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Studies on Blood Parasites of Birds in Coles County, Illinois

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Eastern Illinois University

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STUDIES ON BLOOD PARASITES OF BIRDS

IN COLES COUNTY, ILLINOIS

(TITLE)

BY

Edward G. Fox
B.S. University of Illinois, 1966

THESIS

SUBMITTED IN PARTIAL FULFILLMENT OF THE REQUIREMENTS
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I HEREBY RECOMMEND THIS THESIS BE ACCEPTED AS FULFILLING
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INTRODUCTION

During the Spring of 1967, as part of a problems course, a survey of the blood parasites in some birds of Coles County, Illinois, was carried out and the following result reported.

Seventy house sparrows (Passer domesticus) were captured. Examination of blood smears from these birds revealed a mixed infection of Plasmodium and an unknown microfilariae in one bird, a very light infection of Leucocytozoon in another bird, and six sparrows with Haemoproteus infections.

Hewitt (1940) indicated that Plasmodium infections in birds, in all but a few instances, are characterized by patent periods (the parasite being demonstrable in peripheral blood), followed by periods of indefinite latency (the parasite not demonstrable). It has been shown (Sergent, 1920) that while latent birds are refractory to further infection, sub-inoculation of blood from latent animals into previously uninfected birds results in patent infection of the recipient. This procedure is used to detect latent infections and is called isodiagnosis.

The blood smear sampling technique, used in the survey just described, is effective only for the detection of patent infections and yields no data concerning latent or non-blood tissue infections. According to Cameron (1956) there is no known immunological or other diagnostic procedure for discovering previous infection with microfilariae. The same holds for Haemoproteus. Appearance of these two organisms in circulating blood is the only sure diagnosis.

With the above as a background it was considered feasible that a survey of birds in Coles County, Illinois, using a combination of blood smear techniques and isodiagnostic procedures might yield more information regarding the extent of blood parasitic infection in the bird population, than a survey utilizing blood smears exclusively. Accordingly the present study was undertaken.

LITERATURE REVIEW

History of Avian Blood Parasites

The first published account of blood parasites is perhaps that of Lankester (Opie, 1898) who described, in 1871, sporozoite-like organisms seen in the blood of a frog. In later studies of frogs, Lankester noted the presence of intracorpuseular bodies, now believed to have been malarial parasites. Similar observations were recorded by Gaule in 1880 when he reported seeing "Wurmachen" (literally worm-like animals) in the erythrocytes of frogs. It could be that Gaule was referring to plasmodial sporozoite stages (Opie, 1898).

Laveran, in 1880, described malarial parasites observed in the blood of a French soldier. Five years later Danilewsky's account of intracellular parasites in avian erythrocytes was published. This study based on observation of a 300 bird colony, concluded that malaria was transmitted "per os", and that malaria in man and birds was caused by the same organism.

Marchiafava and Celli proposed the generic name Plasmodium in 1885 to include mammalian and avian malarial parasites. Between 1885 and 1890 numerous descriptions of hemosporidians from birds were reported. Many of these reports appear to be descriptions of Haemoproteus, fewer being Plasmodium (Hewitt, 1940).

Grassi and Feletti distinguished between distinctly different intracorpuseular parasites of sparrows and pigeons. The

halter-shaped form of sparrows they named Laverania, but priority has been given to Haemoproteus, a name proposed by Kruse, in 1890, earlier. The name Haemamoeba praecox was suggested for the oval and crescent-shaped intracorpuseular parasites of pigeons, and was applied to both avian and mammalian malaria. Grassie and Feletti were soon convinced that avian and mammalian forms were different and in 1891 suggested the name Plasmodium relictum for malarial organisms in birds.

Celli and San Felice, in 1891, undertook a study to clarify the taxonomy of avian malaria, but were hampered because they did not recognize mixed infections. These workers were the first to successfully transfer malarial parasites by blood inoculation, and they believed that each avian species harbored its own individual hemosporidian species. Labbe proposed another view in 1894. He suggested Protesoma grassei as the name for all plasmodial infections, and Halteridium danilewskyi for all avial halteridial infections. Labbe's proposal has not been accepted (Hewitt, 1940).

According to Bishop (1955), it was Ross who, in 1897, elucidated the life history of Plasmodium. He discovered the mosquito vector by following the development of sporozoites in the oocyst and observing the subsequent infection of weaver birds and sparrows.

Knowledge of the reproductive cycles of other hemosporidian parasites was gathered by Opie and MacCallum, the first Americans to investigate avian malaria. Opie (1898) described the

differences between Haemoproteus gametocytes and observed ex-flagellation. However, he was not able to decide whether this latter phenomenon was associated with one species, displaying a remarkable polymorphism, or indicative of a different, but concurrent, species. MacCallum (1898), working with the same group of infected birds as Opie, noted pathological changes occurring in visceral organs of birds infected with various haemosporidians. He observed pigment masses and areas of necrosis in the liver and enlargement and darkening of the spleen. In addition, changes in the bone marrow and lung tissue were described. MacCallum also described the phagocytic digestive sequence of infected erythrocytes and, at another time, the fertilization process in Plasmodium falciparum.

Whitmore (1918), in a continuation of Koch's work on transmission of malaria in laboratory canaries, demonstrated continued latency in previously infected canaries for up to three years. His studies on relapse phenomena resulted in the conclusion that relapse could be triggered by lowered resistance of the host and the belief that immunity lasted only as long as the bird remained infected.

Sargent (1920), following Whitmore's work, elucidated five methods for uncovering "La phase chronique" in sub-clinical (latent) infections. In brief these are:

- (1) Immune reaction - Plasmodial infected blood was intraperitoneally injected into a suspect animal. If no reaction occurred (infection), the host was already immune and thus had

previously contracted the disease.

(2) Xenodiagnosis - Culex species were permitted to feed on the suspected host and several days later checked for the presence of sporozont stages.

(3) Recrudescence of infection - the suspect host was made to undergo periods of cold or heat, or human blood injections, in order to lower the corporal resistance.

(4) Splenodiagnosis - a very good index was found to be the conditions of the spleen at autopsy.

(5) Isodiagnosis - blood from a suspect bird might be injected into a canary and then possibly sub-inoculated into a second canary.

For almost twenty years following 1920, field studies on avian malaria were primarily concerned with types and extent of the disease in natural populations. One result of such investigations was the characterization of a few new genera of Haemosporidia and the description of several "new" species. More detail on the work of this period will be covered in sections of the present review dealing specifically with particular genera.

In 1942 a Committee on Terminology and Strains of Avian Malarías was formed to systematize and categorize the work of the previous twenty years. This committee concluded that thirteen distinct species of Plasmodium were evident, with numerous strains and sub-strains. Little was done with genera other than Plasmodium.

Experimental research on avian malaria, while progressing since the very discovery of the causative organisms, was greatly increased during the period following the beginning of World War II. Human malaria was a pressing problem, and avian malarial parasites were excellent screening agents for antimalarial drugs, and had, according to Huff (1963), been involved as investigational tools since the beginning of malarial chemotherapy research. The discovery of P. gallinaceum in 1935, P. lophorae in 1938, both of which are capable of infecting domestic poultry, facilitated the use of bird malaria in screening programs. The total dependency on avian species was discontinued with the discovery of P. berghei in rodents (Chandler, 1961).

The most recent thrust of research effort on avian malarial parasites has been toward an understanding of immunity and susceptibility in vertebrate and invertebrate host. This problem has been approached by studies on the growth, morphology, and chemical composition of parasites. The nature of the intracellular restriction has been investigated, especially with regard to the metabolic dependence of parasites on hosts.

The work on avian malaria has been the subject of two extensive reviews. Hewitt (1940) summarized the information existing up to the time of his writing. Huff (1963) followed with compilations of subsequent work. The latter review places special emphasis on experimental advances.

Avian Plasmodium

Thirty-one species of avian Plasmodium have been characterized

in the literature, and of these 12 are universally accepted (Ridgeway, 1966). These species include not only the ten species listed in Hewitt (1940), but species recently described such as P. gallinaceum and P. lophorae. A number of descriptions have been published indicating developmental variations within species dependent upon the host. These variants are referred to as strains and at present more than 60 have been noted, a great number occurring within the species P. relictum (Ridgeway, 1966).

Attempting to categorize these intracorpuseular parasites into a more useful classification, Corradetti, et al. (1963) further divided the genus into four sub-genera. He based his systematics on the differences in the erythrocytic stages, exo-erythrocytic stages, and vertebrate-host specificity.

Three of the sub-genera have their exo-erythrocytic schizonts located in the lymphoid-macrophage system.

(a) The Haemaboeba are characterized by round gametocytes and large schizonts in the erythrocytes. Type species - Plasmodium relictum Grassi and Feletti, 1891.

(b) The Giovannolaia have elongated gametocytes and large erythrocytic schizonts. It is further sub-divided into the Passerine - type species Plasmodium circumflexum Kikuth, 1931 - and the Game Birds - type species Plasmodium lophurae Coggeshall, 1938.

(c) The Novyella have elongated gametocytes and small schizonts containing a maximum of eight merozoites.

These are also further sub-divided into the Passeriformes - type species Plasmodium vauhani Novy and MacNeal, 1904 - and the Galliformes - type species Plasmodium juxtannucleare Versiani and Gomes, 1941.

The fourth sub-genus has its exo-erythrocytic stages in the hemopoietic system.

(d) The Huffia have small schizonts with a maximum of ten merozoites and elongated gametocytes in the erythrocytes. The type species is Plasmodium elongatum Huff, 1930.

To illustrate the life-cycle of a typical Plasmodium, Chandler's (1961) account has been generalized. The parasite must have an invertebrate vector, consisting of various species of Aedes or Culex in birds and reptiles, and Anopheles in mammals, and a vertebrate host. The ontological cycle may be said to begin with the passing of macrogametocytes and microgametocytes from the host's circulatory system into the stomach of the invertebrate vector during the blood meal. Exflagellation occurs in the gut as does the union of the microgamete and macrogamete and a zygote is formed. The zygote develops into an ookinete which burrows into the stomach wall forming an oocyst. Sporoblasts may be seen developing in the oocysts eventually releasing mature sporozoites into the hemocoel, and these travelling to the salivary glands. With the insect's next blood meal, the sporozoites are injected into the vertebrate host. The sporozoites are then transported by the circulatory system to the endothelial or hemopoietic organs where

the exo-erythrocytic stages of the parasite take place. Cryptozoites (phanerozoites) are formed in the cells of the invaded tissues; metacryptozoites are the second generation located in the same or similar tissues. Trophozoites are then returned to the circulatory system to invade the erythrocytes. Schizonts with merozoites are usually produced in these cells but occasionally gametocytes are produced and the cycle begins anew.

Haemoproteus

Kruse, 1890, was the first investigator to describe the Haemoproteus organism, his type specimen, Haemoproteus columbae, being obtained from Domestic Pigeons in Naples (Garnham, 1966). The Haemoproteids are never found in mammals, but are common in birds, and less common in reptiles. Many species of this genus have been reported from various birds, distinguished not so much because of their morphological features as of their incidence in certain avian species. Garnham (1966) taking this into account stated his reasons for placing all the lesser characterized avian species under one name. "The appearance of the gametocyte in the blood offers few clues for specific differentiation, and still less for separation into two genera. For this reason, the species found in birds will continue to be known under the name of Haemoproteus sensu latu, unless the life-cycle is known."

Kruse's original Haemoproteus has since been enlarged into four genera by Bennett et al. (1965) and given the family

name Haemoproteidae. Two non-avian genera are included in the family. Haemocystidium, Castellani and Willey 1904, are pigmented parasites of lizards with the blood stages infecting erythrocytes, and Simondia Garnham 1966, which includes the parasites of chelonians.

The two genera infecting avian hosts are Haemoproteus and Parahaemoproteus Wenyon 1926. Haemoproteus has exo-erythrocytic stages that are located in vascular endothelium and are not restricted to localized foci. H. columbae is a typical species; hippoboscids are the suggested vectors. Parahaemoproteus is chiefly differentiated on the basis of having unpigmented schizont cytomeres localized in the lung, liver, and kidney, and the vectors are members of the Ceratopogonidae, P. danilewskyi, Kruse 1890, is the type species.

The life-cycle of the Haemoproteidae is typified by H. columbae. There has been found to be an invertebrate vector and a vertebrate host involved in the cycle. The ontogeny of the parasite may be said to begin when the insect vector feeds and inadvertently incorporates gametocytes from the host's blood. The microgametes rapidly become active and fertilize the macrogamete. The resultant zygote develops into a motile ookinete which then burrows through the mid-gut epithelium, encysting between the muscular layers to form an oocyst. The oocyst grows and enlarges, due to the multiplying and maturing of the internal sporozoites, until it eventually ruptures. The freed sporozoites migrate to the salivary gland, appearing in

this gland approximately ten days after ingestion. With the next feeding, the sporozoites are injected into the vertebrate host. The exo-erythrocytic stages then develop. In the case of H. columbae these may be either macro- or micro-schizonts in the endothelium, or in the case of a Parahaemoproteus the exo-erythrocytic stages will grow as large compartmentalized foci in the visceral organs. After a period of 25 days of the insect's bite, merozoites will be freed into the circulatory system. A few of these may return to reinfect the tissues, but the plurality of the organisms parasitize the erythrocytes and these undergo gametogenesis to complete the cycle.

Leucocytozoon

Credit for establishing the genus Leucocytozoon commonly (Mohammed, 1958; Kudo, 1966; Levine, 1961; Hewit, 1940) is given to Danilewsky 1890, but according to Garnham (1966) the discovery should be attributed to Berestnev 1904. This genus of parasites utilizes avian species exclusively as its vertebrate host (Manwell, 1935) and Simuliid Flies as the definitive vector (Fallis et al., 1951).

Mohammed (1958), drawing on the ideas of Fallis (1951), Manwell and Hermann (1935a), and results of his own studies concluded that the incidence of Leucocytozoon is not worldwide. Its distribution is correlated to the distribution of the Black Fly vectors which require very specific conditions of rapidly flowing streams with rocky bottoms for the deposition and attachment of larvae.

The original genus Leucocytozoon has been elevated to the family Leucocytozoidae. The family has two genera, the Leucocytozoon Berestnev 1904, and Akiba Fallis et al. 1965. Bennett, Garnham, and Fallis (1965) have devised a system for classifying the unpigmented parasites in order to cope with the known differentiating characteristics of the group.

The genus Leucocytozoon is characterized by simuliids as vectors, sporozoites with one pointed end, and megaloschizonts with a ventrally placed host cell nucleus. The type species is L. danilewskyi Zieman, 1898. Akiba is characterized by Culicoides as a vector, sporozoites with two tapered ends, megaloschizonts with a laterally placed host cell nucleus. A. caulleryi, Mathis and Leger 1909, is a typical example.

A general description of the life history of a typical member of the Leucocytozoidae is described as illustrated by Garnham (1966). Gametogenesis occurs in lymphoid-macrophage cells and erythrocytic cells of the vertebrate host. The parasitized cells may be ingested by the feeding simuliid or culicoid insect vector. Exflagellation of the microgametocyte occurs--eight microgametes being released per microgametocyte--and the mobile gametes fertilize the immobile macrogamete. The resulting zygote elongates to form the slightly motile ookinete (which is formed within twelve hours of the initial ingestion). The ookinete burrows into an epithelial cell of the hemocoel forming an oocyst which gives off sporozoites. These sporozoites travel to the salivary glands, or directly

to the proboscis. Garnham (1966) listed some particularly striking features of the Leucocytozoidae sporogony which include: (1) the rapidity, complete sporogony within less than five days at 22°C; (2) the small size of the oocyst when mature - 12 μ ; (3) the small number of sporozoites per oocyst - 30-50; (4) the slow discharge of sporozoites from the oocysts.

When feeding upon the vertebrate host the infected vector will inject the sporozoites into the prey's vascular system. They are then carried to any one of a number of locations whereupon the exo-erythrocytic stage of development begins. Two different types of schizonts develop, apparently concurrently. The smaller hepatic schizonts that mature in the parenchymal and macrophage cells of the liver grow only up to 20 μ in length and the host cell's nucleus does not enlarge. The megaloschizonts, which may appear as early as four days after injection, may grow in the spleen, heart, liver, brain, lungs, lymph glands, adrenals, thymus, thyroid, and gonads. They may vary in size from 80-170 μ and contain at least a million merozoites. The host cell's nucleus enlarges even when the parasite is quite young, a clear differentiating feature apparently due to an initial generic signal from the parasitic genotype, as yet undetermined.

When the merozoites are released from these schizonts they are free to infect either the erythrocytic or lymphoid-macrophage cells, where full gametocytic maturation occurs. Fallis et al. (1951) found that the pre-patent period may be less than 5-1/2 days with the acme of the infection being reached in 8-1/2 days.

He and his co-workers reported that they were able to transfer the infection by blood inoculations. This was in direct contradiction to Hewitt (1940) who stated that the infection was not blood transferable. Fallis stated in his paper that their success was probably due to the schizonts in the lymphocytes and the free merozoites which eventually come to rest or infect the circulating cells and form gametocytes.

As opposed to the Haemoproteidae, the Leucocytozoidae are the most important of the hemal protozoa in veterinary medicine. Fallis et al. (1951) noticed that many of the ducks they were observing died from the infection during the months of June and July concurrently when the Black Flies were most abundant. Death in these cases is directly due to the destruction of vital tissue elements, anemia, or shock. Measurable changes in the blood include those of anemia and leucocytosis--sometimes a tripling of the leucocytic elements for a period of up to two weeks. Several of the visceral organs are affected including the liver which may become enlarged and necrotic, and the spleen may be hypertrophied up to twenty times its normal size. Death may occur less than 24 hours after the onset of the initial symptoms.

Avian Microfilariae

Microfilaria is a collective term referring to the circulating blood forms of embryonic nematodes which belong to the class Nematoda, subclass Phasmodia, and order Filarioidea, as classified by Cheng (1964). The adults are filiform worms of

moderate to large size, with the males considerably smaller than the female (Hyman, 1951). They may be located, depending on the species, in the blood, lymph cysts, subcutaneous tissues, or the body cavities (Cameron, 1956). Parturition is either ovoviviparous, in which case the embryo may or may not be enclosed in a sheath (the remains of the egg shell), or oviparous.

The first record of human filarial worms observed was by Pigafetta (Odetoyinbo, 1960) who mentioned an apparent Loa loa infection in his book Travels in the Congo, published in 1589. Wiesenenthal in 1799 was supposedly the first investigator to find an avian filarioid. However, according to Skrjabin (1961), Morle in his book, History of Veterinary Medicine, made reference to a fifteenth century work by Guillaume Tardif on falconary in which he observed a symptom of "frequent gaping" caused by worms in the trachea.

Manson's discovery of the complete life-cycle of Wuchereria bancrofti, the causative organism of elephantiasis, was the first to positively indict insects as vectors of disease, leading the way for Ross' discovery of the Anopheline Mosquitoes as the vector of malaria.

The life history of filarial nematodes is well known. The female adult filarial worm produces the first form embryo, either sheathed or unsheathed, measuring between 0.2 to 0.4 mm in length. When the insect vector feeds on the vertebrate host it may ingest the microfilaria. (It is interesting to note here that Hawking (1962) noticed a greater concentration of the embryos in the

vector immediately after feeding than were found in a comparable amount of blood from the host. He theorized that perhaps there was some sort of chemotactic effect produced by the vector's saliva.) The embryo then loses its sheath and becomes established in the thoracic muscles where it metamorphosises into a short sausage-shaped body. It undergoes a second molt before migrating into the vector's proboscis. When next the vector feeds, the larva is deposited on the epidermis of the host where it then burrows into the skin until the circulatory system is reached, then being carried to the infective site and two final molts occurring. There is no reproduction in the insect. It serves only as a mechanical vector, the only reproduction taking place in the vertebrate host, be that amphibian, reptilian, avian, or mammalian.

One of the most intriguing questions in the realm of parasitology has to do with the as yet unexplained phenomenon of the periodicity of the first-form embryos, the microfilaria, demonstrated by many filarial worms. Other cases of parasitic periodicity are rare in nature. The oocysts of Isospora have been found to appear more in the feces at one time of day, in sparrows, than at other times. The ancient schizogonic periodicity of the malarial organisms is as yet unexplained. Hawking (1962) mentioned a possible reason being that the parasites perhaps took advantage of the heightened presence of certain DNA precursors at one time of day. The periodicity of the microfilarioid worms has at times produced some bizarre explanations.

Incidental Avian Hemal Parasites

Other intracorpuseular hemal parasites have been observed in avian blood. These include the Toxoplasma, Hepatozoans, Microsporidians, Piroplasmas, and the unknown category of intraleucocytic parasites. The last being a kind of "catch-all" group.

Toxoplasma was discovered by Nicolle and Manceaux, 1908, in a North African rodent, Ctenodactylus gondi, (Faust et al., 1962). Herman (1937) attributed the original discovery to Splendore in 1908. The first Toxoplasma infection of a North American bird, Passer d. domesticus, was reported in 1934 (Herman, 1937). Since that time several cases of avian Toxoplasma infection have been reported. They are occasionally found in the peripheral blood infecting white cells (lymphocytes or polymorphonuclear leucocytes), but usually are located in the liver, spleen, and lungs of birds and mammals. The shape is characteristically vermiform with slightly pointed ends and no hematin is present in the cell. The infection is usually pathogenic and may be fatal. Blood transfers in avian hosts have been attempted without success (Hewitt, 1940). Several workers reported that animals became infected when fed the large intracellular "pseudocysts" (Chandler, 1961) of the Toxoplasma. The life-history of these organisms is an enigma. Mechanical insect vector activity was intimated by Lairman in 1956 when he allowed stable flies and fleas to feed on infected animals (Faust, et al., 1962). When these insects were subsequently fed to other animals,

infection resulted. Hegner and Wolfson (1938) theorized that infections of Plasmodium and Toxoplasma might produce Toxoplasma-like organisms; evidence for this has never been brought forth. Herman (1944) placed Toxoplasma in the family Haemogregarinidae.

Another genus of this family, the Hepatozoa, are parasites present in birds and mammals. Schizogony occurs in the reticulo-endothelial cells of the liver, spleen, or bone marrow, and the gametocytes mature in the circulating monocytes where they resemble Haemoproteus, but are distinguished by the absence of pigment. Ticks and mites have been implicated as the intermediate hosts (Levine, 1961).

Mohammed (1958) mentioned Bethram's 1944 discovery of Piroplasmosis in Colinus virginianus. This is perhaps the only record of this parasite in birds.

Many reports of Haemoflagellates, collectively referred to as either Trypanosoma sp., or Trypanosoma avian, are found in the literature. They are not intracorpuseular, being habitants of plasma. No pathogenic effects have been witnessed in birds (Herman, 1944).

MATERIALS AND METHODS

General Considerations

The present study was carried out from June through December of 1967. Birds utilized in the survey were obtained from a number of locations in Coles County, Illinois. Traps were set up on the T. Phipps farm located at "southeast $\frac{1}{4}$ of section 16, Township 11 north, R-9E" south of Charleston. Here, also mist nets were erected in flyways into and among farm buildings. The upper floors of a vacant three story hotel in downtown Mattoon, Illinois, harbors a large population of pigeons (Columba livia) which gain access through broken windows. Birds were collected from this source by snaring with a hand net. Some sparrows were donated by citizens in and about Charleston, Illinois. These animals had been captured in live traps set up in yards, with the view that elimination of sparrows would encourage the presence of more aesthetic song birds. A few birds were contributed by faculty and students who picked them up, incidentally, in the course of field work. In every case, with one exception, the birds were released, usually in the area where captured, after enough blood was obtained for smears and isodiagnosis. The exception being the few grackles that were obtained by shooting. Before its release each bird was examined to determine the general state of its health. Routine examination was made for gross external parasites, deformities, and condition of breast muscles.

Trapping

On the Phipps farm five wire funnel traps were set up in various settings along the periphery of a corn field. These

small automatic funnel traps are modeled after those described by Quay (1951), but with a modified, narrower, access tunnel. The traps were baited with commercial wild bird seed, scattered about just inside the mouth of the access tunnel. All traps were tended twice each day during the duration of the study.

Black nylon, one inch mesh, mist nets were erected across the entrances of two farm buildings. These were set just inside the structure so that shading would make them less conspicuous, and perhaps more effective. Another net was utilized in flyways among trees in a small orchard adjacent to the farm buildings. Nets were checked at four hour intervals during the entire period they were in place. Every effort was made to remove birds as soon as possible after they became entangled in a net. Mist netting procedures were used for periods of 48 hours, discontinued for four days, and then redone. This method was used only during parts of June and July of 1967.

Another method utilized intermittently was the capturing of House Sparrows (Passer domesticus) while they were roosting during the evening hours. Several locations in the Phipps' area were inspected, comprising almost every niche that was available and protected. These included sheds, chicken houses, barns, house eaves, and even laundry poles. With the aid of a flashlight and a ladder, it was possible to pick some birds off the roost. In one instance, all but one exit to a machine shed were closed and a mist net set up in front of the open exit. An automobile headlight was directed into the exit to attract the birds to the exit. Attempts were also made to dislodge them from their

roosting sites in the building. Once in the net, the birds were immediately removed.

A novel method of live-catching attempted was that elucidated by McIlhenny (1942). This consisted of driving a car at night through an open field and thus flushing any ground dwelling birds. The birds would then fly only a short distance. One could approach them on foot, and while directing a hand held light, which would supposedly act as a mesmerizing agent, net the specimens. A special net was constructed for this procedure. It consisted of two 15-foot bamboo poles shaped into a giant "badminton" racket with a seining net and a wooden supporting bar. This was the least effective method tried and was soon abandoned.

Pigeons gathered from the hotel site were netted using long handled aluminum, 18-inch nets of the type used by fishermen for landing fish. Flushed birds were "snagged" in flight as they flew through the hallways, and temporarily caged in portable wire cages for transport to the laboratory. Usual precautions were taken to prevent contracting pigeon-spread respiratory diseases.

Experimental Canaries

Canaries (Serinas canarius) used as hosts for isodiagnosis procedures were obtained from the American Bird Company, Chicago, Illinois, and the National Pet Supply Company, St. Louis, Missouri. Each bird was carefully examined for evidence of disease prior to being housed in laboratory flight cages.

Water and a balanced ration were constantly available. All canaries were females and presumably infection free. In all cases, birds selected for experiments were inspected for ectoparasites and pre-infection blood films made and examined. Canaries actually being utilized in isodiagnosis were housed in separate individual cages.

Description of Techniques

(a) Blood Films and Parasite Counts: Blood films were made from blood obtained by making a small incision, in the toe of the bird, with a lancet. In the case of canaries, and other small birds, blood was obtained by puncturing a wing or leg vein. Following a period of air drying for 20 minutes, the films were fixed by placing them in absolute methyl alcohol for three minutes. When removed from the alcohol, slides were air dried and immersed in Giemsa stain, prepared by diluting 1 ml of stock stain to 40 ml of distilled water. After a staining period of 40 minutes the slides were rinsed in water and air dried.

Examination of all stained blood films was accomplished using an American Optical binocular microscope with 10X oculars and 100X oil immersion objective.

Parasite counts were made using the method described by Ginrich (1932). This procedure has been demonstrated to have a probable error of 10%, which is sufficiently accurate for this study.

(b) Isodiagnostic Techniques: Transfer of blood between vertebrate hosts was accomplished using a 2 cc hypodermic syringe fitted with a 23 gauge needle. Blood was drawn from a wing vein, or a cardiac puncture, into a syringe containing enough heparin solution (1,000 units/cc distilled water) to wet the inside. When handling canaries, or other small birds, a 1 cc syringe and a 25 gauge needle were used according to the method of Hewitt (1940). In such birds, blood could best be drawn from and/or injected into the tarso-metatarsal vein. Isodiagnosis was used only after the usual blood smear methods of parasite detection had produced negative results. Because the number of experimental canaries available for use was limited and the number of some birds captured was great (i.e., Passer domesticus, the House Sparrow), pooled blood samples were used. One fourth cc of blood was drawn from each of a group of five birds being subjected to isodiagnosis. Animals in each group were of the same sex and of course the same species. Blood from the five donors was placed in a clean test tube, mixed thoroughly but gently, and drawn into a clean heparinized syringe. A recipient experimental canary was then inoculated with one-fourth cc of this sample. Donor group birds were kept together in a quarantine cage until the end of the experiment. Blood smears were taken daily (for 12 days) from the canary recipient, stained, and examined for evidence of parasites. Negative smears for 12 days were accepted as evidence that all five birds in the group were free of latent infections. The onset of a patent infection, on the other hand, indicated that at least one bird of the donor

group harbored a latent malarial infection. If the latter was the case, the isodiagnostic procedure was repeated, on a one donor to one recipient basis, in order to isolate the particular latent bird or birds in the donor group pool.

A variation in the isodiagnostic protocol, incorporating a method of dilution conveyed personally to the author by Schneider was used in the latter part of the study. No pooled samples were used, direct one-to-one transfers being feasible. Blood samples were obtained as usual. One four cc of blood was withdrawn and mixed at a 1:1 dilution with Solution "A" of Schneider's (1967) blood preserving medium. This medium is said to contain materials which will be anticoagulative and stable. The dilution was then spun down and the supernatant discarded. Blood cells were resuspended in two-tenth cc of Solution "A" and this mixture injected intraperitoneally into the experimental canaries.

(e) Histological Techniques: Canaries that died during the course of an infection, or following the patent phase of an infection, were autopsied and their tissues examined for phanerozoites. Small pieces of liver, brain, lung, spleen, and kidney were removed. The tissue was sliced and the cut surface applied lightly to a clean dry microscope slide. This impression was stained with Giemsa in the same manner used for blood films. The remaining tissue was then fixed with Zenker's fluid, washed in dilute iodine solution, and stored in 70% alcohol for later histological examination. Tissue impressions were examined using an oil immersion lens.

RESULTS AND DISCUSSION

Trapping and Collection

Various trapping procedures carried out from June through December of 1967, resulted in the capture of 114 birds. In addition, three specimens were obtained by shooting and six specimens collected as road injuries, displaced nestlings, and injured as a result of having flown into a local television tower. The total number of birds examined for blood parasites was 123.

Of the five trapping procedures utilized for collecting specimens for this survey, three were very successful, one moderately so, and one not successful at all. Table 1 illustrates the relative effectiveness of trapping methods.

Hand netting of 36 pigeons was most successful in terms of numbers (Table 2). The birds were virtually trapped in the abandoned building from which they were taken, and snaring was an easy task. The capture of 29 House Sparrows and two Bank Swallows by removing them from their roosting sites was surprisingly rewarding when one considers the energy expended. This procedure was applicable regardless of weather.

Mist netting was disappointing considering the total number of individuals caught. But the greatest variety of birds (six different species) were obtained using this method. By and large, the mist nets were most effective on sunny days, and when positioned just inside the doorways of darkened farm

Table 1
SUCCESS OF TRAPPING METHODS

<u>Method</u>	<u>Number Captured</u>	<u>% of Total</u>
Funnel Trap Cage	1	0.8
Hand Net	36	29.2
Roosting	31	25.1
Mist Net	12	9.7
Hav-a-hart Cage	34	27.6

Table 2

SPECIFIC COLLECTING RESULTS OF A BLOOD PARASITE SURVEY OF BIRDS IN COLES COUNTY, ILLINOIS

Common Name	Scientific Name	Numbers Obtained by Collecting Method							Total
		F.T.*	H.N.*	Rst.*	M.N.*	H.H.*	Shot	Other	
House Sparrow	<u>Passer domesticus</u>			29	4	33		3	69
Pigeon	<u>Columba livia</u>		36						36
Song Sparrow	<u>Melospiza melodia</u>				3	1			4
Grackle	<u>Quiscalus quiscula</u>						3		3
Bank Swallow	<u>Riparia riparia</u>			2	1				3
Barn Swallow	<u>Hirundo rustico</u>				2				2
Kinglet	<u>Regulus satrapa</u>							1	1
Brown Thrasher	<u>Toxostoma rufum</u>	1							1
Cardinal	<u>Richmondia cardinalis</u>				1				1
Gold Finch	<u>Spinus tristis</u>				1				1
Great Horned Owl	<u>Bubo virginianus</u>							1	1
Bobwhite Quail	<u>Colinus virginianus</u>							1	1
Total Collected--All Methods									123

*F.T. - Funnel Trap; H.N. - Hand Net; Rst. - Roosting; M.N. - Mist Net; H.H. - Hav-a-hart

buildings. It is possible that the pupillary responses of the birds were not sufficiently rapid to accommodate to the shadows as they flew into the buildings from bright sunlight. Conversely, on cloudy days this rapid adjustment was not necessary and the birds could see and avoid the nets.

Funnel traps were not as productive as anticipated, since the only captured specimen was a Brown Thrasher. A variation of the funnel cage trap, the repeating, displaced opening, balance trap, manufactured by the Hav-a-hart Trap Company, proved to be very effective for the capture of House Sparrows. No other kinds of birds were taken in this particular trap, and most of the captures were on cloudy days. This could have been due to the visual accessibility of the bait in the trap compared to other feeding materials during these periods. When comparing the circumstances under which the Hav-a-hart trapped birds and the mist netted birds were captured, the phenomenon of weather acting as a factor is conspicuous. It is suggested that any bird trapping program take into consideration not only time of day but meteorological features as well.

Kinds of Birds

The 123 birds utilized in this blood parasite survey include representatives of four orders, nine families, and eleven species. The order Passeriformes was represented by the greatest number of individual birds and the most families. Of this order, members of the family Ploceidae to

which Passer domesticus (the House Sparrow) belongs were most abundant. Sixty-nine individuals were taken of which 59% were females. Hirundinidae was represented by three Bank Swallows (Riparia riparia) and two Barn Swallows (Hirundo rustico). Four Song Sparrows (Melospiza melodia), one Cardinal (Richmondia cardinalis), and one Goldfinch (Spinus tristis) were all of the Fringillidae. Three Grackles (Quiscalus quiscula) of the Icteridae were also collected. The families Sylviidae (a golden crowned Kinglet, Regulus satrapa), Mimidae (a Brown Thrasher, Toxostoma rufum), of which one bird each was utilized, concludes the Passeriformes families. The three other orders present were: Strigiformes (Strigidae, Bubo virginianus, Great Horned Owl), Galliformes (Phasianidae, Colinus virginianus, Bobwhite Quail), and Columbiformes (Columbidae, Columba livia, Pigeon). One owl, one quail, and 36 pigeons were examined. Table 2 illustrates specific collecting results.

Incidence of Protozoan Blood Parasites

Twenty (16.2%) of the 123 birds examined were found to harbor hemal parasitic infections. Of the 20 cases, 17 were patent and were discovered using the usual blood smear techniques, while three latent infections were revealed using isodiagnostic procedures.

Haemosporidian parasites of the genera Haemoproteus, Plasmodium and Leucocytozoon were found. In addition, at least two kinds of microfilariae were observed in the plasma of House Sparrows and Grackles. The incidence of each kind

of parasitic infection present in the bird survey is as follows: Haemoproteus - 9.7%; Plasmodium - 4.85%; microfilarial infections - 2.45%; and Leucocytozoon - 0.8%. Table 3 tabulates the incidence of infection by number of different kinds of birds infected with particular parasites and shows percents positive. Pigeons demonstrated the greatest incidence of infection, with ten birds out of 36 examined having Haemoproteus in circulating erythrocytes.

Haemoproteus: The gametocytes of the Haemoproteus in pigeons compare morphologically to the descriptions of H. columbae given by Levine (1961). According to Wood and Herman (1943) and Mohammed (1958), this species is the most prevalent in pigeons, so it can be assumed that one is correct in calling it H. columbae. The infections were light in pigeons. Parasitemias ranged from 0.10% to 0.25%, based on the percent of erythrocytes infected.

Comparing the incidence factors of Haemoproteus of pigeons in the present survey with those in other studies, the findings are similar. Coatney and Roudabush (1937) after examining 451 pigeons found 11% with Haemoproteus; Mohammed (1958) reported 73.5% of 106 birds had the parasite. It would seem that the relatively high incidence of Haemoproteus in Columba livia depicts a rather well dispersed, entrenched, benign host-parasite relationship. Other hemal parasites have never gained such a foot-hold in the pigeon community. Plasmodium is fairly well known from the group (Ridgeway, 1966). Wood and Herman

Table 3.

INCIDENCE OF BLOOD PARASITES IN BIRDS EXAMINED DURING A SURVEY IN COLES COUNTY, ILLINOIS

Birds		Parasitic Infections				Total	
Systematic Position and Scientific Name	Common Name	Haemo- proteus	Plasmo- dium	Leuco- cytozoon	Micro- filaria	Exam- ined	Positive # %
Galliformes							
Phasianidae							
<u>Colinus</u> <u>virginianus</u>	Bobwhite Quail					1	0 0.0
Columbiformes							
Columbidae							
<u>Columba</u> <u>livia</u>	Domestic Pigeon	10	1			36	10* 27.8 T 27.8 H 2.78P
Strigiformes							
Strigidae							
<u>Bubo</u> <u>virginianus</u>	Great Horned Owl	1		1		1	1* 100.0
Passeriformes							
Hirundinidae							
<u>Riparia</u> <u>riparia</u>	Bank Swallow					3	0 0.0
<u>Hirundo</u> <u>rustico</u>	Barn Swallow					2	0 0.0
Mimidae							
<u>Toxostoma</u> <u>rufum</u>	Brown Thrasher	1				1	1 100.0

Table 3 (cont'd)

INCIDENCE OF BLOOD PARASITES IN BIRDS EXAMINED DURING A SURVEY IN COLES COUNTY, ILLINOIS

Birds		Parasitic Infections				Total		
Systematic Position and Scientific Name	Common Name	Haemo- proteus	Plasmo- dium	Leuco- cytozoon	Micro- filaria	Exam- ined	Positive #	%
Sylviidae <u>Regulus</u> <u>satrapa</u>	Golden- Crowned Kinglet					1	0	0.0
Icteridae <u>Quiscalus</u> <u>quiscula</u>	Common Grackle				1	3	1	33.3
Ploceidae <u>Passer</u> <u>domesticus</u>	House Sparrow		6		2	69	7*	10.1 8.7 P 2.9 M
Fringillidae <u>Melospiza</u> <u>melodia</u>	Song Sparrow					4	0	0.0
<u>Richmondia</u> <u>cardinalis</u>	Cardinal					1	0	0.0
<u>Spinus</u> <u>tristis</u>	Goldfinch					1	0	0.0
Totals		12	7	1	3	123	20	16.2 T 9.7 H 5.75P 0.81L 2.45M

*Double Infection

T - Total, H - Haemoproteus, P - Plasmodium, L - Leucocytozoon

M - Microfilaria

(1943) have a host-record of Leucocytozoon from a Band Tail Pigeon, and Levine (1961) mentions a Leucocytozoon record in a Domestic Pigeon. Trypanosoma has never been reported in pigeons.

Two other hosts, examined during the present study, were infected with Haemoproteus. A 0.40% parasitemia was observed in blood smears obtained from a Brown Thrasher (Toxostoma rufum) and the gametocytes resembled H. beckeri var. toxostoma of Coatney and Roudabush (1935). Haemoproteus has been recorded from the Brown Thrasher in Illinois. Huff (1939) found 29 birds out of 48 examined had patent infections. Other records in North America are included in the papers of Wetmore (1941) and Coatney (1938).

The one Great Horned Owl (Bubo virginianus) had a 0.50% parasitemia of what appeared to be Haemoproteus noctuae as described by Coatney and Roudabush (1937). This infection existed concurrently with an infection of Leucocytozoon smithi. Perhaps the most intensive recent survey of blood parasites in the Great Horned Owl, in the midwest, is that of Farmer (1960) who examined 25 hosts and found five with Haemoproteus and three with Leucocytozoon infections.

Leucocytozoon: The Great Horned Owl, referred to above, demonstrated the only infection of Leucocytozoon encountered during the present study. The morphology of this parasite corresponds to the descriptions of L. smithi outlined by Levine (1961). The parasitemia was 0.25% in the owl and the

gametocytes appeared to have not distorted the leucocyte host cells to the extent characteristic of many Leucocytozoon species. The host cells were enlarged, but not drawn out into the elongated spindles that are characteristic of L. simondi or L. caulleryi infections.

The absence of Leucocytozoon in the birds constituting this survey is not unusual. Sixty-nine of the total 123 birds surveyed were Passer domesticus. According to Herman (1944), the only record of Leucocytozoon in this species is a 1912 report by Maya and David. Thirty-six of the birds presently surveyed were Columba livia. The only mention of Leucocytozoon infection in pigeons is that of Wood and Herman (1943) in Band Tails and the mention by Levine (1961) of a South African record for Domestic Pigeons. Pigeons and House Sparrows composed 85% of the birds utilized in this study. Both are closely associated with the habitats of man and not the most likely hosts to be fed upon by numbers of Black Flies, the known vectors of Leucocytozoon.

Galliform birds are among the preferred hosts for Leucocytozoon infections. Levine (1961) mentions L. caulleryi and L. sabrazesi as utilizing domestic chickens as hosts. L. smithi is a common pathogen of turkeys, perhaps using wild Strigiform birds as reservoir hosts. L. bonasae is a parasite of Ruffed Grouse (Bonasa umbellus) (Kudo, 1966). Other undescribed species of Leucocytozoon have been reported from Jungle Fowl (Gallus lafayetti) and the Guinea Fowl (Numida meleagris).

However, the negative blood smear obtained from the only member of the Galliformes examined during the present study is not unusual. Not only is one host a very small sample on which to base a conclusion, but there is no record of Leucocytozoon in Bobwhite Quail (Collinus virginianus).

The fact that Leucocytozoon was absent in blood smears of passeriformes examined in this study is in keeping with the results obtained in previous surveys (Appendix 1). Data presented in Appendix 1 suggests that only the Grackle (Quiscalus quiscula) and the Cardinal (Richmondia cardinalis) are commonly parasitized with Leucocytozoon.

Plasmodium: Plasmodial infections were found in six of the 123 birds examined. All six positive hosts were Passer domesticus. Three of the plasmodial infections were patent and demonstrated by blood smears. The other three infections were latent infections revealed by isodiagnosis. One questionable infection was encountered in a pigeon. Only one "infected" erythrocyte was observed in this bird, and, while the inclusion in the cytoplasm was suggestive of a malarial trophozoite, no nucleus or pigment granules could be seen.

Parasitemias in House Sparrows showing patent infections were all less than 0.01%. So few uninucleate trophozoites and/or schizonts were present that identifications of the particular species was not possible. The three latent infections revealed by isodiagnosis will be discussed in the section of this paper dealing with isodiagnostic results.

A number of species of Plasmodium are well known from birds. Hewitt (1940) lists ten species and since the writing of his book several other species have been described. Of the 11 different species of birds examined in the course of this survey, all except Riparia riparia (Bank Swallow), Regulus satrapa (Golden-crowned Kinglet), and Spinus tristis (Goldfinch) have been recorded as hosts of Plasmodium. The number of papers dealing with Plasmodium infections in House Sparrows are too numerous to list, but excellent summaries are provided by Manwell and Herman (1935a) and Jordan (1943). The latter paper also refers to the high incidence of infection in Cardinals (Richmondia cardinalis) and Brown Thrashers (Toxostoma rufum). Appendix 1 outlines percents of Plasmodium infections by host in selected surveys.

The reasons for Plasmodium not being found in most of the hosts surveyed revolves around two points. First, this malarial parasite is not one with an extended patent period. Infections may only be demonstrable with blood smear methods during the first few days after contact by a vector. If these vectors were present only during early spring, then hosts would have been latent at the time of this study. The second point is that made by Manwell and Herman (1935b). These authors theorized that malarial infections of migratory birds were acquired in the southern bases of their range because this is where the vectors can survive. If this be true, and assuming a short patent period, then it is reasonable that migratory hosts should be latent by the time they reach the northern climes.

Results of Isodiagnostic Procedures

House Sparrows (Passer domesticus) were utilized exclusively in attempts to discover latent infections by isodiagnostic means. A previous local preliminary survey had given evidence of very low incidences of patent Plasmodium infections in these birds and it was believed that the actual population incidence should be higher. Further, the number of experimental canaries available was limited.

The first canary inoculated by the intravenous route died within the first few hours of the injection, either of shock or of blood loss due to the anticoagulatory effects of too much heparin. Future injections were made using less heparin in the needles and syringes. After this alteration, no fatalities were recorded as a result of inoculating procedures.

Samples for the first isodiagnostic series were obtained from groups of five donor House Sparrows. Of the six canary hosts receiving one-quarter cc each of the pooled blood samples, three subsequently developed patent plasmodial infections. The manner of inoculation in all three cases was by the intravenous route. One of the positive experimental birds exhibited a pre-patent period of fourteen days and a patency of only two days. In this bird the parasitemia was very low, never more than 0.01%; one trophozoite and one immature schizont was observed. The duration of the pre-patent and patent periods suggests an infection of Plasmodium polare as described by Hewitt (1940). This would place the infection in the sub-genus Giovannolaia of Corradetti's

(1963) classification system.

The second positive isodiagnostic host had an incubational period of seven days and remained patent for five days. On preliminary observation, the parasite was originally identified as a Haemoproteus, but when later smears were examined definite schizont and trophozoite stages were found placing the infection in the genus Plasmodium. The parasitemia was approximated to be 0.40%. According to Hewitt's classification the infection would appear to be either P. circumflexum, P. elongatum, or P. hexamerium. Corradetti would classify this infection in the subgenera Huffia or Novyella because of the shape of the gametocyte, and of the three species only P. elongatum is included as a member of either of these two subgenera.

The third latent infection exhibited an incubational period of twenty-eight days, and a patent time of at least three days, the parasitemia being moderate to low (average 0.20%). The infection may have been either P. hexamerium, P. nucleophilum, P. polare, or possibly P. vauhani, according to the representations of Hewitt. These fall in the subgenera Novyella and Giovannolaia of Corradetti. No conclusion could be drawn as to the exact suspect species.

For the second isodiagnostic series, blood from one donor House Sparrow to one host canary was injected by way of the intraperitoneal route. Six canaries were used in this experiment. No deaths were recorded immediately after inoculation, although internal bleeding did develop in two birds, as evidenced by the

appearance of blood in the oral and nasal passages. One death did occur in this group eighteen days after injection. Tissue impressions were made of the spleen, which was not enlarged or otherwise abnormal in appearance, and the brain of the deceased host. Examination of these slides proved negative. All the blood smear slides examined during this series were negative and it is assumed that all of the six House Sparrows were free of latent infections.

The three latent infections discovered by the isodiagnostic procedures were apparently different species, provided one assumes that distinction of species can be made on the periodic differences of the course of an infection. The latter two infections suggested species that have been commonly identified in Passer domesticus, while the canary host harboring Plasmodium polare illustrates an infection that has not previously been reported in the House Sparrow. Since this would be the first recorded incidence of this species in the House Sparrow, it may be argued that the canary itself might have originally harbored this infection prior to its use in this experiment. This opposition may be removed on the basis that for all the canary hosts pre-inoculation blood smears were made and found to be negative.

The incidence of infection demonstrated in the hosts subjected to isodiagnosis in this study was 10.0%. One previous investigator (Thompson, 1943), employing similar isodiagnostic tools, showed that 31% of his birds collected in Georgia, found negative by blood smear techniques for hemal infections,

actually harbored latent plasmodial infections. Percentage results in the present study were not as high as those of Thompson's experiments, but several factors may be of importance when considering the differences between these two isodiagnoses. The present study was conducted in a more northerly latitude than Thompson's, possibly giving credence to the premise held by Manwell and Herman (1935b) that hemal infections are more prevalent in the tropical climes than in the northern realms. There has not been any positive evidence for this theory. Possible proof for this hypothesis might lie in an extensive comparative isodiagnostic study of similar species captured in each of two different climes. Such a survey might finally answer the question as to whether or not hemal infections are more common in the tropical or semi-tropical nesting sites or those of the north.

Incidence of Metazoan Blood Parasites

One Grackle and two House Sparrows examined during this survey harbored microfilarial infections. Blood smears from the Grackle showed three juvenile worms (Plates V and VI) per thirty fields (100X), six worms per thirty fields (Plates I and II) and eight worms per thirty fields (Plates III and IV), respectively, were found for each of the House Sparrows.

Measurements of several parameters were made in all three cases (Table 4), especially for comparisons of the two sparrow infections to estimate the probability of their being the same or different species. Dimensions representing distances from

the anterior to the (1) excretory pore, (2) inner body, and (3) anal pore were converted to percentage factors of the total individual length, and then reconverted to average lengths in microns (u). Probability analyses were then made of the inner body parameter and also of the total body length, utilizing the Student's T-Test and tables as described by Lacey (1964). The findings were tabulated as follows:

Student's T-Test of the Total Body Length of
the Two Microfilarial Infections

<u>Sparrow No. 69</u>			<u>Sparrow No. 50</u>		
<u>Length (microns)</u>			<u>Length (microns)</u>		
<u>n</u>	<u>x</u>	<u>x²</u>	<u>n</u>	<u>y</u>	<u>y²</u>
53.5	2.6	6.8	83.1	5.6	31.5
52.5	1.6	2.6	82.0	4.5	20.3
51.9	1.0	1.0	93.0	16.5	272.0
45.0	- 5.9	35.0	75.0	- 2.5	6.2
42.5	- 8.4	71.0	61.1	- 16.4	270.0
47.5	- 3.4	11.6	64.5	- 13.0	170.0
65.0	14.1	197.0	64.5	- 13.0	170.0
48.7	- 2.2	4.85	98.0	20.5	420.0
<hr/>		<hr/>	<hr/>		<hr/>
406.6		329.8	621.2		1360.0

mean x = 50.9 u

$$S_x = \sqrt{\frac{x^2}{n-1}} \quad S_x = 6.85$$

$$S_{\bar{x}} = \frac{S_x}{\sqrt{n}} \quad S_{\bar{x}} = 2.41$$

$$S_D = \sqrt{S_{\bar{x}}^2 + S_{\bar{y}}^2} \quad S_D = 5.45$$

$$D = \text{mean } y - \text{mean } x = 26.6$$

$$t = \frac{D}{S_D} = 4.87$$

mean y = 77.5 u

$$S_y = 13.9$$

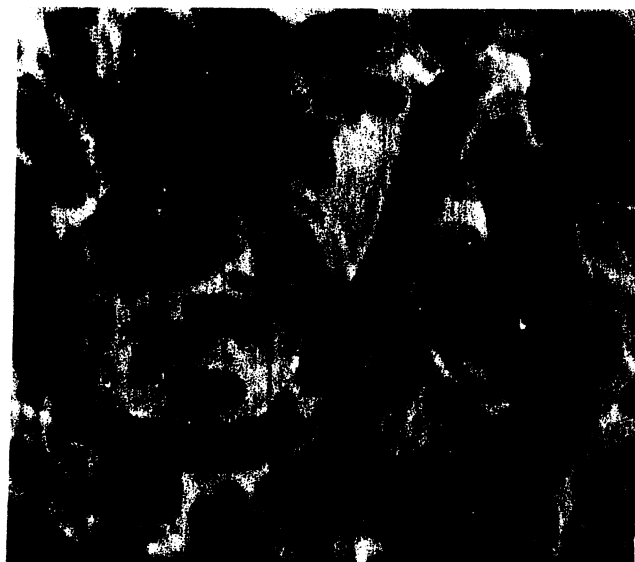
$$S_{\bar{y}} = 4.90$$

Plate I

An Unknown Microfilaria from a House Sparrow (69)
(Passer domesticus) Captured in Coles County, Illinois

Plate II

An Unknown Microfilaria from a House Sparrow (69)
(Passer domesticus) Captured in Coles County, Illinois



6.2 μ

Plate I



6.2 μ

Plate II

Plate III

An Unknown Microfilaria from a House Sparrow (50)
(Passer domesticus) Captured in Coles County, Illinois

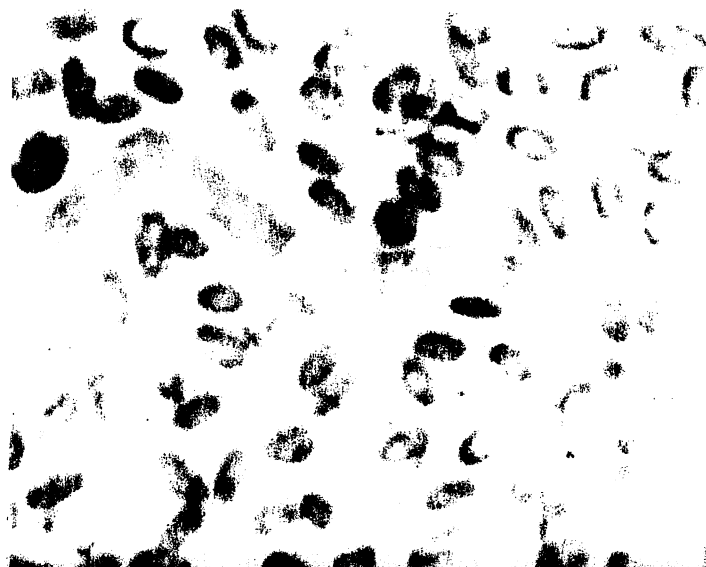
Plate IV

An Unknown Microfilaria from a House Sparrow (50)
(Passer domesticus) Captured in Coles County, Illinois



6.2 μ

Plate III



6.2 μ

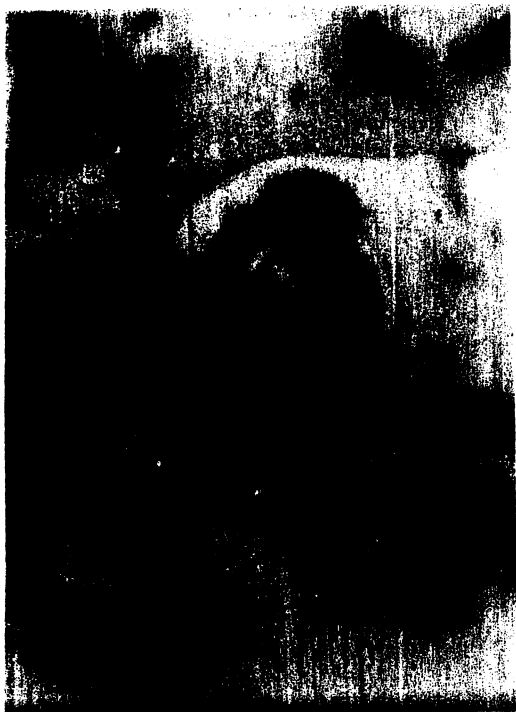
Plate IV

Plate V

An Unknown Microfilaria from a Grackle
(Quiscalus quiscula) Captured in Coles County, Illinois

Plate VI

An Unknown Microfilaria from a Grackle
(Quiscalus quiscula) Captured in Coles County, Illinois



6.2 μ

Plate V



6.2 μ

Plate VI

Table 4

DIMENSIONS OF MICROFILARIA OBSERVED IN SPARROWS AND GRACKLES

Specimen	Length μ	Anterior to Excretory Pore μ	(%) *	Anterior to Inner Body μ	(%) *	Anterior to Anal Pore μ	(%) *	Width at Inner Body μ	Posterior Space μ
<u>Sparrow No. 69</u>									
1	53.5	9.25	17.3	13.6	20.5	---	---	3.0	---
2	5.25	9.25	17.6	21.5	41.0	41.3	78.5	3.0	1.85
3	51.9	7.40	14.2	18.4	35.5	40.7	78.5	4.3	1.85
4	45.0	7.40	16.4	17.8	39.5	34.5	77.0	3.08	3.08
5	42.5	5.55	13.0	11.1	26.1	---	---	3.08	---
6	47.5	8.60	18.0	17.8	37.5	32.6	69.0	3.39	1.85
7	65.0	8.65	13.3	19.1	29.5	39.5	60.5	3.70	1.85
8	48.7	9.25	19.0	17.8	36.5	38.2	78.0	3.08	1.24
Average, μ	50.9	8.20		16.8		37.4		3.70	1.95
%			16.1		33.2		73.3		
<u>Sparrow No. 50</u>									
1	83.1	20.8	25.0	30.6	36.6	---	---	3.67	2.45
2	82.0	19.6	24.0	25.7	31.3	---	---	---	1.84
3	93.0	23.2	24.9	33.7	36.2	---	---	3.67	1.84
4	75.0	17.2	23.0	24.5	32.7	51.9	69.0	3.67	2.15
5	61.1	8.6	14.0	18.4	30.0	---	---	3.67	2.45
6	64.5	12.2	18.9	22.7	35.0	---	---	3.37	3.07
7	64.5	13.5	20.9	18.4	28.5	---	---	---	3.07
8	98.0	25.7	26.1	39.0	39.7	77.0	78.4	3.67	3.07
Average, μ	77.5	17.1		26.2		57.0		3.62	2.49
%			22.0		33.7		73.3		
<u>Grackle No. 2</u>									
1	105	---	---	38.0	36.4	---	---	3.08	---
2	93.0	19.0	20.4	39.5	42.5	64.0	68.9	3.08	2.45
3	65.0	---	---	27.7	42.5	38.0	58.2	3.67	3.07
Average, μ	87.5	19.0		35.3		55.5		3.08	2.45
%			20.4		40.3		63.2		

*% means percent of total body length.

Student's T-Test of the Anterior to Inner-Body Length
for the Two Microfilarial Infections

Sparrow No. 69			Sparrow No. 50		
Length (microns)			Length (microns)		
n	x	x ²	n	y	y ²
17.3	1.2	1.44	25.0	3.0	9.0
17.6	1.5	2.25	24.0	2.0	4.0
14.2	-1.9	3.61	24.0	2.9	8.41
16.4	0.3	0.09	23.0	1.0	1.0
13.0	-3.1	9.61	14.0	-8.0	64.0
18.0	1.9	3.61	18.9	-3.1	9.61
13.3	-2.8	7.85	20.9	-1.1	1.21
19.0	2.9	8.41	26.1	4.1	16.9
<hr/>			<hr/>		
128.8		36.87	176.8		114.1

mean x = 16.1

mean y = 22.0

Sx = 2.29

Sy = 4.03

Sx̄ = 0.81

Sȳ = 1.41

$$S_D = 1.63$$

$$D = 5.9$$

$$t = 3.62$$

Saunders (1955) stated that human microfilarial species may be distinguished on the basis of differences in bodily measurements. Thus, we may conclude that statistically the two infected House Sparrows were each found to harbor a different microfilarial infection, the t values having proved significant at least to the 0.05 level.

Odetoyinbo (1961) described two different microfilariae in the Grackles he examined. One type was given the generic name Microfilaria "X" because its adult form was not located

in the host. The embryo was characterized as being a sheathed organism with a blunt anterior and pointed posterior end. The other microfilarial specimen was identified as the larval form of the adult filaria, Splendidofilaria quiscali, and thus given the title Microfilaria quiscali. This immature form was distinguished by being sheathed with both extremities appearing round and blunt. The average length of this species was 187 μ , ranging from 154-208 μ . Other measurements (percentage distance of the total body length from the anterior end in parenthesis) were:

- (a) Width: 3-6 μ , average 5.2 μ
- (b) Anterior to excretory pore: 37-86 μ ,
average 59 μ , (31.5%)
- (c) Anterior to inner body: 68-139 μ ,
average 101.2 μ , (54.0%)
- (d) Anterior to anal pore: 86-177 μ ,
average 137.7 μ , (73.1%)

When these figures are compared to those of the Grackle specimens in Table 4, differences are clearly established in all measurements. The most evident morphological distinction is that our species was anteriorly pointed, while the posterior was blunt. Apparently, the specimens obtained from the Grackle in this survey are a new and as yet undescribed species.

GENERAL DISCUSSION

The question of whether malarial and other hemal parasitic infections are incurred in the southern climes or the northern climes has been debated since the earliest surveys were conducted. Manwell and Herman (1935b) found that avian malarial cases have been found almost worldwide, except at the poles and various sequestered localities. They theorized that the annual avian migrations were necessary for the dissemination of the microfilarial and malarial infections because of the warm temperature and humidity requirements of the intermediate hosts, which are lacking at northern migrational points. As part of their survey they noted that of the robins (Turdus migratorus) they examined, all the adults were positive for the malarial parasites while all the juveniles were negative, leading them to conclude that the infection was incurred while at the southern migratory bases.

The antithesis to Herman and Manwell's theory was that as proposed by Micks (1949). He believed that infection might be acquired in any clime. In the northern regions this would naturally depend on the season. Compiling twenty-four surveys from thirteen states that included 9,577 birds, he calculated that 7% of the specimens were positive. One of the findings that fostered his theory was that the House Sparrows, which are a non-migratory species, were tabulated to have a higher malarial incidence than the average. Mohammed (1953) raised the objection that Micks failed to take into consideration the variances in susceptibility of

different hosts.

To defend Micks it must be said that many investigators have agreed with his theory. Working with the introduction of microfilarial infections of the Common Crow, (Corvus brachyrhynchos), and the Blue Jay, (Cyanocitta cristata), Robinson (1955) was forced to conclude that the infection was acquired in the north because these birds were not migratory, even though the specific vector was not found. As for the Leucocytozoan infection, Fallis, et al. (1951) found that the incidence of infection in the migratory ducks was directly related to the presence of two simuliid Black Fly species in Canada. Huff (1932), during his studies of Haemoproteus in Mourning Doves (Zenaidura macroura), was not able to find hippoboscids on the infected doves. He thus concluded that trasference of the infection was limited exclusively to the southern part of the migrational routes. A rather informative survey was recently reported by Huff and Whitmore (1967) from specimens collected in Panama. They compared their results with those of Huff's (1939) extensive survey. With an infection incidence of 44.4% in the U.S., the corresponding figure was only 15.2% in Panama. Breaking this down into types of infection:

	Panama	United States
<u>Haemoproteus</u>	39%	80%
<u>Plasmodium</u>	17	15
<u>Leucocytozoon</u>	3.5	2.7
<u>Trypanosoma</u>	10.7	2.7
<u>Microfilaria</u>	28.5	(not recorded)

Apparently, latitude is no true indication of the certainty of discovering infection of avian hosts.

If one concludes that Micks' theory is the most credible, the problem of recrudescence of infection in northern realms must then be discussed. Several investigators have noticed the heightened incidence of the several hemal infections during the warmer months when the suspected or certified vectors are present. Jordan (1943) cited a definite increase in the incidence of Plasmodium and Haemoproteus infections from the beginning of June through the rest of the summer months; this he theorized as being apparently due to the susceptibility of the juveniles. Micks' (1949) survey illustrated with his captured sparrows this seasonal rise very explicitly:

<u>Month</u>	<u>No. Examined</u>	<u>No. Positive</u>	<u>% Positive</u>
January	33	0	0
February	33	0	0
March	57	2	3.5
April	42	7	16.6
May	23	5	21.7
June	42	5	12.0
July	19	4	21.0
August	30	3	10.0
September	150	24	15.0
October	35	5	11.0
November	16	1	6.2
December	<u>26</u>	<u>2</u>	<u>7.7</u>
	516	58	11.2

It would seem apparent that the infections remain latent during winter months and parasitemia ensues almost simultaneously with the appearance of the vectors, the transferring and perpetuating of infection being conducted with relative ease. The problem then arises as to what biological

factors are functioning to initiate the recurrence of the parasites in the peripheral blood, specifically as Bishop (1955) mentioned Huff's noting a higher gametic parasitemia during the reproductive months. One possible reason for this may be a change in the endocrinological factors that occur during the spring months. With the increase of androgens and estrogens which trigger the host reproductive processes, perhaps there is a corresponding triggering of the parasites reproductive capacities and resultant reappearing patency.

Proceeding from accepted parasitological evolutionary theory, whereby organisms tend to develop from the generalized to the specialized, it is possible to envision the means by which the hemal parasites have evolved. Initially, Manwell and Herman's theory was probably exclusively functioning for dispersion of these parasites, but as the avian hosts dispersed so were their parasites. These malarias adapted to more specialized environments, which were composed of the various hosts, vectors, and seasonal reproductive and viable periods of the climes. At the present time, many of these infections have progressed to the stage that is encompassed by Micks' theory. This is to say that in these organisms genetic adaptation has evolved to the degree that the need for continual trasference of the infection is not necessary, but only annually. Perhaps infection may now occur at either pole of the migratory routes, and that sub-species have evolved to such an extent that one specific parasitic organism may be at one extreme and a different species at the other and various hybrids in between.

SUMMARY AND CONCLUSIONS

1. A survey of the blood parasites in the local avian fauna of Coles County, Illinois, was conducted during the summer and fall of 1967. Birds were captured from various sites including rural and urban areas.
2. One hundred and twenty-three birds were examined. These included: Passer domesticus (69), Columba livia (36), Melospiza melodia (4), Hirundo rustico (3), Riparia riparia (3), Quiscalus quiscula (3), Bubo virginianus (1), Colinus virginianus (1), Regulus satrapa (1), Richmondia cardinalis (1), Toxostoma rufum (1).
3. Of the five trapping procedures utilized for collecting specimens for this survey, three were very successful, one moderately so, and one not successful at all.
 - a. Hand netting was most productive in terms of numbers of captured avian specimens, while mist netting gave the greatest variety of birds.
 - b. Removal of birds from their roosting sites was most rewarding when considering the energy expended.
 - c. The Hav-a-hart repeating balance trap proved to be very useful for the capture of House Sparrows (Passer domesticus).
 - d. Funnel traps were not as productive as anticipated, the only captured specimen was a Brown Thrasher (Toxostoma rufum).
 - e. Night-lighting was ineffective.

4. Twenty (16.2%) of the 123 birds examined were found to harbor hemal parasitic infections. Of the 20 cases, 17 were patent and were discovered using the usual blood smear techniques, while three latent infections were revealed using isodiagnostic procedures.
 - a. Haemosporidian parasites of the genera Haemoproteus, Plasmodium, and Leucocytozoon were found. The relative incidence of each type of infection was 9.7%, 4.85%, 0.8%, respectively.
 - b. One of the three latent infections uncovered by the isodiagnostic procedures suggested the presence of Plasmodium polare in one of the donor House Sparrows. If this interpretation is correct, the finding would be a new host record for this avian malarial species.
 - c. Two injection routes were utilized for the isodiagnostic tests. The three positive cases were found in hosts inoculated by way of the intravenous method, while all hosts injected by the intraperitoneal route were negative.
 - d. One Grackle (Quiscalus quiscula) and two House Sparrows examined during this survey harbored microfilarial infections, each host apparently infected with a different species.
5. There are essentially two phases in the study of avian hemal protozoan and microfilarial infections that are in need of continued research. Primarily, studies should be conducted in areas of the world in which sufficient surveys

have not been made. It is suggested that these researches include not only immediate blood smear examinations, but also one-to-one isodiagnostic techniques.

Secondly, much work is needed in correlating the results of the many surveys already reported. Lacking is a collective map illustrating the prevalence of infections, especially the different plasmodial species, in order to allow for the possible uncovering of some previously unknown physiological, ecological, or geographical relationship that might exist between the hosts, parasites, and vectors.

Literature Cited

1. Bennett, G. F., Garnham, P. C., and Fallis, A. M., "On the Status of the Genera Leucocytozoon Ziemann, 1898 and Haemoproteus Kruse, 1890 (Haemosporidiida: Leucocytozoidae and Haemoproteidae)", Can. Jour. of Zool., 43:927-932, 1965.
2. Bishop, A., "Problems Concerned with Gametogenesis in Haemosporidia, with Particular Reference to the Genus Plasmodium," Parasitology, 45:163-185, 1955.
3. Boughton, D. C., "Microfilarial Periodicity in the Crow," Journ. of Paras., 24:161-165, 1938.
4. Boyd, M. F., Malariology: Vol. I, W. B. Saunders Co., Philadelphia, 1949.
5. Cameron, T.W.M., Parasites and Parasitism, John Wiley and Sons, Inc., New York, 1956.
6. Chandler, A. C., Introduction to Parasitology, John Wiley and Sons, Inc., New York, 1961.
7. Cheng, T. C., The Biology of Animal Parasites, W. B. Saunders Co., Philadelphia, 1964.
8. Coatney, G. R., "A Check-List and Host-Index of the Genus Haemoproteus," Journ. of Paras., 22:336-340, 1938.
9. _____, "A Catalog and Host-Index of the Genus Leucocytozoon," Journ. of Paras., 23:202-212, 1937.
10. _____, "Some Blood Parasites from Birds of the Lake Okajobi Region," Amer. Mid. Nat., 20:336-340, 1938.
11. Coatney, G. R. and Jellison, D., "Some Blood Parasites from Montana Birds," Journ. of Paras., 26:158-160, 1940.
12. Coatney, G. R. and Roudabush, R. L., Haemoproteus beckeri N. Sp. and Trypanosoma laverani var. toxostomae N. Var. from the Brown Thrasher (Toxostomae rufum), Iowa State College Journ. of Science, 10:1-6, 1935.
13. _____, "Some Blood Parasites from Nebraska Birds," Amer. Mid. Nat., 18:1005-1030, 1937.
14. Coatney, G. R. and West, E., "Some Blood Parasites from Nebraska Birds II," Amer. Mid. Nat., 19:601-612, 1938.
15. Corradetti, A., P.C.C. Garnham and M. Laird, "New Classification of the Avian Malaria Parasites," Parasitology, 5:1-4, 1963.

16. Fallis, A. M., et al., "Life History of Leucocytozoon simondi Mathis and Leger in Natural and Experimental Infections and Blood Changes Produced in the Avian Host," Can. Journ. of Zool., 29:305-328, 1951.
17. Farmer, J. N., "Some Blood Parasites from Birds in Central Iowa," Iowa Acad. of Sci., 67:591-597, 1960.
18. Faust, E. C., et al., Animal Agents and Vectors of Human Disease, Lea and Febiger, Philadelphia, 1962.
19. Garnham, P.C.C., Malarial Parasites and Other Haemosporidia, Blackwell Scientific Publications, Oxford, 1966.
20. Ginrich, W., "Methods of Estimating Haemosporidial Infections," Journ. of Prev. Med., 6:197-246, 1932.
21. Hawking, F., "Microfilaria Infestation as an Instance of Periodic Phenomena seen in Host-Parasitic Relationship," Annals of the New York Acad. of Sci., 98:940-953, 1962.
22. Hegner, R. and F. Wolfson, "Association of Plasmodia and Toxoplasma-Like Parasites in Birds," Annals of the Journ. of Hyg., 28:437-454, 1938.
23. Herman, C. M., "Toxoplasma in North American Birds and Attempted Transmission to Canaries and Chickens," Amer. Journ. of Hyg., 25:303-312, 1937.
24. _____, "The Relative Incidence of Blood Protozoa in Some Birds from Cape Cod," Trans. of the Amer. Micro. Soc., 57:132-141, 1938.
25. _____, "The Blood Protozoa of North American Birds," Bird Banding, 15:89-112, 1944.
26. Hermis, W. B., et al., "Blood Parasites of California Birds," Journ. of Paras., 25:510-512, 1939.
27. Hewitt, T., Bird Malaria, John Hopkins Press, Baltimore, 1940.
28. Huff, C. G., "Studies on Haemoproteus of Mourning Doves," Amer. Journ. of Hyg., 16:618-623, 1932.
29. _____, "A Survey of the Blood Parasites of Birds Caught for Banding Purposes," Journ. of the Amer. Vet. Med. Assoc., 94:615-620, 1939.
30. _____, "Experimental Research on Avian Malaria," Naval Medical Research Institute, Bethesda, Maryland, 1963.

31. Huff, C. G. and A. Whitmore, "Blood Parasites of Birds Collected in Four Successive Years in Panama," Bull. Wildlife Disease Assoc., 3:179, 1967.
32. Hyman, L., The Invertebrates: Vol. II, McGraw-Hill, New York, 1951.
33. Jordan, H. B., "Blood Protozoa of Birds Trapped in Athens, Georgia," Journ. of Paras., 29:260-293, 1943.
34. Kudo, R. R., Protozoology, Thomas Pub. Co., Springfield, Ill., 1966.
35. Lacey, O. L., Statistical Methods in Experimentation, MacMillan, New York, 1964.
36. Levine, N. D., Protozoan Parasites of Domestic Animals and Man, Burgess Pub. Co., Minneapolis, Minnesota, 1961.
37. MacCallum, W. G., "Notes on the Pathological Changes in the Organs of Birds Infected with Haemacytozoa," Journ. of Exp. Med., 3:103-116, 1898.
38. McIlhenny, E. A., "Results of 1940 Bird Banding at Avery Island, Louisiana, with Special Account of a New Banding Method," Bird Banding, 13:19-28, 1942.
39. Manwell, R. D., "How Many Species of Avian Malarial Parasites are There?" Amer. Journ. of Trop. Med., 15:268-283, 1935.
40. Manwell, R. D. and C. Herman, "Duration of Malarial Infections in Birds," Amer. Journ. of Hyg., 19:532-538, 1934.
41. _____, "The Occurrence of the Avian Malarias in Nature," Amer. Journ. of Trop. Med., 15:661-673, 1935a.
42. _____, "Blood Parasites of Birds and Their Relation to Migratory and Other Habits of the Host," Bird Banding, 6:130-133, 1935b.
43. Micks, D. W., "Malaria in the English Sparrows," Journ. of Paras., 35:543-544, 1949.
44. Mohammed, H., Systematic and Experimental Studies on Protozoal Blood Parasites of Egyptian Birds, Cairo Univ. Press, 1958.
45. Novy, F. G. and W. J. MacNeal, "Trypanosomes and Bird Malaria," Proc. of the Soc. of Exp. Biol. Med., 2:23-28, 1905.

46. Odetoyinbo, J. A., "Biology of Splendidofialria guiscali (Von Linstow, 1904) N. Comb. (Nematoda: Onchocercidae)," Iowa State Univ. Ph.D. Thesis, 1960.
47. Opie, E. L., "On the Haemocytozoa of Birds," Journ. of Exp. Med., 3:76-101, 1898.
48. Quay, T. L., Mourning Dove Studies in North Carolina, Pamphlet of N.C. Wildlife Res. Comm., Raleigh, N.C., 1951.
49. "Report of the Committee on Terminology of Strains of Avian Malaria," Journ. of Paras., 28:250-254, 1942.
50. Ridgeway, B. T., "Host-Parasite Relationships of a Strain of Plasmodium relictum," Univ. of Mo., Ph.D. Thesis, 1966.
51. Robinson, E. J., "Observations on the Epizootology of Filarial Infections in Two Species of the Avian Family Corvidae," Journ. of Pars., 41:209-214, 1955.
52. Saunders, D. C., "The Classification of Microfilariae in Birds, Avifilaris tryannidarum and A. fringillidarum," Trans. Amer. Micro. Soc., 74:37-45, 1955.
53. _____, "Microfilariae and Other Blood Parasites in Mexican Wild Doves and Pigeons," 45:69-74, 1959.
54. Sergent, E., "LeDiagnostic de L'Infection Latente dans le Paludisme des Oiseaux (Plasmodium relictum)," Comptes Rendus des Seances et Memoires de la Societe de Biologie, 83:1063-1064, 1920.
55. Schneider, M., Illinois Institute of Technology, Research Institute, Chicago, Illinois, 1967.
56. Skrjabin, K. I., Key to Parasitic Nematodes, Sivan Press, Jerusalem, 1961.
57. Smith, D. G., "A Survey of the Occurrence of Blood Parasites in the Local Bird Population in the Horam, Ohio Area," Bull. Wildlife Dis. Assoc., 3:185-6, 1967.
58. Thompson, R. E., "Relative Incidence of Blood Parasites in Some Birds from Georgia," J. of Parasites, 29:153-5, 1943.
59. Wetmore, B. W., "Blood Parasites of Birds of the District of Columbia and Patuxent Research Refuge Vicinity," J. of Paras., 27:379-393, 1941.

60. Whitmore, E. R., "Observations on Bird Malaria and the Pathogenesis of Relapse in Human Malaria," *John Hopkins Hospital Bull.*, 29:62-67, 1918.
61. Wood, S. F. and C. M. Herman, "The Occurrence of Blood Parasites in Birds from Southwestern U.S.," *J. of Paras.*, 29:187-196, 1943.
62. Wood, F. D. and S. F. Wood, "Occurrence of Haematozoa in Some California Birds and Mammals," *J. of Paras.*, 23: 197-202, 1937.

APPENDIX I

INCIDENCE OF PROTOZOAN PARASITES OBSERVED IN SELECTED AVIAN HOSTS IN SURVEYS 1935 TO 1967

Host	Exam.	Plasmodium		Haemoproteus		Leucocytozoan		Trypanosoma		Observer
		+	%	+	%	+	%	+	%	
<u>Hirundo</u> <u>erythro-</u> <u>gaster</u>	31	0	0.0	0	0.0	0	0.0	0	0.0	Coatney and West (1938), Coatney and Roudabush (1937), Manwell and Herman (1935), Herman (1938), Hermis (1937)
<u>Riparia</u> <u>riparia</u>	19	0	0.0	0	0.0	0	0.0	0	0.0	Manwell and Herman (1935)
<u>Columba</u> <u>livia</u>	1	0	0.0	0	0.0	0	0.0	0	0.0	Jordan (1943)
	31	0	0.0	11	35.5	0	0.0	0	0.0	Saunders (1959)
	2	0	0.0	1	50.0	1	50.0	0	0.0	Wood and Herman (1943)
	106	0	0.0	78	73.5	0	0.0	0	0.0	Mohammed (1958)
	451	1	0.22	50	11.0	0	0.0	0	0.0	Coatney and Roudabush (1937)
Total	591	1	0.17	140	23.6	1	0.17	0	0.0	
<u>Colinus</u> <u>virginianus</u>	93	1	1.1	1	1.1	0	0.0	0	0.0	Wetmore (1941)
<u>Spinus</u> <u>tristis</u>	4	0	0.0	2	50.0	0	0.0	0	0.0	Huff (1939)
	33	0	0.0	0	0.0	1	3.0	4	12.1	Wood and Herman (1943)
Total	37	0	0.0	2	5.4	1	2.7	4	10.8	

APPENDIX I (cont'd)

<u>Richmondia</u>										
<u>cardinalis</u>	3	0	0.0	2	66.7	0	0.0	0	0.0	Smith (1967)
	7	3	43.0	2	28.6	1	14.3	2	28.6	Thompson (1943)
	8	3	37.5	1	12.5	4	50.0	0	0.0	Wetmore (1941)
	19	0	0.0	8	43.0	0	0.0	1	5.25	Huff (1939)
	25	18	72.0	7	28.0	0	0.0	2	8.0	Jordan (1943)
Total	62	24	38.8	20	32.2	5	8.07	5	8.07	
<u>Passer</u>	77	0	0.0	0	0.0	0	0.0	0	0.0	Hermis (1939), Wetmore
<u>domesticus</u>										(1941), Wood and Wood
<u>domesticus</u>										(1937), Coatney and
										Roudabush (1937), Smith
										(1967), Herman (1938)
	245	7	2.9	0	-	-	-	-	-	Manwell and Herman (1935)
	41	3	7.3	0	0.0	0	0.0	0	0.0	Thompson (1943)
	489	71	19.5	63	29.4	0	0.0	0	0.0	Jordan (1943)
	125	13	10.4	0	0.0	0	0.0	2	1.6	Huff (1939)
	5	1	20.0	0	0.0	0	0.0	0	0.0	Coatney and West (1938)
(P.d.nilo-	176	72	40.9	70	39.7	0	0.0	5	2.8	Mohammed (1958)
<u>ticus)</u>	41	2	4.9	0	0.0	0	0.0	0	0.0	Wood and Herman (1943)
Total	1,249	169	13.5	133	13.2	0	0.0	7	0.69	
<u>Toxostoma</u>	48	0	0.0	29	60.5	0	0.0	2	4.3	Huff (1939)
<u>rufum</u>	12	2	17.0	5	41.5	0	0.0	0	0.0	Wetmore (1941)
	2	0	0.0	0	0.0	0	0.0	1	50.0	Coatney and West (1938)
	9	3	33.3	0	0.0	0	0.0	1	11.1	Thompson (1943)
	215	41	19.0	63	29.4	0	0.0	0	0.0	Coatney (1938)

APPENDIX I (cont'd)

<u>(T.r. red- ivurium)</u>	2	0	0.0	0	0.0	0	0.0	0	0.0	Wood and Wood (1938)
	14	0	0.0	0	0.0	1	7.1	0	0.0	Wood and Herman (1943)
Total	303	46	15.2	98	32.3	1	0.33	4	1.3	
<u>Bubo virginianus</u>	3	0	0.0	0	0.0	0	0.0	0	0.0	Herman (1938), Coatney and Jellison (1940), Manwell and Herman (1935)
	1	1	100.0	1	100.0	0	0.0	0	0.0	Wetmore (1941)
	1	1	100.0	1	100.0	1	100.0	1	100.0	Coatney and Roudabush (1937)
	3	0	0.0	1	33.3	0	0.0	0	0.0	Coatney (1938)
	25	0	0.0	5	20.0	3	12.0	0	0.0	Farmer (1960)
	33	2	6.0	8	24.2	4	12.5	1	3.1	
<u>Quiscalus quiscula</u>	2	0	0.0	0	0.0	0	0.0	0	0.0	Coatney (1938), Coatney and Roudabush (1937)
	3	2	67.7	1	33.3	0	0.0	0	0.0	Jordan (1943)
	16	1	6.2	1	6.2	5	31.2	1	6.2	Farmer (1960)
	19	1	5.3	-	-	-	-	-	-	Manwell and Herman (1935)
	26	2	7.7	13	50.0	16	61.5	0	0.0	Smith (1967)
	89	14	15.7	21	23.5	41	46.0	2	2.2	Wetmore (1941)
	130	15	11.5	45	34.5	4	3.1	1	0.8	Huff (1939)
	Total	285	35	12.3	81	31.0	66	24.8	4	1.5
<u>Melospiza melodia</u>	6	0	0.0	0	0.0	0	0.0	0	0.0	Smith (1967)
	20	3	15.0	2	10.0	0	0.0	0	0.0	Huff (1939)
	41	1	2.4	0	0.0	0	0.0	0	0.0	Wood and Herman (1943)
	62	22	35.5	-	-	-	-	-	-	Manwell and Herman (1935)
	204	9	4.4	1	0.49	0	0.0	0	0.0	Herman (1938)
Total	333	35	10.5	3	9.0	0	0.0	0	0.0	