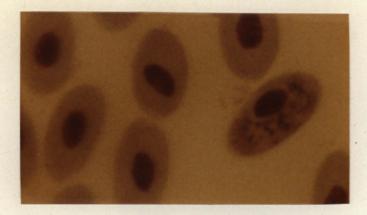


Fig. 1. Haemoproteus meleagridis gamont in turkey blood.

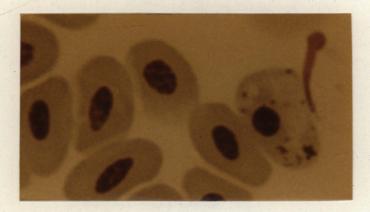


Fig. 2. Unidentified organism in turkey blood.

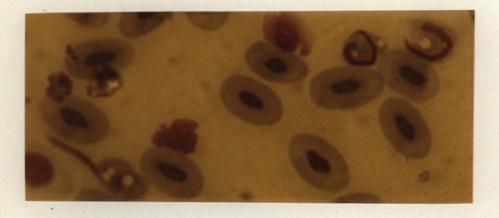


Fig. 3. Unidentified organism in turkey blood.

in 1975 and least numerous in 1974. Their distribution was like  $\underline{H}$ .

meleagridis; Pope county had the lowest rate of infection (15%) and

Jackson county had the highest (40%). There was no significant difference between infection rates in juvenile (27.5%) and adult (30.0%) turkeys.

#### DISCUSSION

meleagridis than most other states where the birds have been studied.

Only 27% of the 152 Illinois turkeys carried H. meleagridis while in

Georgia one of two birds was infected (Love et al., 1953). Eve et al.

(1972) reported 49% of 76 turkeys infected in West Virginia. Texas had an infection rate of 80% in 133 birds (Cook et al., 1966) and in Florida,

84% of 605 turkeys were infected (Forrester et al., 1974). Only Pennsylvania (5%) (Kozicky, 1948) and South Carolina (13%) (Noblet and Moore,

Lower rates of infection in Illinois might be due to several factors.

Geographic location is one apparent factor since accounts in the literature suggest that infection rates decline from south to north. The southern states have higher temperatures and greater precipitation.

Within Illinois, differences in climatic and topographic conditions influenced the infection rates. The three counties located along the Mississippi River, Alexander, Union, and Jackson, are in part subject to frequent flooding with swampy areas adjacent to much of the turkey habitat. The rates of infection were similar in birds of these three counties (Table 1), probably due to the fact that they are adjacent and allow an essentially continuous turkey population. Pope county, located

approximately 40 miles east of the other three counties, is considerably higher and drier; its turkeys had a significantly lower rate of infection. Similar findings were reported in South Carolina (Noblet and Moore, 1975) where <u>Haemoproteus meleagridis</u> was found in turkeys from coastal, but not from inland areas.

The described vectors for <u>Haemoproteus</u>, hippoboscid flies and biting midges require water for breeding (Borror and DeLong, 1971). If other organisms are capable of transmitting the parasite, they too are probably tied to an aquatic habitat. The lack of water and consequently the effect of precipitation on the rate of infection was evident in 1975. While levels of <u>H. meleagridis</u> infection remained nearly the same for three years, they decreased markedly in 1975 (Table 1). Precipitation in 1974 was about 20% below the five-year average for southern Illinois (Climatological data for Illinois published by U. S. Department of Commerce).

Another factor that might cause lower infection rates of <u>Haemoproteus</u> in Illinois is the density and distribution of the turkey population. No recent data are available but in 1965 Aldrich reported only 500 birds in Illinois. Alabama and Florida, however, had 80,000 and most other southern states had more than 30,000 wild turkeys (Aldrich, 1967). Based on the number of turkeys killed each year, Illinois density still is relatively low. A low-density, dispersed population reduces the chance of vector contact between birds. Hippoboscid flies are less likely to contact turkeys scattered over a large area because they tend to be carried by their host rather than fly (Lapage, 1962). Perpetuation of <u>H. meleagridis</u> occurs by means of infected adults as a source for juveniles each year.

Control of this parasite is in part a matter of reducing the numbers of the vector. But <u>Haemoproteus</u> has not been shown to be pathogenic (Levine, 1961) so there is no need for control measures.

Leucocytozoon was not found in Illinois turkeys. The parasite has been prevalent in other flocks infecting 40% of 268 turkeys in Virginia (Mosby and Handley, 1943), 43% of 76 turkeys in South Carolina (Noblet and Moore, 1975), and 100% of 5 turkeys in Pennsylvania (Kozicky, 1948). A possible explanation for the complete absence of this parasite in Illinois turkeys would be the lack or insufficient number of vectors. But according to personal communication with Dr. G. T. Riegel, Simulium blackflies are numerous in southern Illinois. Wild turkeys may also have some degree of immunity against Leucocytozoon as suggested by Noblet and Moore (1975).

Trypanosomes were not seen on any of the 152 blood smears. They are rare in birds and more common in mammals. The only report of this parasite in wild turkeys is by Eve et al. (1972) in which a single trypanosome was found in the blood of one bird in West Virginia.

Both <u>Plasmodium</u> and microfilaria which are common in many birds are reported only once each in the literature for turkeys (Telford and Forrester, 1975; Anderson and Prestwood, 1969). Neither one of these organisms was encountered in this study. <u>Plasmodium</u> might have been latent. Isodiagnosis could have been used to detect the parasite by subinoculating heparinized blood of wild turkeys into domestic turkey poults. A comparison between uninoculated control poults and recipient poults might have revealed Plasmodium.

Our sampling techniques unfortunately were limited since the turkeys of this study were already dead, some for several hours. Bone marrow or

tissue studies were not possible because the hunters kept the birds. If the conditions had been more suitable, probably other parasites besides <a href="Haemoproteus"><u>Haemoproteus</u></a> would have been found.

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#### LITERATURE REVIEW

Both protozoan and metazoan blood parasites have been reported from avian hosts. The majority of the protozoans belong to two classes, the Sporozoasida and the Zoomastigophora.

All species in the class Sporozoasida require two types of host:

a definitive invertebrate host where the parasite reaches sexual maturity,
and an intermediate vertebrate host in which asexual multiplication (schizogeny) occurs (Garnham, 1966). Sporozoans are found within the blood
cell and three genera of the family Plasmodiidae are of importance:

Plasmodium, Haemoproteus and Leucocytozoon (Levine, 1961).

The class Zoomastigophora includes the genus <u>Trypanosoma</u> which inhabits the body fluid. Trypanosomes are flagellated, leaf-life protozoans and they multiply by binary fission (Manwell, 1968).

Metazoan hemoparasites of birds are microfilaria belonging to the class Nematoda. Microfilaria contain a heterogenous assortment of forms that are only known in the larval stage (Levine, 1968).

Class Sporozoasida Genus: Plasmodium

The genus <u>Plasmodium</u> was first reported from avian hosts by Danil-evskii in 1885 but not until 1913 was a strain from sparrows actually isolated (Herman, 1944). It is most readily observed within erythrocytes and has been the subject of numerous biological studies because of its similarity to the malarias of man.

The first report of <u>Plasmodium</u> in turkeys was from Kenya (Herman, 1941) where the parasite was described as extremely pathogenic and fatal to domestic poults. The new species was called <u>P. durae</u>. It forms elongate gamonts which often occupy an oblique position at the polar extremity of the host cell.

Garnham (1966) considered <u>P</u>. <u>durae</u> to be a natural parasite of African birds; turkey infections therefore represented a veterinary zoonosis acquired from a feral source. This would explain why no <u>Plasmodium</u> had been recognized in natural populations of wild turkeys in the new world until 1975. Telford and Forrester (1975) found a parasite in wild turkeys in Florida and called it <u>P</u>. <u>hermani</u>. The parasite bears some morphological similarities to <u>P</u>. <u>durae</u> but exhibits low parasitemia and is nonlethal.

<u>Plasmodium</u> must have a mosquito vector of the species <u>Aedes</u> or <u>Culex</u> to complete its life cycle. Accounts of this life history can be found in Garnham (1966) or Hall (1953). Species of <u>Plasmodium</u> differ in their number of erythrocytic schizogonic cycles, their periodicity and consequently their pathogenicity. When the erythrocytes are destroyed at the end of schizogeny, merozoites, metabolic byproducts and hemoglobin are released into the plasma. This results in the characteristic fever (Weinman and Ristic, 1968).

Avian malarias respond to treatment with chloroquine; however, as a practical matter chemotherapy is impossible in wild birds. Preventive measures such as vector control are recommended instead (Levine, 1961).

Class Sporozoasida
Genus: Haemoproteus

The genus <u>Haemoproteus</u> causes a malaria-like disease primarily in birds but may occur rarely in reptiles. Levine (1961) named the one species of <u>Haemoproteus</u> found in turkeys in the United States <u>H. meleagridis</u> and summarized information on this species.

Haemoproteus was first reported in wild turkeys of Pennsylvania (Kozicky, 1948) where 5.2% of 97 turkeys were infected. Since then the parasite has been found in 1 of 2 wild turkeys in Georgia (Love et al., 1953), in 13% of 76 birds in South Carolina (Noblet and Moore, 1975), as well as in 9 of 46 turkeys in Alabama, Mississippi, Arkansas and West Virginia (Eve et al., 1972b). Forrester et al. (1974) reported 84% of 605 wild turkeys from Florida infected and Cook et al. (1966) recorded an 80% infection in 133 birds from Texas.

The species has been found in domestic turkeys from the District of Columbia (Wetmore, 1941), Texas (Morehouse, 1945), North Dakota (Goldsby, 1951), Georgia (Atchley, 1951) and South Carolina (Bierer et al., 1959).

Immature turkeys tend to have higher levels of infection, but the pathogenicity of  $\underline{H}$ .  $\underline{meleagridis}$  is unknown. It is apparently not an important cause of disease in wild birds and as a result parasitemias observed by other workers may have gone unreported (Cook et al., 1966).

Haemoproteus parasitizes erythrocytes in one stage of its development but in other stages occurs only in internal organs (Herman, 1944). The sexual stages are found in erythrocytes and here the gamonts are picked up by the arthropod vector when feeding on the vertebrate. Vectors are hippoboscid flies of the genus <u>Pseudolynchia</u> or biting midges of the genus <u>Culicoides</u>. A detailed description of the life history can be found in Weinman and Ristic (1968).

The gamonts generally are sausage-shaped, filling about one half the cytoplasm of the erythrocyte and curving slightly around each end of the nucleus. This is the typical "Halteridium" form. Pigment is present, indicating ingestion of hemoglobin (Bierer et al., 1959).

Electron microscope studies suggest another theory about the location of the halteridium. Instead of the parasite being inside the cell it is thought to be attached to the surface of the erythrocyte (B. T. Ridgeway, personal communication).

## Class Sporozoasida Genus: Leucocytozoon

In 1890 Danilevskii gave the name <u>Leucocytozoon</u> to parasites which he found in the blood of birds. Since then 58 species of <u>Leucocytozoon</u> have been described (Weinman and Ristic, 1968). Only one species has been reported in the wild and domestic turkey.

Leucocytozoons were first reported in turkeys by Theobald Smith in 1895 in domestic birds from Rhode Island and Massachusetts. Ten years later, Laveran and Lucet (1905) observed similar organisms in France and named them <u>L. smithi</u> in honor of the American scientist. The parasite has also been reported in domestic turkeys in Minnesota and North Dakota (Volckmar, 1929), Nebraska (Skidmore, 1932), and Alabama, South Carolina, Florida and Missouri (Travis et al., 1939).

Mosby and Handley (1943) first reported the occurrence of <u>L. smithi</u> in wild turkeys of Virginia. They found 40% of 45 wild birds infected.

Noblet and Moore (1975) subsequently reported them in South Carolina where 43% of 76 turkeys were infected. Kozicky (1948) found 100% of 5 birds in Pennsylvania and Banks (1943) 28 out of 106 birds in Texas infected.

These reports suggest that <u>L</u>. <u>smithi</u> has a wide distribution in both wild and domestic turkeys. Domestic turkeys and poults are more susceptible to infections than wild turkeys, possibly due to the fact that wild birds, having long been exposed to infections, have developed some degree of immunity (Byrd, 1959). In domestic turkeys the parasite causes droopiness, loss of appetite, incoordination, diarrhea and occasionally convulsions that end in death (Johnson et al., 1938). The acute stage usually lasts two to three days, after which the birds die or start to recover. Recovered birds may carry the organism in their blood and serve as carriers, being largely responsible for perpetuation of the disease from year to year (Wehr, 1962).

Jones et al. (1972) showed that turkey hens infected with  $\underline{L}$ .  $\underline{smithi}$  had a highly significant decrease in egg production and hatch of fertile eggs as compared to healthy hens. The infection had no effect on fertility, however. Overall the disease causes large economic losses to the turkey industry.

Control is possible only through flock management. Turkeys should not be raised in regions where there is running water which serves as a breeding place for blackflies of the genus <u>Simulium</u>, the only known vector at the present time. Removal of parasitized birds from the flock would also limit the rate of infection (Biester and Schwarte, 1943).

Leucocytozoon smithi is an elongated, oval or cigar-shaped organism. The host cell normally is a leucocyte but can also be a young erythrocyte and the invasion of the parasite results in a spindle-shaped blood cell. The host cell nucleus becomes either bean-shaped or split with one half being displaced to each side of the cell. The whole cell stretches to about four to five times its normal length (Volckmar, 1929).

Leucocytozoon sporozoites enter the blood of the turkey during the bite of an infected blackfly. They then develop into two different kinds of schizonts. One is found in cells of the liver of about 20 µL length which does not enlarge the nucleus; the other type occurs in bloodvessels, is called megaloschizont, is 80-170 µL long and does enlarge the nucleus (Newberne, 1955). A detailed life history of Leucocytozoon can be found in Atchley (1951).

Class Zoomastigophora Genus: Trypanosoma

This genus is widespread in birds and the relatively few species look very much alike. They become large,  $20-60\mu$  long and the body is often striated. The species found in fowl was named  $\underline{T}$ .  $\underline{gallinarum}$  by Bruce et al. (1911). Only one trypanosome organism has been reported in the blood of a wild turkey in West Virginia (Eve et al., 1972a).

Only one complete life cycle has been worked out by Baker and others in 1956 for <u>T. avium</u>; others are probably similar. The avian host becomes infected when ingesting an infected insect (mosquito and hippoboscids). No multiplication occurs in the bird. This would account for the sparse number of trypanosomes in the blood, especially in turkeys, since their diet usually does not consist of small biting insects (Levine, 1961).

Class Nematoda Genus: <u>Singhfilaria</u>

Filariid infections are not of great importance in domestic birds, but they are common in many wild birds. Anderson and Prestwood (1969) reported the first occurrence of microfilaria of the genus Singhfilaria

in 27 of 52 wild turkeys from Alabama, Arkansas and Mississippi. The new species was named S. hayesi.

They are medium-sized worms, about 150 µ long and the anterior half of the body is slightly wider than the posterior half. The cuticle is raised with innumerable thickenings. They have well defined sheaths and the posterior extremity is rounded. Filariid worms are transmitted by bloodsucking arthropods and a majority are viviparous, releasing microfilaria into the tissue of their vertebrate host. A complete life history account can be found in Hyman (1951) and Levine (1968).

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