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Black flies (Diptera: Simuliidae) occurring in Mississippi, and their medical, veterinary, and economic impacts

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By

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Little is known about black fly pest species in Mississippi, other than research from the 1930s. A better understanding of the pest species that occur in Mississippi is important for human and animal health. My research focused on what species of black flies occur in Mississippi, their seasonality and distribution, and a detailed systematic survey of the primary pest species. Lastly, I attempted to quantify nuisance effects and economic impacts of black flies on people, backyard poultry, and livestock.

I examined scientific literature and records of adult black flies occurring in the southeastern U.S., and particularly Mississippi. This search revealed several unpublished manuscripts by Dr. George H. Bradley on the biology, ecology, and control of black flies in the Mississippi Delta during the 1930s. These publications were curated and made available to the scientific community.

I identified and compiled an annotated list of larval, pupal, and adult stages of black flies occurring in Mississippi, derived from specimens housed in the Mississippi State University Entomological Museum (MEM). These specimens had been collected over several decades by a

variety of entomologists, students, and the public. In addition, I included data from thousands of black flies collected during this current project.

To assess seasonality and relative abundance of the primary pest black fly species in Mississippi, I systematically collected adult black fly specimens for two years, documenting species present, seasonality, adult emergence patterns, and associated meteorological conditions. These ten sites were selected based on Dr. George Bradley's extensive work and complaints from local county extension agents, veterinarians, and municipal public works personnel.

For economic, human, and animal health impacts of black flies, I employed a four-tiered approach: 1) a survey of lay and medical literature for reports of human health problems from black fly bites, 2) a query of city and county public works personnel concerning black fly nuisance effects, 3) an analysis of statewide hospital outpatient International Classification of Diseases-9 (ICD-9) discharge data and lastly, 4) a statewide survey of backyard poultry owners to ascertain animal health and monetary impacts from black flies.

DEDICATION

This dissertation is dedicated to my daughters, Josephine Isabel and Peyton Elizabeth, my driving force and inspiration through this entire process. You are my earthly angels that have kept me flying. Remember to always keep your dreams in focus and reach for the stars. I am also grateful to my family and friends, for without your love and support, I would have quit a very long time ago. This has been a hard road to travel, and your energy has kept me moving.

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At Mississippi College, I embarked on a journey. I had the wonderful experience of working under Dr. Bill Stark. He introduced me to aquatic entomology, and I loved it. If it had not been for his kind and helpful guidance, I would have never found entomology.

Lastly, I would like to thank my committee members. Your quiet suggestions, comments, and powerful words have been a contributing factor to this degree. I hope to make all of you proud.

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CHAPTER I

BACKGROUND AND LITERATURE REVIEW

Background and History

Black flies, members of the insect family Simuliidae, are significant pests throughout the world (WHO 1989) that have tremendous medical and veterinary importance; about ten percent of species attack humans (Mock and Adler 2002, Adler and McCreadie 2009). The species that do not bite may still be worrisome pests by swarming around the head and landing on the ears, face, and neck, a behavior sometimes called "dive-bombing" (Weed 1904, Knowlton 1935, Davies and Peterson 1956, Fallis 1964). While on other animals, they attack sites with little or no pelage, like the teats, udders, and underbelly (Webster 1887, Cameron 1922, Millar and Rempel 1944, Jones 1961, WHO 1971, Jones et al. 1977). Species of black flies that bite birds on the head and neck may attack in great numbers and cause exsanguination or suffocation (Webster 1887, Bradley 1933, Noblet et al.1975, Rohner et al. 2000, Jones et al. 2014).

Black flies are such vicious pests that they have been referred to with strong language such as "blood-sucking demons" (Gudgel and Grauer 1954, Adler and McCreadie 1997). Females require a blood meal for nourishment of their eggs; and this blood-feeding behavior has caused enormous economic losses due to human and animal stress and avoidance behaviors (Getting 1945, Davies and Peterson 1956, Fallis 1964, Adler and McCreadie 1997).

In North America, black flies are one of the few arthropod groups that have killed animals during massive and severe attacks by exsanguination, the process of blood loss to a

degree sufficient to cause death (Webster 1887, Millar and Rempel 1944). The first known reported documentation of black fly attacks on animals was in 1859 in Greenville, Mississippi and Clarendon, Arkansas, with these gnats harassing and killing livestock (Webster 1904, Atwood and Meisch 2004). There was additional documentation of an abundance of black flies in May 1888, with a severe outbreak in spring of 1891, attacking livestock in the White Rock Mountains near Vineland, Arkansas (Webster 1904, Atwood and Meisch 2004). The pest black fly species, *Simulium meridionale*, was documented frequently from 1886 – 1891 from Somerset Landing to Madison, Arkansas (Atwood and Meisch 2004).

Most of the important black fly pest species belong to the genus *Simulium*. In North America, *Simulium vittatum* is annoying to livestock. This species crawls and constantly probes the skin of livestock (Webster 1887). The turkey gnat, *Simulium meridionale*, is a common species during the spring, occurring throughout the Mississippi Valley and southeastern United States. This species attacks the wattles and combs of poultry. Another common species that harasses people is *Simulium venustum* (Peterson 1955). This species often attacks fisherman and campers. They are found in mass numbers during June and July but can continue through the summer season in the United States, Canada, and Alaska (Peterson 1955).

Subsequently from 1861 – 1864, the buffalo gnat, *Cnephia pecuarum*, became a severe nuisance, killing every horse and mule in its path. In 1866, the alluvial countryside in Louisiana was literally taken over by this pest. Within a few days in Tensas, Madison, and Concordia Parishes in Louisiana, over 4,000 mules and horses were killed by black flies (Webster 1904). Some planters would lose up to 25 to 75 head a day on their plantations (Webster 1904). There were additional massive outbreaks from 1881 – 1884. In 1882, black flies were so bad that wild deer were pushed out of swamps and almost exterminated in Louisiana between Ouachita and

Mississippi Rivers. It was during this time that physicians verified the death of several people, in Louisiana, and Arkansas, who had been attacked by gnats (Atwood and Meisch 2004). Hundreds of horses and mules were killed while working on plantations, before they could be taken to an area of protection from these biting insects. In addition, cities like Memphis, Tennessee, and Vicksburg, Mississippi had horses and mules attacked and killed on streets and in stables. The attacks on livestock, during the spring in March and April when planters needed to plant their springtime crops, were so severe it was almost impossible to run a horse and cart during that time (Webster 1904, Atwood and Meisch 2004). A black fly attack in 1874, in a west Tennessee county, killed an estimated $500,000 worth of livestock (Webster 1887, 1904).

At one point, most of the country bordering the Mississippi River between Cairo, Illinois and New Orleans, Louisiana, suffered economic losses due to black flies. These pests made raising livestock in these areas unprofitable and almost impossible (Webster 1904, Atwood and Meisch 2004). Further documentation in these areas from 1944 – 1948 from black fly outbreaks show more than 1,100 animals died annually from the immense number of flies that attacked livestock (Webster 1904, Atwood and Meisch 2004).

Figure 1.1 Black fly on a person's body

Black flies are small insects that have caused detrimental effects to humans, livestock, and backyard poultry owners. Even though they are small, black flies have large-scale economic impacts on farmers and backyard poultry owners. Estimated adult size, 1 – 5 mm. Photo credit: Centers for Disease Control (CDC) website.

Dr. George H. Bradley – Historical Outbreaks in Mississippi

During the 1930s, a USDA scientist named George H. Bradley studied black flies extensively in the southern U.S., particularly, in Mississippi. He reported outbreaks of the buffalo gnat, *Cnephia pecuarum*, in Mississippi and Arkansas and conducted fieldwork on seasonality, location of breeding sites, duration and modes of attacks, conditions of river habitats, treatments, and control methods used during outbreaks (Gray et al. 1996). He found when floodwaters were consistent during winter season, outbreaks of *Cnephia pecuarum* were mild. When the winter season was dry and floodwater came during the spring, *Cnephia pecuarum* would erupt in large masses. This caused serious livestock losses, especially during 1931 and 1934, with isolated cases occurring in 1932 and 1933 (Bradley 1935b, Atwood and Meisch 2004). In 1931, almost

1,000 mules were killed in Arkansas and Mississippi, and in 1934 over 500 mules were killed in Arkansas (Bradley 1934, 1935a, b, WHO 1971, Mock and Adler 2002). Since the 1930s, black fly problems have subsided in the southern states; however, there have been reports of black fly problems since 2009 (Zema 2011, Jones et al. 2014).

Figure 1.2 Farmer using smoke plumes to protect mules from black flies

Dr. George Bradley documented protection measures that farmers used during planting season to protect their livestock from black flies. Photo credit: Dr. George Bradley (1932), USDA.

Feeding Behavior and Bite Reactions

Female black flies are daytime feeders (Balfour 1906, Bradley 1937, Davies and Peterson 1956, Adler and McCreadie 2009), and most species do not enter homes or buildings looking for hosts (Stone and Snoddy 1969, Jones et al. 2014). Humans may be attacked viciously, causing localized reaction with reddening, itchy wheals, and other conditions that vary due to sensitivity of the person and number of bites (Gudgel and Grauer 1954, WHO 1971, Zema 2011, Chen 2013). “Black fly fever,” a syndrome resulting from bites, is characterized by headache, fever, nausea, dermatitis, and allergic reactions. Some people have severe reactions including swollen

arms, face, and other exposed body parts, often on the lower legs (Balfour 1906, Gudgel and Grauer 1954, Gallis 1964, DeFoliart and Rao 1965, Choudhary and Choudhary 2005). Black flies are especially troublesome in recreational areas. Even a small population is considered annoying and large populations can become simply intolerable. Annoyance from black flies is usually seasonal, occurring in spring to early summer. When black flies are in season, vacationing and tourist activity can be greatly reduced (Weed 1904, Newson 1977, Freeden 1985, Gray et al. 1996, Zema 2011, Anonymous 2013).

Diseases Associated with Black Flies

The main disease threat worldwide from black flies is onchocerciasis, caused by the nematode *Onchocerca volvulus* (De Kock 1928, Getting 1945, Collins 1979, WHO 1995). Onchocerciasis occurs in 34 countries: 27 in Africa, 6 in Latin America, and one in the Arabian Peninsula (WHO 1995). There are possibly 90 million people at risk, and at least 37 million people are infected with onchocerciasis (WHO 1995). The number of people who have developed blindness from onchocerciasis is approximately 270,000, with possibly another 500,000 with severe ocular disability (WHO 1989, 1995). Specific vectors of this disease belong primarily in the *Simulium damnosum* sensu lato complex (Crosskey 1996). This complex includes groups separated into three clades by habitat and mitochondrial DNA sequencing: 1) savanna clade, *Simulium damnosum* sensu stricto and *Simulium sirbanum*; 2) rain forest clade, *Simulium yahense* and *Simulium squamosum*; 3) a transition clade, *Simulium leonense* and *Simulium sanctipauli* (Peterson et al. 1981, WHO 1989, 1995, Richards et al. 2001).

Black flies in the areas listed above, especially in Africa, can travel long distances due to the wind currents. Trade winds allow black flies to make their way from one area to another. In fact, over a period of 3 – 4 weeks, black flies can travel 150 km (De Kock 1928). This is one

reason why onchocerciasis is so endemic in Africa (De Kock 1928) as opposed to other tropical locations (De Kock 1928, Fallis 1964, WHO 1989).

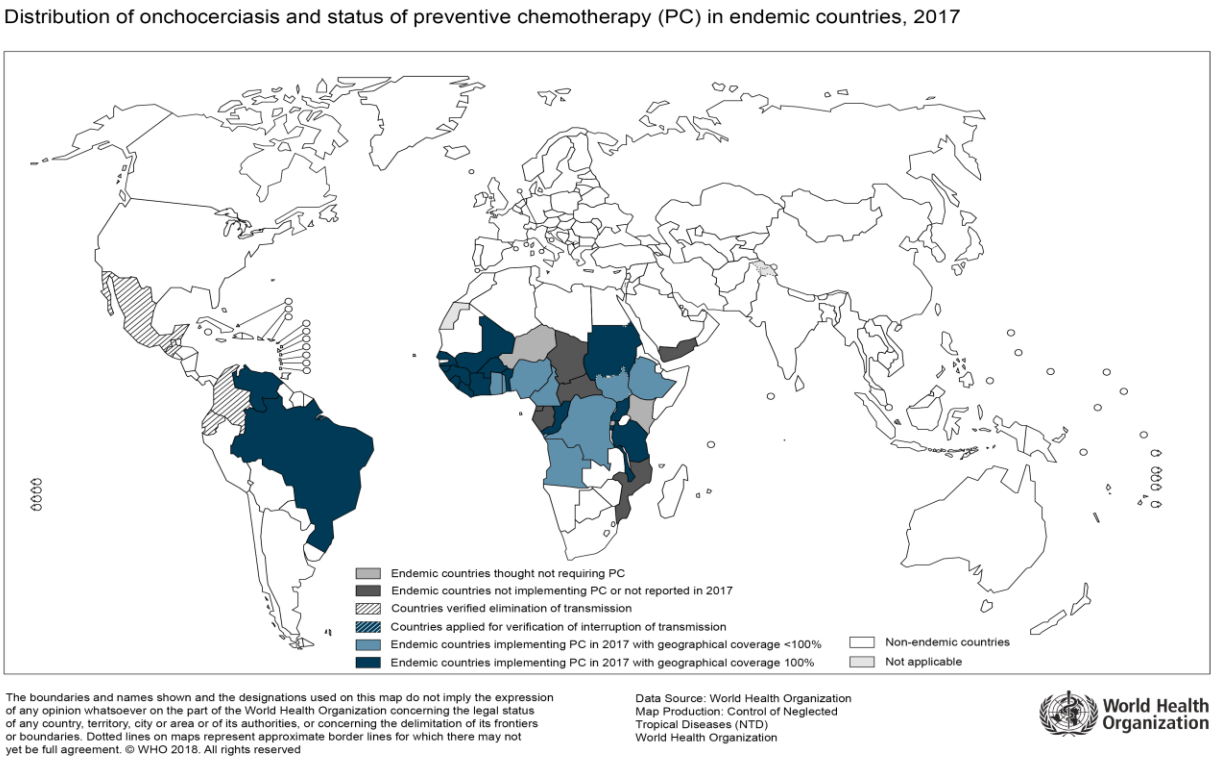

Figure 1.3 Reported worldwide distribution and status map of preventive chemotherapy for onchocerciasis

Dark blue highlighted areas are endemic populations using chemotherapy treatments for onchocerciasis. Light gray areas are endemic populations not implementing this strategy and dark gray areas are populations that have not reported onchocerciasis outbreaks in 2017. Photo credit: World Health Organization (WHO) (2017).

Other diseases include *Haemoproteus*, *Plasmodium*, *Trypanosoma*, and *Leucocytozoon*, blood invasive protozoa of birds that are transmitted by arthropod vectors, such as black flies (Simuliidae) and biting midges (Culicoides) (Getting 1945, WHO 1971, Noblet et al. 1975, Rohner et al. 2000, Sato et al. 2008, Jones et al. 2014). Of these, *Leucocytozoon* is perhaps the most economically important, causing much morbidity and mortality among commercial poultry

annually. In 1945, leucocytozoan infections killed 5,000 turkeys out of a flock of 8,000 in Manitoba (Freeden 1977). From 1942 – 1951, leucocytozoonosis caused an estimated three-quarters of a million dollars in domestic turkey losses (Adler and McCreadie 2009). Many of these poultry and domestic birds were housed in outdoor poultry houses. The risk for leucocytozoonosis transmission has decreased immensely over the past decade, possibly due to indoor poultry housing protection (Adler and McCreadie 2009). Additional disease agents sometimes transmitted by black flies include Venezuelan equine encephalitis (VEE) and eastern equine encephalitis (EEE) (WHO 1971, Jones et al. 1977, Adler and McCreadie 2009).

Purpose of this study

To-date, there has not been a lot of information on the economic impacts that black flies have in Mississippi. They are a competent vector of leucocytozoonosis and have potential detrimental effects on livestock, poultry, and humans. This project was a preliminary study to obtain base-line data on how these insects negatively affect Mississippi communities. This work was accomplished by surveying Mississippi black flies, with particular emphasis on the pest species. By systematically collecting and identifying black fly species occurring in Mississippi, seasonality and factors affecting outbreaks were determined. In addition, using health department data and survey results obtained from backyard poultry owners, we assessed nuisance effects and economic impacts on people, poultry, and livestock.

Dissertation Objectives

1. Obtain as much information as possible about black flies occurring in Mississippi and their biology/ecology to help determine nuisance effects and economic impacts they have produced in Mississippi.

2. Re-type and make available to the public six lost (unpublished) works about black flies in Mississippi from the 1930s. These, conducted by Dr. George H. Bradley, Associate Entomologist with the United States Department of Agriculture, were apparently never published. They were ultimately donated to the Mississippi Entomological Museum at Mississippi State University. A careful search of the USDA Pest Survey Bulletins and the other bibliographic sources from 1928 – 1937 revealed; they were never published anywhere and were not available to the scientific community.
3. Compile an annotated list of black flies occurring in Mississippi using our (current) collection data and records from the Mississippi State University Mississippi Entomological Museum (MEM).
4. Systematically collect and identify black flies from ten sites in Mississippi over a two-year period to determine primary pest species in the state.
5. Evaluate how black flies affect people, domestic animals, and the economic impacts they have in Mississippi.

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CHAPTER II

THE GEORGE H. BRADLEY BLACK FLY PAPERS (DIPTERA: SIMULIIDAE)

Results from this chapter have been previously published in The Midsouth Entomologist (2015) 8(2): 73 – 75. This publication is available at http://www.midsouthentomologist.org.msstate.edu.

Abstract

Dr. George H. Bradley, a former USDA and Army entomologist, conducted research on a variety of medically important insects during his career, most notably mosquitoes (Diptera: Culicidae) and black flies (Diptera: Simuliidae). Much of Dr. Bradley's personal library was donated to the Mississippi Entomological Museum at Mississippi State University after his death. Six of Dr. Bradley's papers in that library, apparently never published, were discovered concerning the ecology and biology of the southern buffalo gnat, which he called *Eusimulium pecuarum* (now known as *Cnephia pecuarum*). These unpublished works provide much needed and never before seen information on this medically important pest. This current paper now makes those papers freely available with open access to interested researchers at the link provided.

Introduction

George H. Bradley (1893 – 1983) was an American medical entomologist who had a long and distinguished career during the 20th Century as a public health official in the U.S. Centers for

Disease Control and the U.S. Department of Agriculture. He was an Associate Entomologist at the Division of Insects Affecting Man and Animals, Bureau of Entomology and Plant Quarantine, U.S. Department of Agriculture during the 1930s; Head of the Entomology Division, Office of Malaria Control, Public Health Service, during World War II; and later, Chief Entomologist, Centers for Disease Control, Atlanta, Georgia. Dr. Bradley was one of the primary workers in the National Malaria Education program which ended in 1951 (Bradley 1966). Dr. Bradley published on a variety of medical and veterinary pests, but most notably mosquitoes (Diptera: Culicidae) (Bradley and McNeel 1928, Bradley et al. 1940, Bradley 1948, King et al. 1960, Bradley 1966) and black flies (Diptera: Simuliidae) (Bradley and McNeel 1928, Bradley 1935a, b). During the early 1930s, he conducted detailed experiments on black flies in various small rivers in the Mississippi Delta. Results of at least two of these experiments were published (Bradley 1935a, b), but several others were not. The purpose of this chapter is to make these unpublished papers available to the public.

Figure 2.1 A “skinned” mule that was greased as a control method in Crowder, MS

Dr. George Bradley recorded and documented treatments for protection on livestock. Photo credit: Dr. George Bradley (1932), USDA.

Materials and Methods

Much of Dr. Bradley's personal library was donated to the Mississippi Entomological Museum (MEM), Mississippi State University after his death via Col. Bill Pearson, the U.S. Army consultant to the Surgeon General who received his Master of Science degree in Entomology from Mississippi State University. MEM personnel and the authors closely examined his handwritten and typed papers and discovered six of his black fly papers were (apparently) never published other than one-paragraph summaries in the USDA Insect Pest Survey Bulletin (Bradley and McNeel 1928, Bradley 1934). Some of the pictures from his research are in the figures shown in Appendix B (Figures B1 – B11).

Results and Discussion

Six never-published papers of Dr. Bradley's concerning the ecology and biology of the southern buffalo gnat that he called *Eusimulium pecuarum*, now known as *Cnephia pecuarum* (Adler et al. 2004), have been re-typed, the photos and figures scanned, and digital files stored on a server at the MEM, Mississippi State University. They are now available for free and open access to interested researchers at the link below. *Note: the research and writing associated with these projects were performed while Dr. Bradley was employed by the USDA and are most likely public domain. We have made no attempt to claim copyright for these works. Further, we made no effort to scientifically review, correct, or edit the works. They are presented in their original form.* http://mississippientomologicalmuseum.org.msstate.edu//MEM.Pubs/BradleyPapers.html

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CHAPTER III

AN ANNOTATED LIST OF BLACK FLIES (DIPTERA: SIMULIIDAE) OCCURRING IN MISSISSIPPI

Results from this chapter have been previously published in The Transactions of the American Entomological Society (2018) 144: 551 – 562. This publication is available at http://taes.entomology-aes.org/.

Abstract

Little is known of black fly species occurring in Mississippi, other than that provided in the historical work by Dr. George Bradley in the 1930s. There has been a resurgence of black flies in the state over the past decade, prompting renewed interest in these blood-sucking pests. For this study, adult black flies were collected by hand netting from various locations throughout the state from 2015 through 2018. Samples were also periodically obtained/submitted from veterinarians and concerned citizens since 2009, and especially during outbreak years (2009, 2011, 2012, 2018). In addition, larval and adult specimens were found in the Mississippi State University Entomological Museum (MEM), that had been collected over several decades by a variety of entomologists and students. Data from approximately 14,471 black fly specimens were analyzed to compile a list of species from the state and to describe their seasonal and geographic distributions. Twenty-seven species of black flies were confirmed from Mississippi, including four new state records.

Introduction

Black flies (Diptera: Simuliidae) are significant medical and veterinary pests throughout the world. There are more than 2,000 known black fly species worldwide, and about, ten percent are known to attack humans (Adler and McCreadie 2009). Species that do not bite people may still be worrisome pests by swarming around the head and landing on ears, face, and neck (Weed 1904, Stone 1963, Mock and Adler 2002). On animals, they often attack sites with little or no pelage, like the eyes, teats, udders, and underbelly (Webster 1887, Fallis 1964, WHO 1971, Jones et al. 1977, Freeden 1985). In large swarms, black flies can cause exsanguination or suffocation (Jones et al 2014, Nations et al. 2015) and make outdoor life intolerable during spring and early summer (Weed 1904, Davies and Peterson 1956, Gray et al. 1996, Adler and McCreadie 2009, Zema 2011). These intense nuisance effects from black flies have sometimes led to descriptions such as “blood-sucking demons” (Gudgel and Grauer 1954, Adler and McCreadie 1997).

Figure 3.1 Young girl being attacked by black flies in Natchez, MS

Black fly reemergence has had tremendous and detrimental effects on outdoor activities. This young girl, from Natchez, MS, Adams County, was attacked by black flies. Photo credit: The Natchez Democrat (June 1, 2011). www.natchezdemocrat.com/2011/06/01/buffalo-gnats-a-temporary-annoyance/

Disease agents that can be transmitted by black flies include those causing human onchocerciasis (river blindness), bovine onchocerciasis, masonellosis, avian leucocytzoonosis, and several arboviruses (Getting 1945, Noblet et al. 1975, Collins 1979, WHO 1989, Richards Jr et al. 2001, Hoerauf 2011, Dhiman et al. 2014, Jones et al. 2014). Not much is known about black fly species in Mississippi with the exception of the historical work by George H. Bradley in the 1930s (Nations 2015) wherein he conducted extensive field studies on the seasonality, location of breeding sites, duration and mode of attacks, conditions of river habitats, treatments, and control methods for the black fly species, *Cnephia pecuarum*, the buffalo gnat (Gray et al. 1996, Nations et al. 2015). Since the 1930s, black fly problems subsided in the southern states; however, there has been a resurgence of black flies since 2009 (Zema 2011, Jones et al. 2014).

The purpose of this study was to determine which species of black flies occur in Mississippi (both historically and currently), as well as their seasonality and geographic distribution.

Materials and Methods

Adult specimens were systematically collected monthly by hand netting year-round for two years (2015-16) in 10 counties in Mississippi. Some of the research sites described in Dr. Bradley's papers were chosen in this present study. Non-scheduled adult collections were also made in other counties between 2009 – 2018. Specimens were placed in 70% ethanol, identified at the Mississippi State University Entomology Department, and subsamples sent to Dr. P.H. Adler (Clemson University) for confirmation.

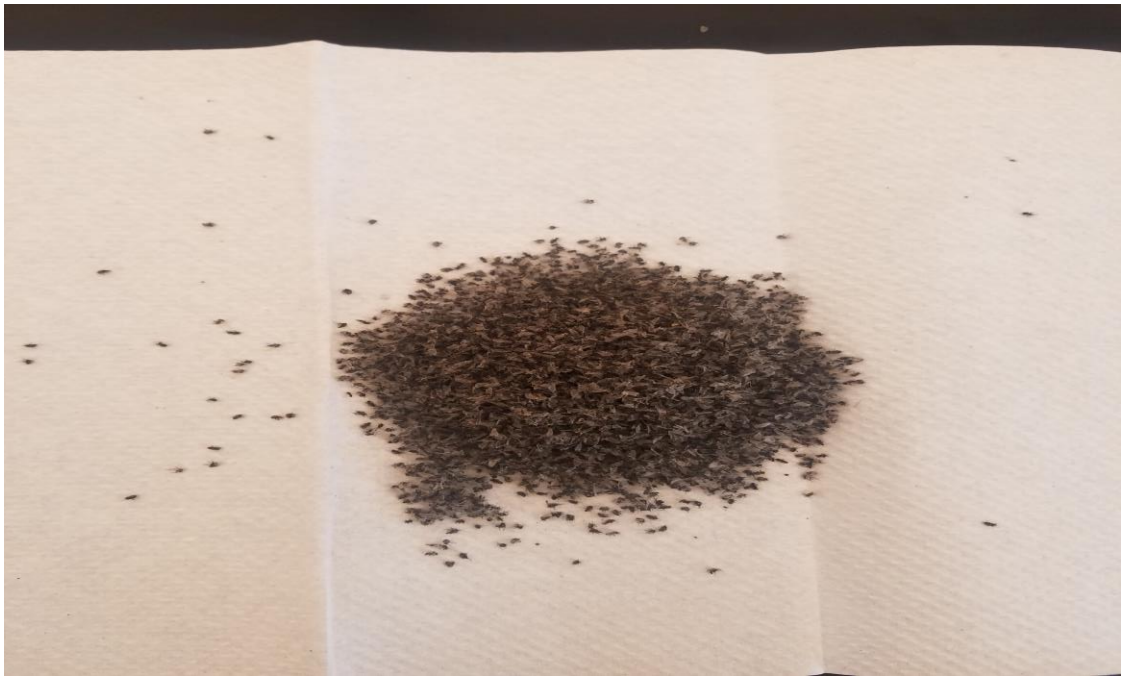

Figure 3.2 Black flies collected from one trap during the 2018 outbreak near the Pearl River

Over 1500 black flies were collected in one trap during the 2018 outbreak near the Pearl River. Estimated adult(s) size, 1 – 5mm. Photo credit: Tina Nations (2018).

Additional adult, larval and pupal specimens from the Mississippi Entomological Museum (MEM) are included in this study. Also, some records are drawn from the database associated with these specimens by Adler et al. (2004). Below is an annotated list of black fly species found in Mississippi. Voucher specimens are deposited in the MEM. New state records are noted in parentheses.

Figure 3.3 CDC CO_2-baited light trap with black flies

To document the 2018 black fly outbreak near the Pearl River, CDC CO_2-baited light traps were set out daily for a week. Over 8,000 black flies were collected. Photo credit: Tina Nations (2018).

Annotated List of Mississippi Black Flies

Prosimulium arvum Adler & Kim

Large numbers of larvae and pupae of this species have been collected in north-central Mississippi during February and March.

Material examined – Mississippi: Carroll County, 2nd tributary of Little Sand Creek, intersection with Hwy 82, 3.6 miles North of the Montgomery County line, 21.II.1988, T.L.

Schiefer and M.F. Hodges Jr (155 larvae). Lowndes County, Ellis Creek and intersection with East Nashville Ferry Road, 28.II.1988, T.L. Schiefer (7 larvae and 2 pupae). Webster County, 2nd tributary of Lindsay Creek, The Cove, 4.III.1988, T.L. Schiefer (35 larvae); 2nd tributary of Lindsay Creek, The Cove, 11.III.1988, T.L. Schiefer (72 larvae).

Number of specimens examined – 271

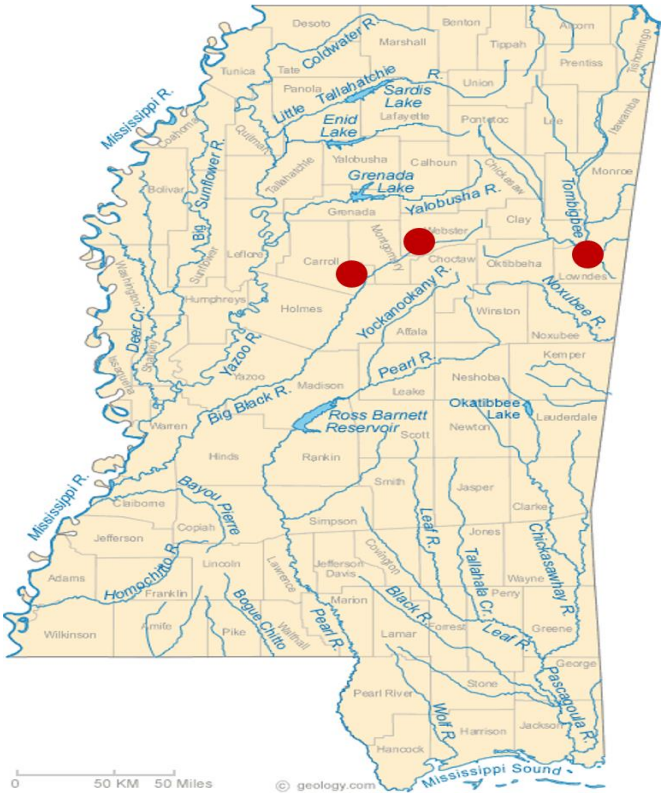

Figure 3.4 *Prosimulium arvum* Adler & Kim distribution map

Mississippi counties where *P. arvum* have been collected or documented. Map credit: Geology.com.

***Prosimulium magnum* Dyar & Shannon (New State Record)**

We have only two collections of *Prosimulium magnum* complex from Mississippi. Two larval specimens were collected in extreme northeastern Mississippi during March.

Material examined – Mississippi: Tishomingo County, Tishomingo State Park at Bear Creek, 7.III.1987, T.L. Schiefer (1 larva); Tishomingo State Park, Rock Quarry Branch, 7.III.1987, T.L. Schiefer (1 larva).

Material examined – 2

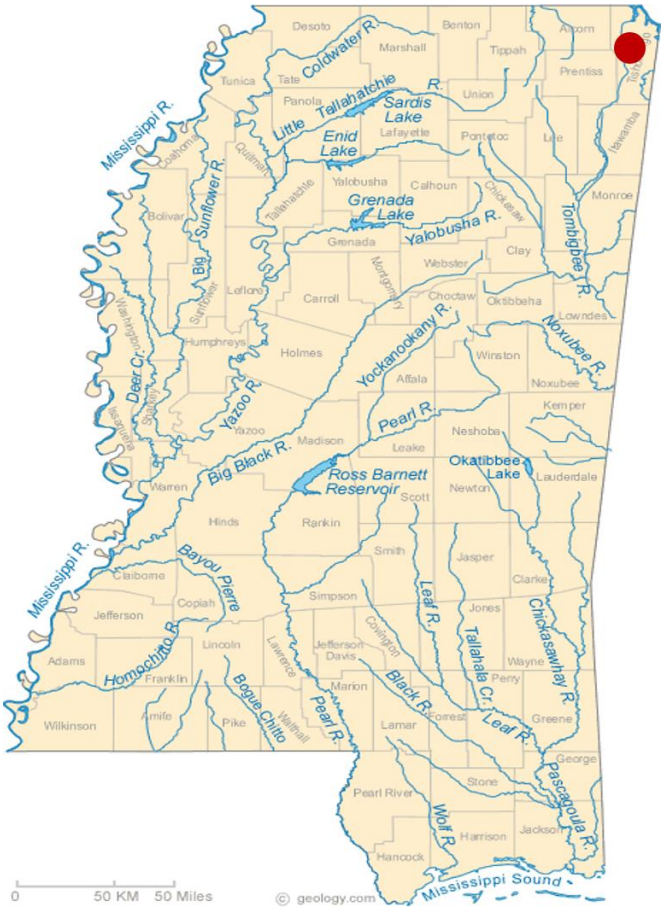

Figure 3.5 *Prosimulum magnum* complex Dyar & Shannon distribution map

Mississippi counties where *P. magnum* complex have been collected or documented. Map credit: Geology.com.

***Stegopterna diplomutata* Currie & Hunter**

Adult specimens were not collected during this project, but Adlter et al. (2004) recorded collections from the northeastern part of Mississippi. Larvae of this species were collected in February and March.

Material examined – Mississippi: Webster County, 2nd tributary of Lindsay Creek, The Cove, T20N-R8E-S12, 11.III.1989, T.L. Schiefer (1 larva). Wilkinson County, Clark Creek Natural Area, Clark Creek, 11.III.1989, T.L. Schiefer & J. MacGowan (5 larvae). Webster County, 2nd tributary of Lindsay Creek, The Cove, T20N-R8E-S12, 18.III.1989, T.L. Schiefer (10 larvae). Tishomingo County, J.P. Coleman State Park, Camp Hollow, 11.II.1990, T.L. Schiefer (9 larvae).

Number of specimens examined – 25

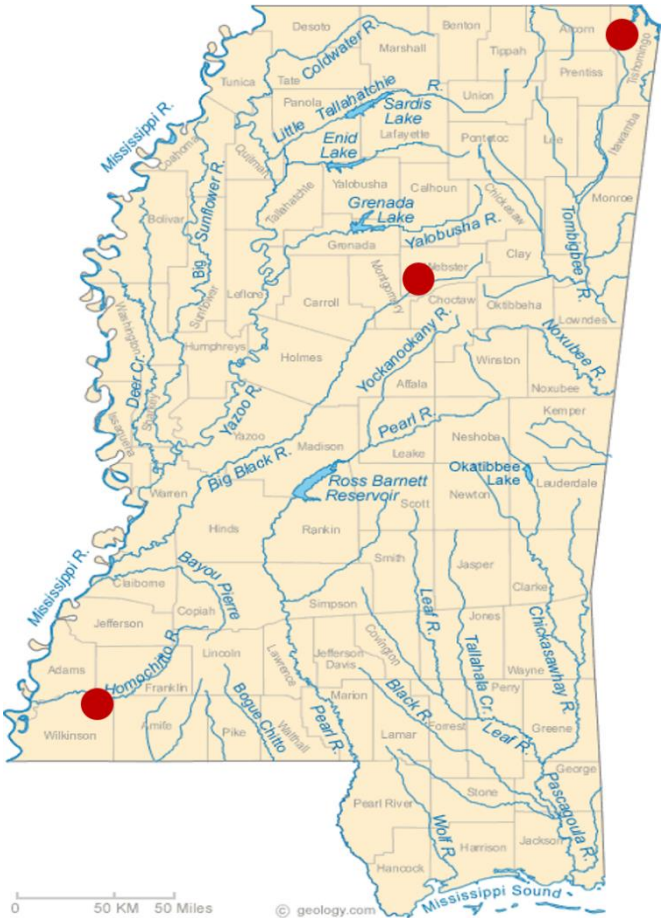

Figure 3.6 *Stegopterna diplomutata* Currie & Hunter distribution map

Mississippi counties where *S. diplomutata* have been collected or documented. Map credit: Geology.com.

***Stegopterna mutata* complex (Malloch)**

No adult specimens have been collected in Mississippi, but larvae and a pupa, probably of *Stegopterna diplomutata* have been collected in March.

Material examined – Mississippi: Tishomingo County, Tishomingo State Park, Bear Creek, 7.III.1987, T.L. Schiefer (1 larva and 1 pupa). Webster County, 2nd tributary of Lindsay Creek, The Cove, 11.III.1988, T.L. Schiefer (3 larvae).

Number of specimens examined – 5

Figure 3.7 *Stegopterna mutata* complex (Malloch) distribution map

Mississippi counties where *S. mutata* complex have been collected or documented. Map credit: Geology.com.

***Cnephia ornithophilia* Davies, Peterson & Wood**

This black fly feeds primarily on birds and has been found in central and southern Mississippi during January and February. Adults have only been collected in February.

Material examined – Mississippi: Noxubee County, Noxubee Wildlife Refuge, near Bluff Lake spillway, 31.I.1987, T.L. Schiefer (1 larva and 1 pupa). Forrest County, Ragland Hills (T3N-R12W-S24W), 17.II.1990, D.M. Pollock (2 adults); near Ragland Hills (T3N-R12W-

S24W), 17.II.1990, T.L. Schiefer (8 adults). Winston County, Tombigbee National Forest (33º10’20”N, 89º03’55”W), 9.II.1999, R. Brown and J. MacGowan (2 adults).

Number of specimens examined – 14

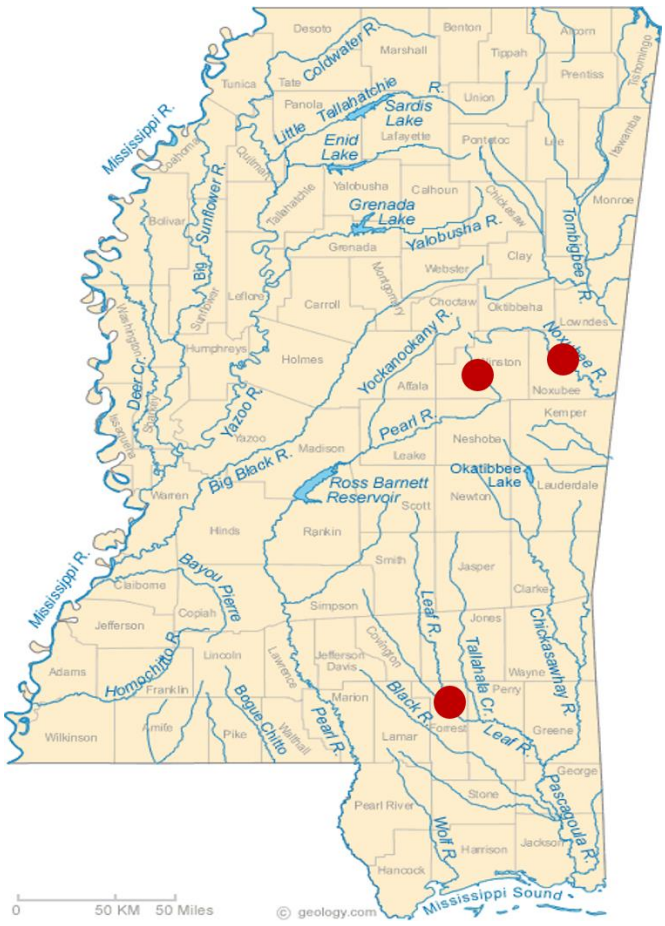

Figure 3.8 *Cnephia ornithophilia* Davies, Peterson & Wood distribution map Mississippi counties where *C. ornithophilia* have been collected or documented. Map credit: Geology.com

***Cnephia pecuarum* (Riley)**

The southern buffalo gnat is probably the most notorious black fly in the Mississippi Delta region. Historically, this black fly caused devastating livestock losses in the early to middle 1930s (Nations et al. 2015). *Cnephia pecuarum* feed primarily on humans and domestic animals,

such as poultry and livestock. This species has been collected in northern and central Mississippi particularly in the Mississippi Delta region from January through August with a peak in March and April.

Material examined – Mississippi: Oktibbeha County, Starkville, 14.VII.1921, A. McIntosh (1 adult). Tallahatchie County, Charleston, 11.IV.1927, G.R. Fulton (4 adults). Yazoo County, Yazoo City, 14.IV.1927, Chesley Hines (4 adults). Sharkey County, Rolling Fork, 26.I.1932, Clay Lyle (6 adults). Leflore County, Greenwood, 16.III.1932, D.W. Grimmes (4 adults); Greenwood, 23.III.1932, D.W. Grimmes (4 adults); Greenwood, 30.III.1932, D.W. Grimmes (1 adult).

Number of specimens examined – 24

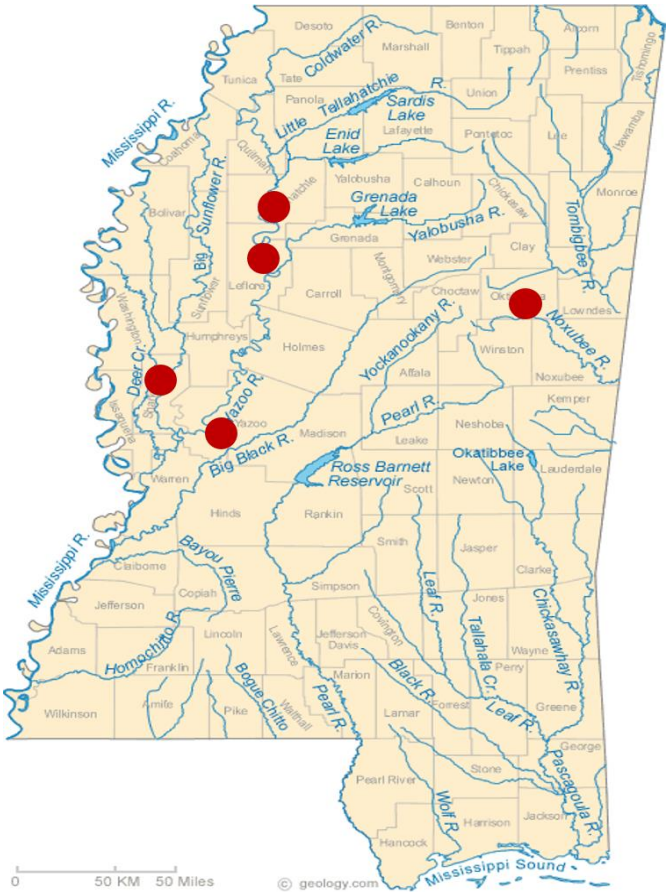

Figure 3.9 *Cnephia pecuarum* (Riley) distribution map

Mississippi counties where *C. pecuarum* have been collected or documented. Map credit: Geology.com.

***Simulium congareenarum* (Dyar & Shannon)**

This species feeds primarily on ducks, turkeys, and chickens. Two specimens have been found in central and southern Mississippi.

Material examined – Mississippi: Webster County, Tributary of Lindsay Creek, The Cove, 4.III.1988, T.L. Schiefer (1 larva). Jackson County, Grand Bay National Wildlife Refuge, Bayou Heron Road (30º25’54.4”N, 88º25’31.9”W), 15.IV.2010, D. Hilderbrandt (1adult female).

Number of specimens examined – 2

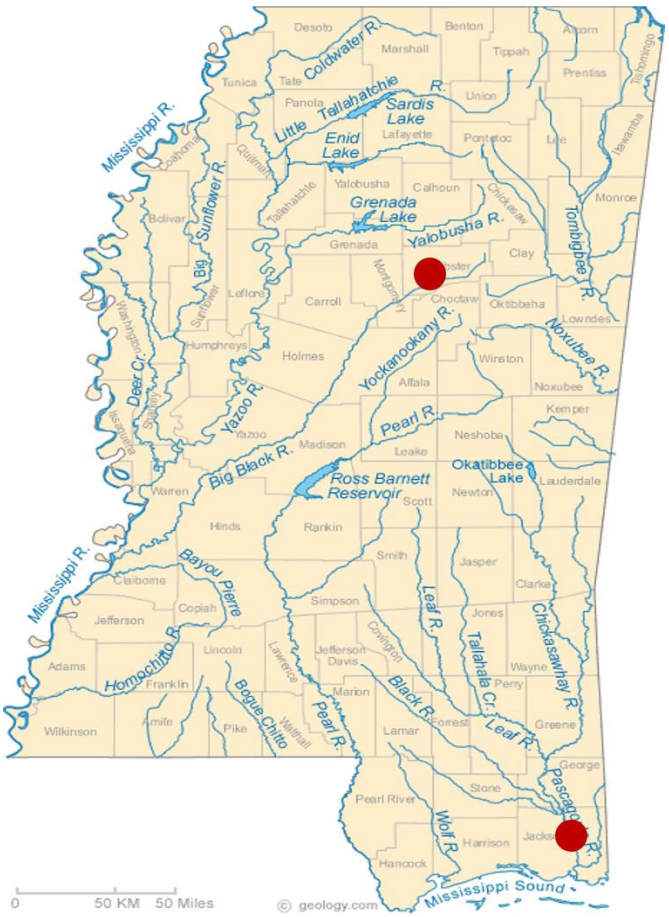

Figure 3.10 *Simulium congareenarum* (Dyar & Shannon) distribution map

Mississippi counties where *S. congareenarum* have been collected or documented. Map credit: Geology.com.

***Simulium johannseni* Hart (New State Record)**

The only specimen we have of *Simulium johannseni* was found during March in southern Mississippi.

Material examined – Mississippi: Covington County, near Seminary, 14.III.2016, J. Goddard (1 adult).

Number of specimens examined – 1

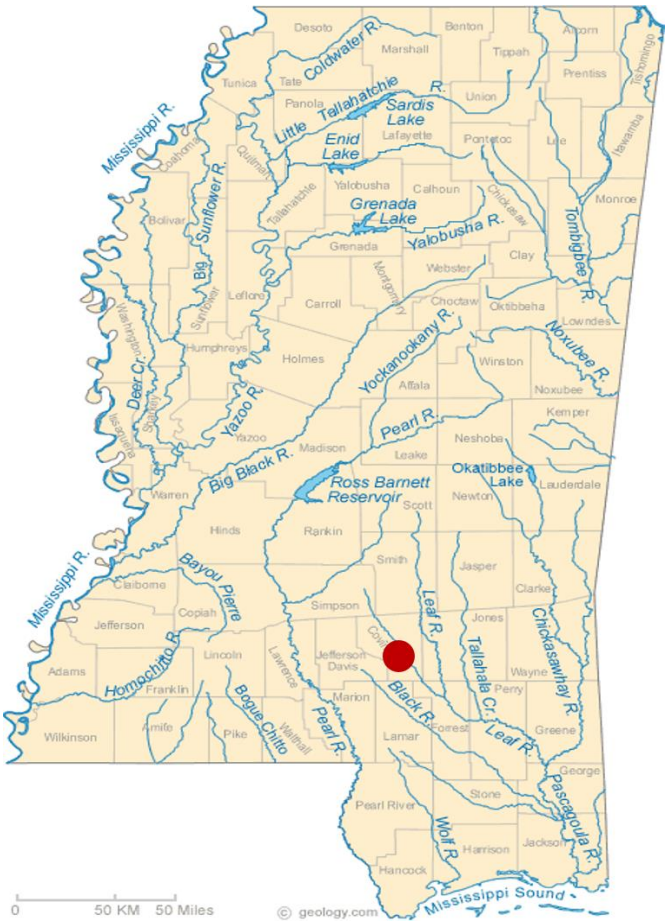

Figure 3.11 *Simulium johannseni* Hart distribution map

Mississippi counties where *S. johannseni* have been collected or documented. Map credit: Geology.com.

***Simulium parmatum* Adler, Currie & Wood (New State Record)**

This species has been collected in March and April, with a peak in April. *Simulium parmatum* is found mainly in southern Mississippi.

Material examined – Mississippi: unknown county, unknown location, 15.IV.1920, F.M.H. Lill (1 adult female). Oktibbeha County, Agr. Coll., 8.IV.1922, E.W. Stafford (3 adult females). Jackson County, Pascagoula River Wildlife Management Area (30º37’46”N, 88º36’10”W), 15.III.1994, D.M. Pollock (2 adults); Pascagoula River Wildlife Management

Area (30º37’46”N, 88º36’10”W), 15.III.1994, D.M. Pollock (1 adult). George County, Mixon Lakes (30º50’28”N, 88º45’11”W) sweeping, 8-9.IV.1994, R.L. Brown and D.M. Pollock (1 adult); Pascagoula River Wildlife Management Area (30º52’40”N, 88º46’05”W), 9.IV.1994, R.L. Brown and D.M. Pollock (1 adult); Mixon Lakes (30º50’28”N, 88º45’11”W), 31.III.1995, D.M. Pollock (1 adult); Pascagoula River Wildlife Management Area (30º52’40”N, 88º46’05”W), 1.IV.1995, D.M. Pollock (3 adults); Mixon Lakes (30º50’28”N, 88º45’11”W) in black light trap, 1.IV.1995, D.M. Pollock (2 adults). Covington County, Seminary, 23.II.2016, J. Goddard (1 adult). Clarke County, 10 miles east of Shubuta, 3.III.2016, J. Goddard (1 adult).

Number of specimens examined – 17

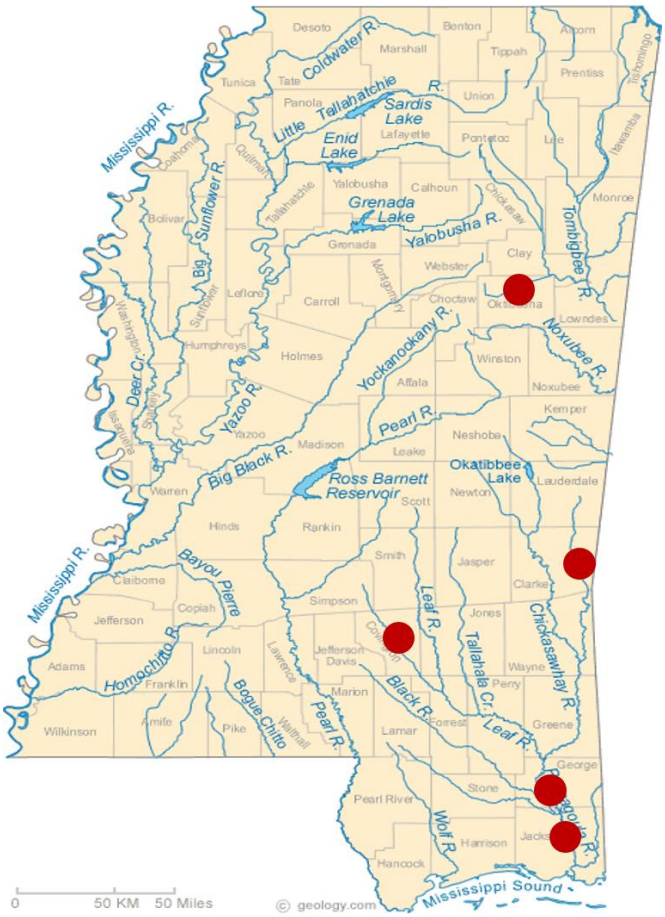

Figure 3.12 *Simulium parmatum* Adler, Currie & Wood distribution map

Mississippi counties where *S. parmatum* have been collected or documented. Map credit: Geology.com.

***Simulium meridionale* Riley**

This is the most common black fly species in Mississippi and is especially troublesome to wild and domestic poultry (Jones et al. 2014). Historically, this species may not have been properly distinguished from *Cnephia pecuarum* (Adler et al. 2004). Adults feed on a variety of birds and mammals but seems to prefer birds. *Simulium meridionale* can be a serious nuisance pest to humans and is probably the black fly targeted in an aerial spraying event at Greenville, MS during June 2011 (Knew 2011). This black fly is found primarily in northern and central

Mississippi, although we have at least one collected from southern Mississippi. Specimens have been reported from February through July with a peak in May and June.

Material examined – Mississippi: Oktibbeha County, Agr. Col., 9.V.1922, unknown collector (1 adult). Washington County, Stoneville, Delta Exp. Forest, 20.IV.1989, J.R. MacDonald (1 adult). Bolivar County, Great River Road State Park (33º50'37"N, 91º03'01"W), 29.IV.1993, D.M. Pollock and R.L. Brown (9 adults); 1.9 miles WNW of Scott (33º36'00"N, 91º06'26"W), 12-13.V.1993, D.M. Pollock (7 adults); Dahomey National Wildlife Refuge (33º42'10"N, 90º55'27"W), 13.V.1993, D.M. Pollock (3 adults). Issaquena County, Mayersville, 29.V.2009. R. Martin (3 adults). Wilkinson County, unknown location, 30.V.2009, D. Adams (4 adults). Warren County, near Vicksburg, 5.VI.2009. K. Jones (101 adults); Still Drive, Vicksburg, 9.VI.2009, K. Jones (20 adults); near Vicksburg, 14.VI.2011, Vicksburg Public Works Department (2000 adults); Redwood, collected from chickens, 18.III.2013, A. Harrison (5 adults); Vicksburg at blockade, 5.IV.2013, J. Goddard (7 adults). Clarke County, near Shubuta, 3.III.2015, J Goddard (1 adult); near Shubuta, 14.IV.2015, J. Goddard (2 adults). Tunica County, near Lula, 23.IV.2015, T. Nations (1 adult). Leflore County, near Money, 23.IV.2015, T. Nations (1 adult). Warren County, Vicksburg, 23.IV.2015, A. Harrison (2 adults). Hinds County, Jackson, 24.IV.2015, W. Varnado (2 adults); Jackson, 6.V.2015, W. Varnado (5 adults). Tunica County, near Lula, 7.V.2015, T. Nations (3 adults). Quitman County, near Sledge, 7.V.2015, T. Nations (2 adults). Tallahatchie County, 6.5 miles east of Webb, 7.V.2015, T. Nations (1 adult). Leflore County, near Money, 7.V.2015, T. Nations (3 adults). Washington County, Greenville, 12.V.2015, J. Goddard (1 adult). Quitman County, near Sledge, 26.V.2015, T. Nations (3 adults). Tallahatchie County, 6.5 miles east of Webb, 26.V.2015, T. Nations (8 adults). Leflore County, near Money, 26.V.2015, T. Nations (3 adults). Washington County, Greenville, 26.V.2015, T.

Nations (1 adult); Greenville, 4.VI.2015, J. Goddard (3 adults). Warren County, Vicksburg, 7.VII.2015, J. Goddard (1 adult). Washington County, Greenville, 14.VII.2015, J. Goddard (2 adults). Covington County, near Seminary, 23.II.2016, J. Goddard (2 adults). Hinds County, Jackson, 27.III.2016, W. Varnado (3 adults); Jackson, Hwy 22 (on chickens), 27.III.2016, A. Harrison (4 adults). Quitman County, near Sledge, 31.III.2016, T. Nations (8 adults). Tallahatchie County, 6.5 miles east of Webb, 31.III.2016, T. Nations (3 adults). Leflore County, near Money, 31.III.2016, T. Nations (2 adults). Hinds County, 15 miles west of Jackson, Hwy 22 (on chickens), 4.IV.2016, A. Harrison (30 adults); Jackson, 13.IV.2016, W. Varnado (3 adults). Quitman County, near Sledge, 15.IV.2016, T. Nations (1 adult). Tallahatchie County, 6.5 miles east of Webb, 15.IV.2016, T. Nations (1 adult). Warren County, Levee Road at Tara Wildlife Refuge, 25.IV.2016, A. Harrison (11 adults). Washington County, near Greenville, 26.IV.2016, J Goddard (1 adult). Tallahatchie County, 6.5 miles east of Webb, 28.IV.2016, T. Nations (2 adults). Leflore County, near Money, 28.IV.2016, T. Nations (4 adults). Hinds County, Jackson, 29.IV.2016, W. Varnado (1 adult). Warren County, Vicksburg, 30.IV.2016, A. Harrison (2 adults). Quitman County, near Sledge, 14.V.2016, T. Nations (2 adults). Tallahatchie County, 6.5 miles east of Webb, 14.V.2016, T. Nations (2 adults).Washington County, Greenville, 19.V.2016, J. Goddard (2 adults). Quitman County, near Sledge, 22.VI.2016, T. Nations (1 adult). Tallahatchie County, 6.5 miles east of Webb, 22.VI.2016, T. Nations (1 adult). Washington County, Greenville, 23.VI.2016, J. Goddard (1 adult). Adams County, Natchez, 31.V.2017, J. Goddard (2 adults); Natchez, 21.VI.2017, J. Goddard (1 adult). Jackson County, Pascagoula Wildlife Management Area, 22.III.2018, J. Goddard (9 adults). Hinds County, Jackson, Westbrook Road, 26.III.2018, W. Varnado (166 adults); Jackson, Westbrook Road, CO_2-baited CDC light trap, 27.III.2018, W. Varnado (6858 adults); Jackson, Westbrook Road,

CO_2-baited CDC light trap, 28.III.2018, T. Nations (1236 adults); Jackson, Westbrook Road, CO_2-baited CDC light trap, 3.IV.2018, W. Varnado (597 adults). Simpson County, Hopewell Road, Harrisville, 5.IV.2018, H. Deerman (27 adults). Adams County, Natchez Trace, Natchez Community, Natchez, 12.IV.2018 (12 adults); Washington Community, Natchez, 12.IV.2018, W. Varnado (5 adults); Margaret Street, Natchez, 12.IV.2018, W. Varnado (12 adults); Highland Street, Natchez, 12.IV.2018, W. Varnado (12 adults). Hinds County, Pearl River, Jackson, 18.IV.2018, W. Varnado (51 adults); Pearl River Outlook, Jackson, 23.IV.2018, W. Varnado (54 adults). Adams County, Mississippi River, Natchez, 30.IV.2018, J. Goddard (3 adults).

Number of specimens examined – 11,338

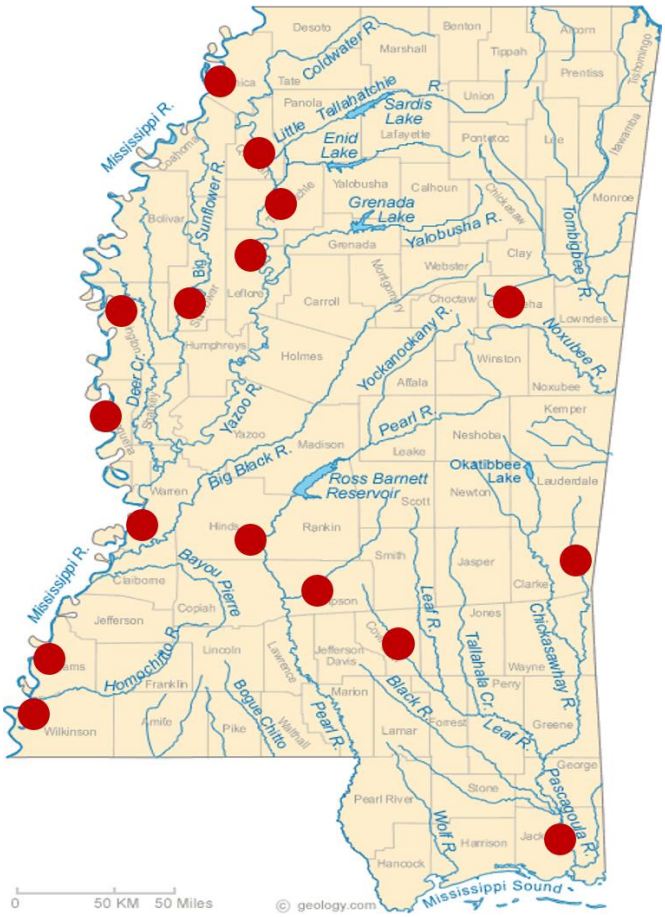

Figure 3.13 *Simulium meridionale* Riley distribution map

Mississippi counties where *S. meridionale* have been collected or documented. Map credit: Geology.com.

***Simulium (Nevermannia) loerchae* Adler**

We did not collect adults of this species, but Adler et al. 2004 recorded specimens collected in the northern and southern parts of Mississippi. Larval specimens were predominately collected during February, while others were collected in March and May.

Material examined – Mississippi: Tishomingo County, J.P. Coleman State Park, Camp Hollow, 11.II.1990, T.L. Schiefer (5 larvae); 0.4 miles east of park entrance in the 2nd tributary of Short Creek, 11.II.1990, T.L. Schiefer (2 larvae). Stone County, 9 miles west of Wiggins,

Sweetbay Bogs Natural Area, 2nd tributary of Kirby Creek, T2S-R13W-S34SW, 13.III.1991, T.L. Schiefer (6 larvae). Marshall County, Wall Doxey State Park, 18.V.1991, J.K. Moulton (4 larvae).

Number of specimens examined – 17

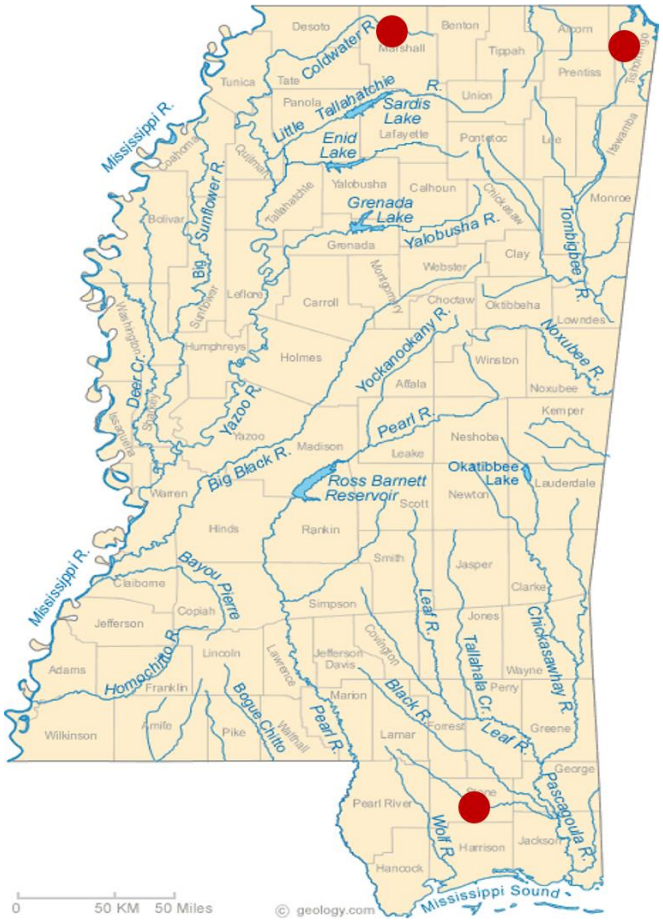

Figure 3.14 *Simulium (Nevermannia) loerchae* Adler distribution map

Mississippi counties where *Simulium (Nevermannia) loerchae* have been collected or documented. Map credit: Geology.com.

***Simulium vittatum* complex Zetterstedt**

No adult specimens of this species have been collected in Mississippi; however, we have larvae and pupae identified by Dr. P.H. Adler from the state. Note: these specimens may actually

be *S. tribulatum*, but we are maintaining them as a separate species (Dr. P.H. Adler, personal communication). Larvae and pupae have been found January through May; larvae peak in March and pupae in April.

Material examined – Mississippi: unspecificed county, unspecified location, unspecified date, unspecified collector (2 larvae). Oktibbeha County, Starkville, Sand Creek, 22.III.1930, William J. Posey (4 larvae); MSU stream, 10.II.1976, W. Gammill (1 larvae); Mississippi State, 25.II.1976, Mizell (4 larvae); Mississippi State, 24.III.1976, L. Thead (8 larvae and 1 pupa); Mississippi State, 5.IV.1978, S. Winters (2 larvae and 14 pupae); unspecified location, 10.IV.1978, P. Ramey (20 larvae); MSU campus, 1.V.1978, B. Schardien (4 larvae and 1 pupa); Mississippi State, 1.III.1980, F.J. Ramolha (3 larvae and 1 pupa); Starkville, 31.III.1982, A. Smith (5 larvae). Noxubee County, Noxubee National Wildlife Refuge, 16.III.1985, N. Harris (1 larva); Noxubee National Wildlife Refuge, 5.V.1985, W.P. Chan (6 larvae and 5 pupae). Oktibbeha County, Starkville, 30.III.1986, A. Ali (20 larvae). Noxubee County, Noxubee National Wildlife Refuge, 1.IV.1986, A. Asquith (1 larva). Tishomingo County, unspecified location, 12.IV.1986, unspecified collector (1 larva). Oktibbeha County, Starkville, Sand Creek at MSU North Farm, 31.I.1987, T.L Schiefer (3 larvae). Noxubee County, Noxubee County Refuge, floodgate at Bluff Lake, 31.I.1987, T.L. Schiefer (30 larvae). Lowndes County, Tributary of Tombigbee River at intersection with Barton Ferry Road, 28.II.1988, T.L. Schiefer (9 larvae and 2 pupae).

Number of specimens examined – 148

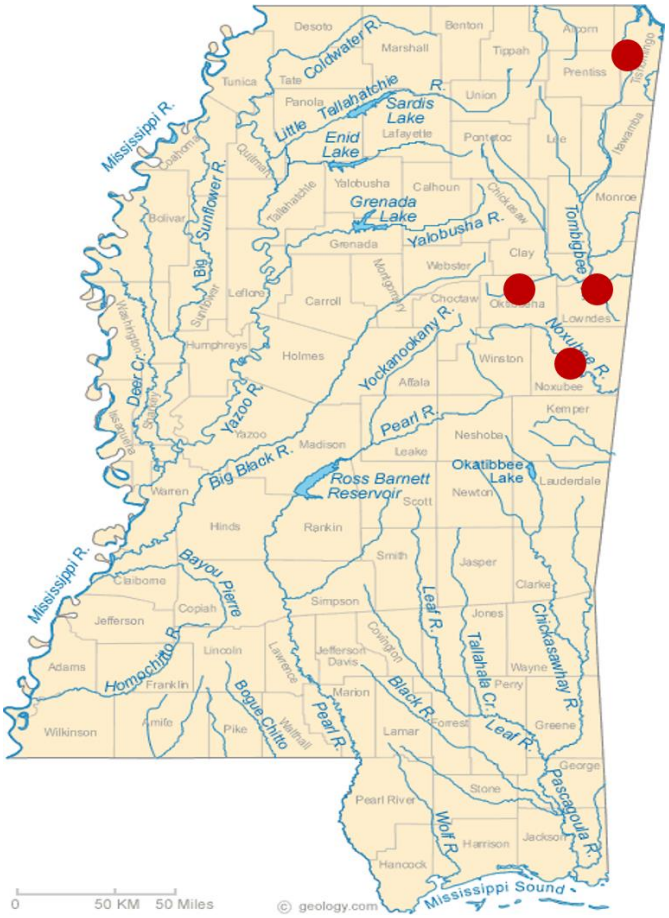

Figure 3.15 *Simulium vittatum* complex Zetterstedt distribution map

Mississippi counties where *S. vittatum* complex have been collected or documented. Map credit: Geology.com.

***Simulium tribulatum* Lugger**

This species has been found in northern and central Mississippi from March through May. Adults have been reported to feed on horses and cattle (Mullens and Dada 1992).

Material examined – Mississippi: Pontotoc County, one-mile South of Ecru in cultivated cotton, 8.V.1981, G.L. Snodgrass (2 adults). Oktibbeha County, Starkville, 29.IV.1981, R.L. Brown (1 adult). Noxubee County, Noxubee National Wildlife Refuge, 5.III.1985, W.P.Chan (1 adult). Oktibbeha County, Sand Creek at MSU North Farm, near Starkville, 31.I.1987, T.L.

Schiefer (3 larvae). Noxubee County, Noxubee National Wildlife Refuge at the floodgate outlet on Bluff Lake, Feb. 1987, T.L. Schiefer (7 larvae). Lowndes County, intersection with Barton Ferry Road, tributary of Tombigbee River, 28.II.1988, T.L. Schiefer (8 larvae and 1 pupa). Washington County, Stoneville, Delta Exp. Forest, 20.IV.1989, J.R. MacDonald (1 adult). Grenada County, Black Creek, T21N-R3E-S7SW, 10.IV.1991, recorded by Adler et al. (2004) (14 larvae and 1 pupae); tributary in Yalobusha River, T22N-R3E-S31NW, 10.IV.1991, T.L. Schiefer (29 larvae and 1 pupa); Black Creek, T21N-R3E-S7SW, 6.VI.1991, recorded by Adler et al. (2004) (6 larvae). Warren County, Vicksburg, 24.III.2015, A. Harrison (1 adult).

Number of specimens examined – 76

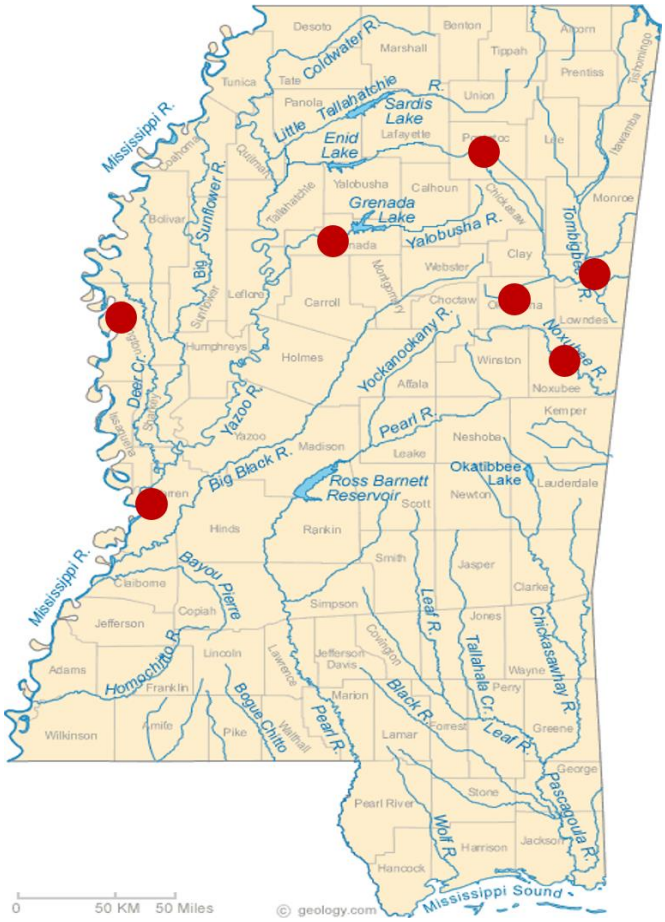

Figure 3.16 *Simulium tribulatum* Lugger distribution map

Mississippi counties where *S. tribulatum* have been collected or documented. Map credit: Geology.com.

***Simulium jenningsi* group Malloch**

This species group is made up of at least 22 species that cannot easily be separated as adults (larvae and pupae can be). In this paper, we have included as separate entities the records of other in the group – *Simulium confusum*, *S. dixiense*, *S. jonesi*, *S. luggeri*, and *S. podostemi* – because they were identified as adults or immatures by Adler et al. (2004). The *S. jenningsi* group is abundant in Mississippi being found virtually statewide. Collections have been made year-round with peak months being April through June. This group is multivoltine and is often

collected in southern Mississippi during mid-winter. Females of *S. jenningsi* group feed on a wide range of mammals and birds (Adler et al. 2004). The females will occasionally bite humans, but humans are not their primary blood meal source.

Material examined – Mississippi: Tishomingo County, Tishomingo State Park, 11.IV.1986, J. MacDonald (1 adult). Choctaw County, Choctaw Lake, 2.V.1995, D.M. Pollock (2 adults). Oktibbeha County, Dorman Lake, 12.V.1995, D.M. Pollock (1 adult). Covington County, near Seminary, 30.III.2012, J. Goddard (8 adults); near Seminary, 30.IV.2012, J. Goddard (6 adults). Simpson County, near Mendenhall, 28.V.2013, J. Goddard (15 adults). Smith County, Taylorsville, near Leaf River, 12.VI.2013, J. Rodgers (4 adults). Covington County, Seminary, 3.XII.2014, J. Goddard (4 adults); near Seminary, 17.I.2015, J. Goddard (8 adults); near Seminary, 26.III.2015, J. Goddard (9 adults). Simpson County, near Mendenhall, 14.IV.2015, J. Goddard (1 adult). Clarke County, near Shubuta, 14.IV.2015, J. Goddard (10 adults). Simpson County, near Mendenhall, 28.IV.2015, J. Goddard (1 adult). Clarke County, near Shubuta, 28.IV.2015, J. Goddard (3 adults). Hinds County, Jackson, 6.V.2015, W. Varnado (3 adults). Covington County, near Seminary, 7.V.2015, J. Goddard (2 adults). Clarke County, near Shubuta, 21.V.2015, J. Goddard (9 adults). Covington County, near Seminary, 21.V.2015, J. Goddard (6 adults). Simpson County, Mendenhall, 9.VI.2015, J. Goddard (2 adults). Clarke County, near Shubuta, 23.VI.2015, J. Goddard (1 adult). Tallahatchie County, 6.5 miles east of Webb, 26.VI.2015, T. Nations (1 adult). Simpson County, near Mendenhall, 9.VI.2015, J. Goddard (2 adults). Clarke County, near Shubuta, 9.VI.2015, J. Goddard (4 adults). Covington County, near Seminary, 9.VI.2015, J. Goddard (1 adult). Simpson County, near Mendenhall, 18.VI.2015, J. Goddard (5 adults). Covington County, near Seminary, 18.VI.2015, J. Goddard (2 adults). Simpson County, near Mendenhall, 3.VII.2015, J. Goddard (10 adults). Covington

County, near Seminary, 3.VII.2015, J. Goddard (10 adults). Clarke County, near Shubuta, 7.VII.2015, J. Goddard (1 adult). Covington County, near Seminary, 17.I.2016, J. Goddard (8 adults). Clarke County, near Shubuta, 23.II.2016, J. Goddard (29 adults); near Shubuta, 14.III.2016, J. Goddard (2 adults). Covington County, near Seminary, 14.III.2016, J. Goddard (2 adults). Warren County, Vicksburg, 15.III.2016, A. Harrison (1 adult). Hinds County, Jackson, 27.III.2016, W. Varnado (4 adults). Clarke County, near Shubuta, 31.III.2016, J. Goddard (1 adult); near Shubuta, 5.IV.2016, J. Goddard (1 adult). Covington County, near Seminary, 5.IV.2016, J. Goddard (16 adults). Simpson County, near Mendenhall, 28.IV.2016, J. Goddard (2 adults). Covington County, near Seminary, 28.IV.2016, J. Goddard (2 adults). Simpson County, near Mendenhall, 12.V.2016, J. Goddard (10 adults). Clark County, near Shubuta, 12.V.2016, J. Goddard (8 adults). Covington County, near Seminary, 12.V.2016, J. Goddard (4 adults). Simpson County, near Mendenhall, 26.V.2016, J. Goddard (7 adults). Covington County, near Shubuta, 26.V.2016, J. Goddard (9 adults). Simpson County, near Mendenhall, 9.VI.2016, J. Goddard (5 adults). Covington County, near Seminary, 9.VI.2016, J. Goddard (21 adults). Clarke County, near Shubuta, 9.VI.2016, J. Goddard (3 adults). Simpson County, near Mendenhall, 21.VI.2016, J. Goddard (2 adults). Covington County, near Seminary, 21.VI.2016, J. Goddard (1 adult). Simpson County, near Mendenhall, 11.X.2016, T. Nations (1 adult). Covington County, near Seminary, 11.X.2016, J. Goddard (11 adults); near Seminary, 15.XI.2016, J. Goddard (7 adults). Itawamba County, Tremont, Bull Mountain Creek, 3.V.2017, J. Goddard (3 adults). Tishomingo County, near Tishomingo, Bear Creek, 25.VII.2017, J. Goddard (4 adults); near Tishomingo State Park, 31.VII.2017, J. Goddard (1 adult); near Tishomingo State Park, 17.VII.2017, J. Goddard (10 adults); Tishomingo State Park, 18.VIII.2017, J. Goddard (2 adults); near Tishomingo, Bear Creek, 24.VIII.2017, J. Goddard (2 adults). Clarke County, near

Carmichael, Buckatunna Creek, 21.IX.2017, J. Goddard (1 adult). Jones County, near Ellisville, Leaf River, 26.IX.2017, J. Goddard (2 adults). Marion County, near Columbia, Pearl River, 26.IX.2017, J. Goddard (12 adults). Jones County, near Ellisville, Leaf River, 5.X.2017, J. Goddard (2 adults). Marion County, Columbia, Pearl River, 5.X.2017, J. Goddard (2 adults). Simpson County, Mendenhall, 11.X.2017, J. Goddard (1 adult). Covington County, Seminary, 28.X.2017. J. Goddard (19 adults). Clarke County, near Carmichael, 7.XI.2017, J. Goddard (1 adult). Tishomingo County, near Tishomingo State Park, 10.XI.2017, J. Goddard (1 adult). Covington County, Seminary, 21.XI.2017, J. Goddard (2 adults). Jackson County, Pascagoula River Wildlife Management Area, 27.II.2018, J. Goddard (1 adult). Jones County, Leaf River, near Ellisville, 22.II.2018, J. Goddard (4 adults). Walthall County, Bogue Chitto Creek, 22.III.2018, J. Goddard (1 adult). Jackson County, Pascagoula Wildlife Management Area, 22.II.2018, J. Goddard (1 adult). Leake County, near Carthage, 27.III.2018, J. Goddard (8 adults); Pearl River, Edinburg, 5.IV.2018, J. Goddard (8 adults). Simpson County, Hopewell Road, Harrisville, 5.IV.2018, H. Deerman (7 adults). Hinds County, Snake Creek, Raymond, 6.IV.2018, W. Varnado (51 adults). Pearl River County, Old River Wildlife Management Area, near Bogalusa, 10.IV.2018, W. Varnado (12 adults). Leake County, Pearl River, near Carthage, 11.IV.2018, J. Goddard (3 adults); Pearl River, Edinburg, 11.IV.2018, J. Goddard (6 adults).

Number of specimens examined – 456

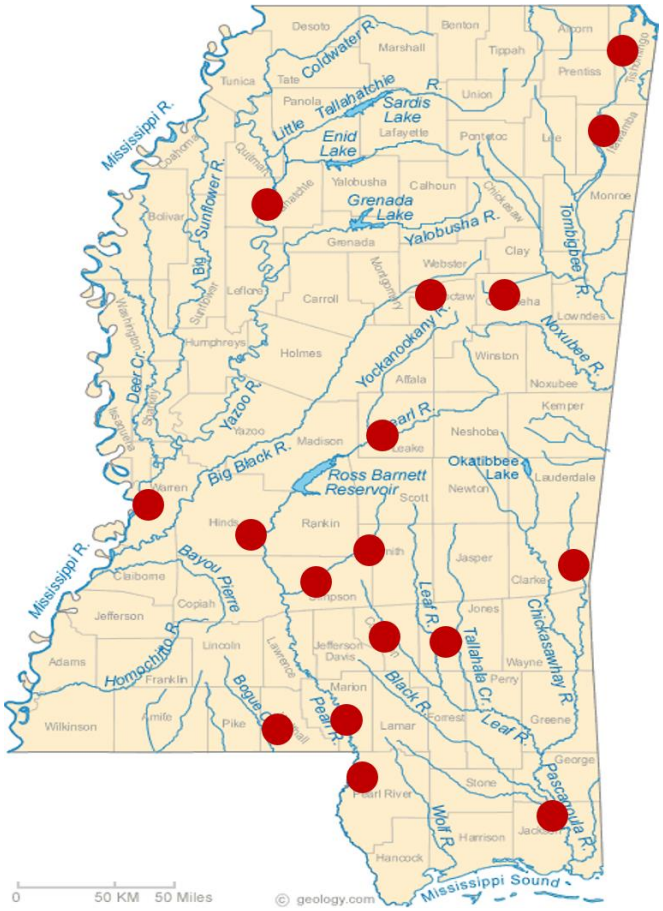

Figure 3.17 *Simulium jenningsi* group Malloch distribution map

Mississippi counties where *S. jenningsi* group have been collected or documented. Map credit: Geology.com.

***Simulium confusum* Moulton & Adler**

We did not collect specimens of *Simulium confusum* in Mississippi during this project, but Adler et al. (2004) documented specimens collected from the central and southern Mississippi Delta.

Material examined – Mississippi: Yazoo County, Eden, 15.IV.1941, A. Stone (1 pupa). Hinds County, Big Black River, 16.IV.1941, A. Stone (2 male adults, 3 female adults with exuvia, and pupae). Grenada County, Black Creek, T21N-R3E-S7SW, 10.IV.1991, T.L. Schiefer (3 larvae); Black Creek, T21N-R3E-S7SW, 6.VI.1991, T.L. Schiefer (83 larvae, 1 pupa, and 1

exuvia); Black Creek, 24.VII.1991, T.L. Schiefer (42 larvae, 5 pupae, and 1 exuvia). Madison County, Big Black River, Rt. 49, 16.III.1992, P.H. Adler and J.K. Moulton (21 larvae, 15 pupae, 21 male adults, 23 female adults with exuvia). Grenada County, Black Creek, T21N-R3E-S7SW, 20.III.1992, T.L.Schiefer (53 larvae). Franklin County, Homochitto National Forest, Middleton Creek, T5N-R4E-S31S, 7.IV.1992, T.L. Schiefer (6 larvae).

Number of specimens examined – 281

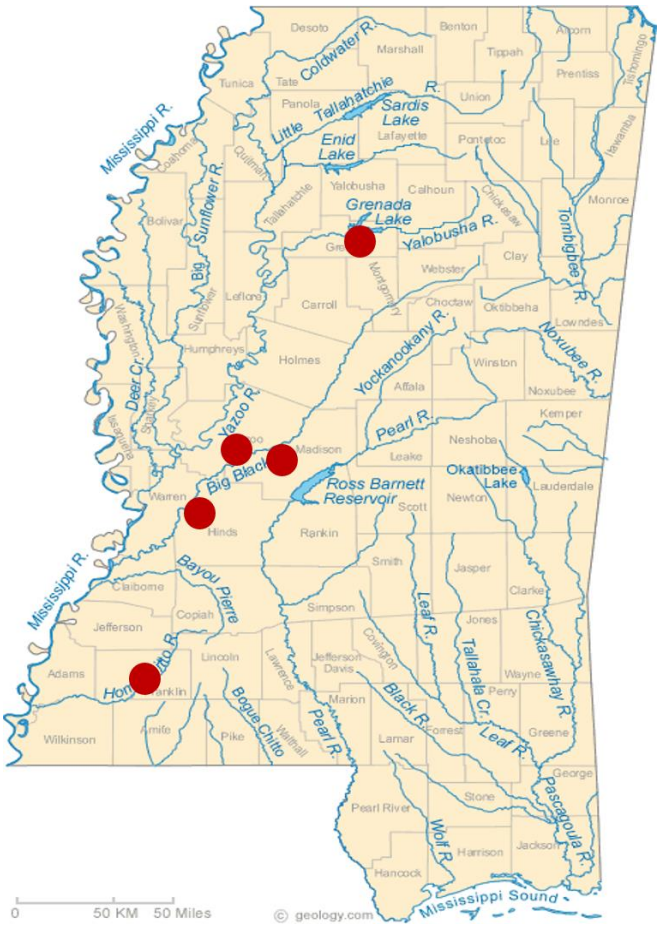

Figure 3.18 *Simulium confusum* Moulton & Adler distribution map

Mississippi counties where *S. confusum* have been collected or documented. Map credit: Geology.com.

***Simulium dixiense* (Stone & Snoddy)**

No specimens were collected in Mississippi during this project, but Adler et al. (2004) recorded specimens collected from southern counties along the Mississippi Gulf Coast.

Material examined – Mississippi: Harrison County, 2.5 miles north northeast of Saucier, Tuxachanie Trail, tributary of McHenry Branch, 10.III.1991, T.L. Schiefer (2 pupae). Jackson County, 2.75 miles northeast of Vancleave, Mounger Creek, 11.III.1991, T.L.Schiefer (6 larvae). George County, 4.5 miles northwest of Lucedale, Skinner Creek, 12.III.1991, T.L. Schiefer (8 larvae).

Number of specimens examined – 16

Figure 3.19 *Simulium dixiense* (Stone & Snoddy) distribution map

Mississippi counties where *S. dixiense* have been collected or documented. Map credit: Geology.com.

***Simulium jonesi* Stone & Snoddy**

This species feeds primarily on mammals and can be a human nuisance pest. There is only one documented collection of *S. jonesi* in Mississippi during July, but there are other Mississippi collections of *S. jonesi* documented by P.H. Adler.

Material examined – Mississippi: Grenada County, (T21N-R2E-S12), 5-11.VII.1991, T.L. Schiefer (1 adult male).

Number of specimens examined – 1

Figure 3.20 *Simulium jonesi* Stone & Snoddy distribution map

Mississippi counties where *S. jonesi* have been collected or documented. Map credit: Geology.com.

***Simulium luggeri* Nicoholson & Mickel**

During this project, no *Simulium luggeri* specimens were collected, but Adler et al. 2004 documented specimens collected from Mississippi.

Material examined – Mississippi: Grenada County, Black Creek, T21N-R2E-S75W, 6.VI.1991, T.L. Schiefer (1 mature larva).

Number of specimens examined – 1

Figure 3.21 *Simulium luggeri* Nicholson & Mickel distribution map

Mississippi counties where *S. luggeri* have been collected or documented. Map credit: Geology.com.

***Simulium podostemi* Snoddy**

No specimens were collected in Mississippi during this project, but Adler et al. 2004 documented specimens from northeastern Mississippi.

Material examined – Mississippi: Tishomingo County, Tishomingo State Park, Bear Creek, 11.II.1990, T.L.Schiefer (13 larvae); Tishomingo State Park, Bear Creek, 2.XI.1990, J.K. Moulton (30 larvae, 31 pupae, 20 male adults, and 22 exuvia).

Number of specimens examined – 116

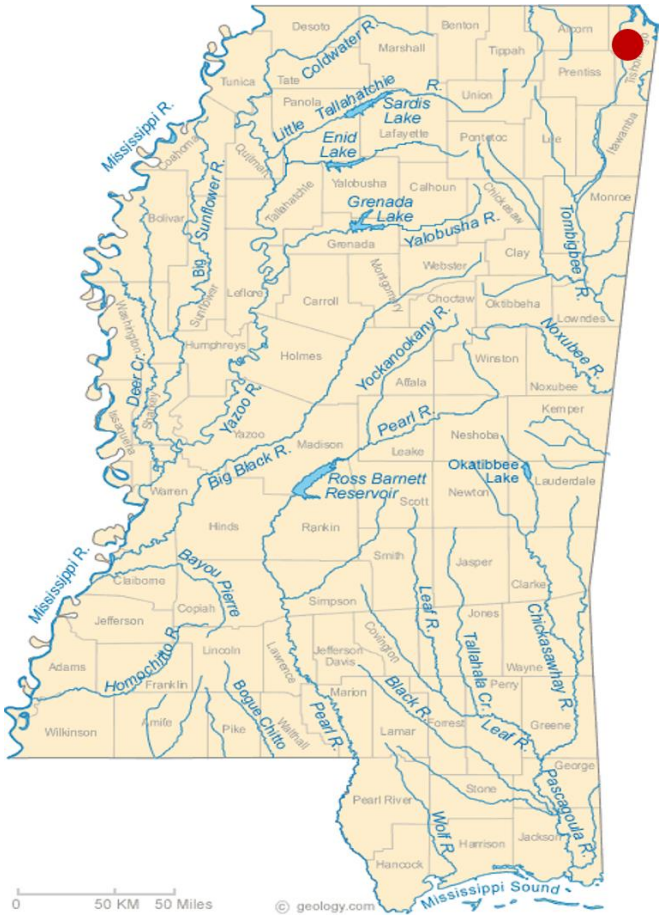

Figure 3.22 *Simulium podostemi* Snoddy distribution map

Mississippi counties where *S. podostemi* have been collected or documented. Map credit: Geology.com.

***Simulium decorum* Walker**

This species has only been documented in one county in Mississippi during January (larvae and pupae) and March (all stages). *Simulium decorum* has a wide range of hosts: large mammals, ducks, geese, crows, grouse, turkeys, chickens, rabbits, foxes, and minks (Davies and Peterson 1956, Anderson and Defoliart 1961). This species may also be a tremendous nuisance pest to humans, flying into the eyes and ears.

Material examined – Mississippi: Noxubee County, Noxubee Wildlife Refuge, 18.III.1984, B.R. Farmer (1 pupa); Noxubee National Wildlife Refuge, 8.III.1984, B. Engber (20 adults); Noxubee National Wildlife Refuge, Bluff Lake Spillway, 19.III.1984, T. Wofford (6 larvae); Noxubee National Wildlife Refuge at floodgate, 21.1.1987, T.L. Schiefer (3 larvae and 8 pupae).

Number of specimens examined – 38

Figure 3.23 *Simulium decorum* Walker distribution map

Mississippi counties where *S. decorum* have been collected or documented. Map credit: Geology.com.

***Simulium slossonae* Dyar & Shannon (New State Record)**

The only specimens of *S. slossonae* were collected in southern Mississippi during March.

Material examined – Mississippi: Jackson County, Pascagoula River Wildlife Management Area (30º37'46"N, 88º36'10"W), 15.III.1994, D.M. Pollock (4 adults).

Number of specimens examined – 4

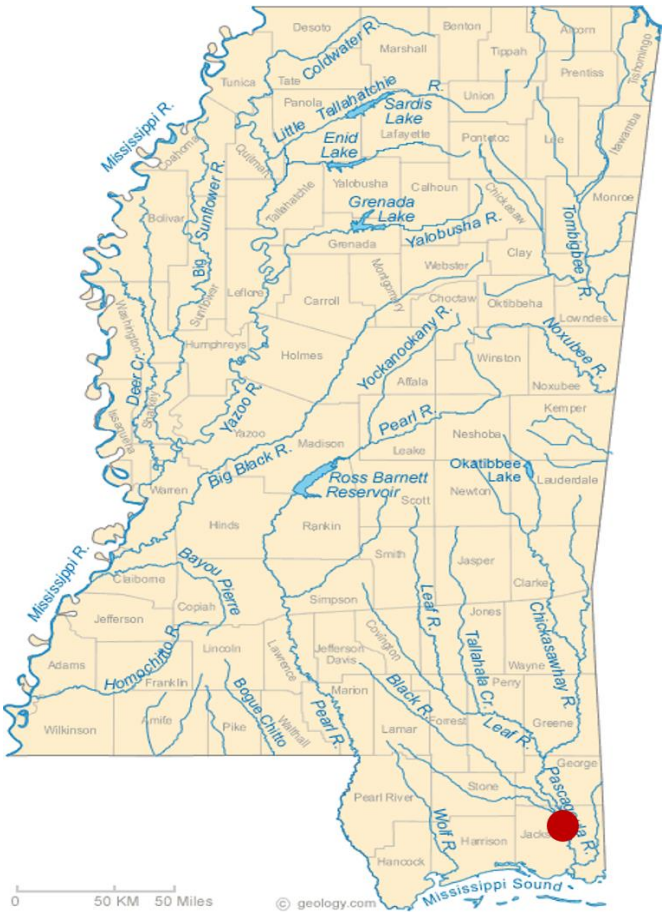

Figure 3.24 *Simulium slossonae* Dyar & Shannon distribution map

Mississippi counties where *S. slossonae* have been collected or documented. Map credit: Geology.com.

***Simulium tuberosum, sensu stricto* (Lundström)**

No adult specimens have been collected of this species in Mississippi, but larvae and pupae have been found in northeastern and southern Mississippi from February through April, with a peak in February and March.

Material examined – Mississippi: Oktibbeha County, Mississippi State, 25.II.1976, Mizell (1 larva); Mississippi State, 24.III.1976, L. Thead (16 larvae and pupae). Tishomingo County, unspecified location, 12.IV.1986, unspecified collector (1 larva); Tishomingo State Park, Rock Quarry Branch, 7.III.1987, T.L. Schiefer (51 larvae and 8 pupae). Lowndes County, Tributary of Luxapilla Creek below beaver dam, 15.III.1987, T.L. Schiefer (4 larvae); Ellis Creek at intersection of Nashville Ferry Road, T19S-R18W-S24, 24.II.1988, T.L. Schiefer (unknown number and stage of specimens); Tributary at Tombigbee River at the intersection with Barton Ferry Road, T16S-R19W-S36, 28.II.1988, T.L. Schiefer (unknown number and stage of specimens. Wilkinson County, Clark Creek Natural Area, Clark Creek, 11.III.1989, T.L. Schiefer (unknown number and stage of specimens). Tishomingo County, Tishomingo State Park, Rock Quarry Branch, 18.IX.1991, T.L. Schiefer (unknown number and stage of specimens). Franklin County, Homochito National Forest, Middleton Creek, T5N-R4E-S31S, 6.II.1992, T.L. Schiefer (many larvae and pupae); Homochito National Forest, Porter Creek, T5N-R4E-S8NW, 8.IV.1992, T.L. Schiefer (138 larvae and 1 pupae); Homochito National Forest, tributary in McGehee Creek, T6N-R4E-S26SW, 8.IV.1992, T.L. Schiefer (249 larvae and 30 pupae).

Number of specimens examined – 504

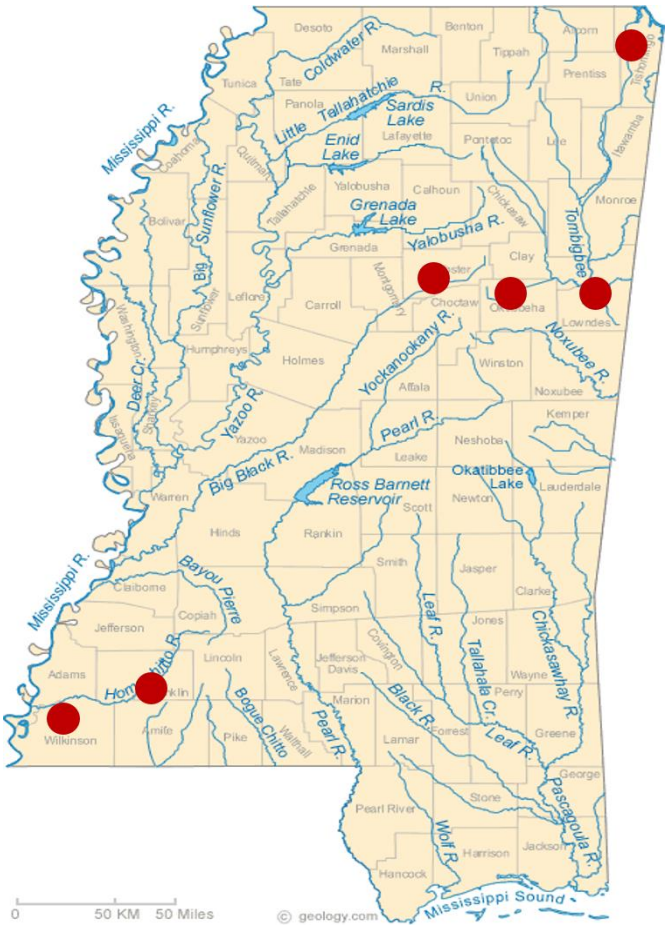

Figure 3.25 *Simulium tuberosum, sensu stricto* (Lundström) distribution map

Mississippi counties where *S. tuberosum, sensu stricto* have been collected or documented. Map credit: Geology.com.

***Simulium perissum* Dyar & Shannon**

We did not collect this species in Mississippi during this project, but Adler et al. (2004) recorded specimens collected in Grenada and Wilkinson counties.

Material examined – Mississippi: Wilkinson County, Clark Creek Natural Area, Clark Creek, 11.III.1989, T.L. Schiefer (1 larva). Grenada County, Black Creek, T21N-R3E-S7SW, 10.IV.1991, T.L. Schiefer (22 larvae); Tributary in the Yalobusha River, T22N-R3E-S31NW, 10.IV.1991, T.L. Schiefer (69 larvae, 9 pupae, and 4 exuvia); Tributary in the Yalobusha River,

T22N-R3E-S31NW, 12.VI.1991, T.L. Schiefer (163 larvae and 5 pupae); Tributary in the Yalobusha River, T22N-R3E-S31NW, 24.VII.1991, T.L.Schiefer (19 larvae and 6 pupae); Black Creek, T21N-R3E-S7SW, 6.II.1992, T.L. Schiefer (40 larvae and 30 pupae); Tributary in the Yalobusha River, T22N-R3E-S31NW, 6.II.1992, T.L. Schiefer (20 larvae and 2 pupae); Black Creek, T21N-R3E-S7SW, 20.III.1992, T.L. Schiefer (many larvae and pupae); Tributary in the Yalobusha River, T22N-R3E-S31NW, 20.III.1992, T.L. Schiefer (many larvae and 1 pupae).

Number of specimens examined – 391

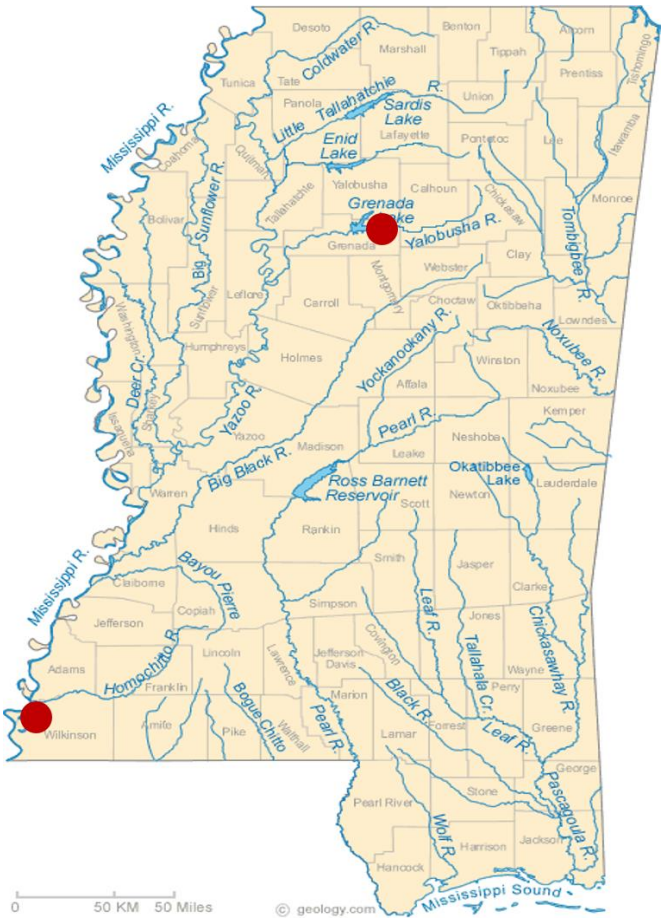

Figure 3.26 *Simulium perissum* Dyar & Shannon distribution map

Mississippi counties where *S. perissum* have been collected or documented. Map credit: Geology.com.

***Simulium ubiquitum* Adler, Currie & Wood**

We have not collected *S. ubiquitum*, but Adler et al. (2004) recorded specimens taken throughout the state of Mississippi.

Material examined – Mississippi: Tishomingo County, Tishomingo State Park, Rock Quarry Branch, 7.III.1987, T.L. Schiefer (unknown number and stage of specimens). Grenada County, Tributary in Clark Creek, T23N-R7E-S16E quarter, 9.IV.1987, T.L. Schiefer (unknown number and stage of specimens). Carroll County, 3.6 miles west of Montgomery County at the intersection of Hwy 82, 2nd tributary of Little Sand Creek, T19N-R4E-S22, 21.II.1988, T.L. Schiefer (unknown number and stage of specimens). Webster County, 2nd tributary of Lindsay Creek, The Cove, T20N-R8E-S12, 12.V.1988, T.L. Schiefer (unknown number and stage of specimens); 2nd tributary Lindsay Creek, The Cove, T20N-R8E-S12, 18.III.1989, T.L.Schiefer (unknown number and stage of specimens). Harrison County, 2.5 miles northeast of Saucier, Tributary at McHenry Branch at Tuxachanie Trail, T4S-R11W-S29W, 4.XI.1990, T.L. Schiefer (16 larvae and 6 pupae); 2.5 miles northeast of Saucier, Tributary at McHenry Branch at Tuxachanie Trail, T4S-R11W-S29W, 10.III.1991, T.L. Schiefer (18 larvae); 2.5 miles northeast of Saucier, West Creek, Tuxachanie Trail, T4W-R11W-S30SE, 10.III.1991, T.L. Schiefer (43 larvae); 2.5 miles northeast of Saucier, McHenry Branch, Tuxachanie Trail, T4S-R11W-S29SW, 10.III.1991, T.L.Schiefer (7 larvae). Jackson County, 2.75 miles northeast of Vancleave, Mounger Creek, 11.III.1991, T.L. Schiefer (44 larvae and 2 pupae). George County, 4.5 miles north-northwest of Lucedale, Skinner Creek, T1S-R6W-S6NE, 12.III.1991, T.L. Schiefer (113 larvae). Stone County, 9 miles west of Wiggins, Sweetbay Bog Natural Area, 2nd tributary of Kirby Creek, T2S-R13W-S34SW, 13.III.1991, T.L. Schiefer (125 larvae, 11 pupae, and 5 exuvia). Forrest County, Paul B. Johnson State Park in an unnamed creek flowing into Geiger

Lake, T2N-R13W-S12, 14.III.1991, T.L. Schiefer (41 larvae). Franklin County, Homochito Ntational Forest, Middleton Creek, T5N-R4E-S31S, 6.II.1992, T.L. Schiefer (unknown number of larvae); Homochito National Forest, tributary to McGehee Creek, T1S-R6W-S6NE, 12.III.1992, T.L. Schiefer (unknown number of larvae). Pearl River County, natural spring that runs in to Perkins Reed Brook, near Kermis Myrick Road, 30.802ºN, 89.466ºW, 18.IV.2002, D.W. Boyd (115 larvae, 6 pupae, and 1 exuvia); natural spring that runs in to Perkins Reed Brook, near Kermis Myrick Road, 30.802ºN, 89.466ºW, 22.XI.2002, D.W. Boyd (6 larvae and 1 exuvia).

Number of specimens examined – 560

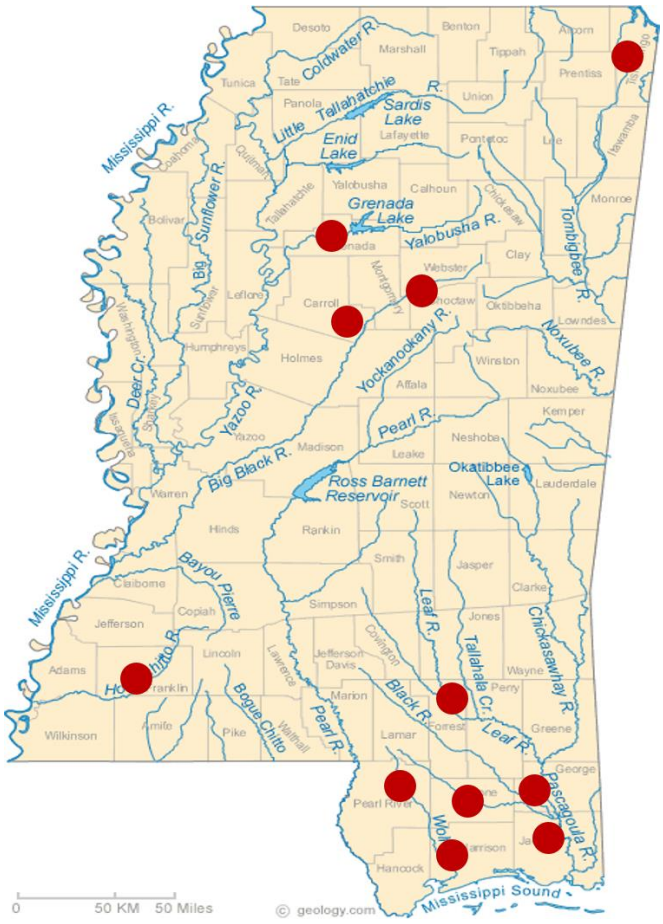

Figure 3.27 *Simulium ubiquitum* Adler, Currie & Wood distribution map

Mississippi counties where *S. ubiquitum* have been collected or documented. Map credit: Geology.com.

***Simulium vandalicum* Dyar & Shannon**

The only specimens of *Simulium vandalicum* were collected in Wilkinson County during March and confirmed by Adler et al. (2004).

Material examined – Mississippi: Wilkinson County, Clark Creek Natural Area, Clark Creek, 11.III.1989, T.L. Schiefer (unknown number and stage of specimens).

Number of specimens examined – not specified.

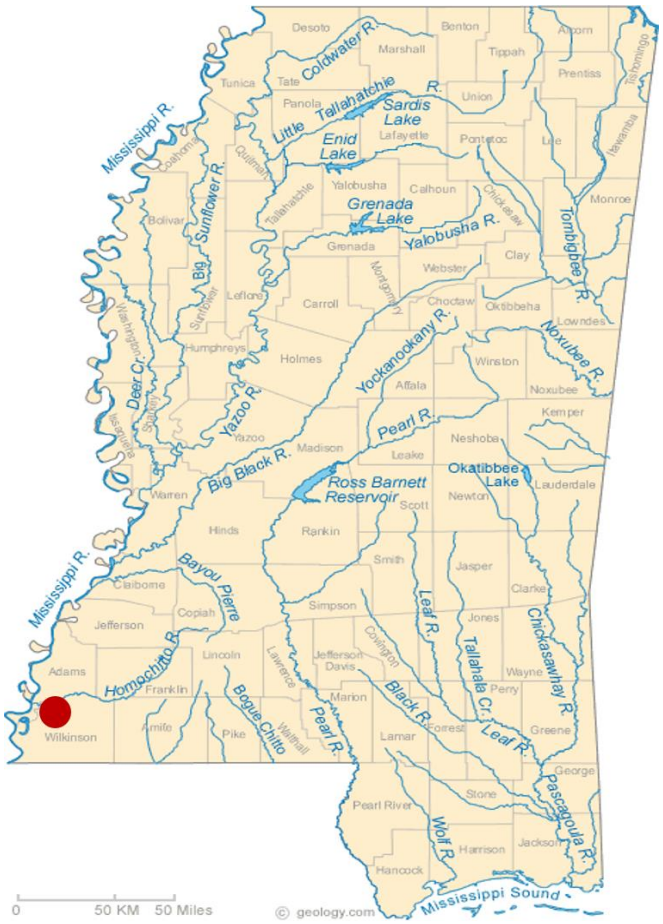

Figure 3.28 *Simulium vandalicum* Dyar & Shannon distribution map

Mississippi counties where *S. vandalicum* have been collected or documented. Map credit: Geology.com.

***Simulium venustum* Say**

This is one of the most abundant black fly species in North America. In Mississippi, *Simulium venstum* has been collected mostly from northern parts of the state (Tishomingo and Oktibbeha counties). Larvae and pupae have been found in January through April with a peak in February. Adults have only been collected in April.

Material examined – Mississippi: Oktibbeha County, Starkville, Sand Creek, 22.III.1930, William J. Posey (1 larva); Mississippi State, 24.III.1976, L. Thead (3 larvae); Dorman Lake in a

hardwood forest, malaise trap, 3-5.IV.1981, R.L. Brown (5 adults); Starkville in a creek, 31.III.1982, S. Winters (2 larvae). Tishomingo County, Tishomingo State Park, 11.IV.1986, J. MacDonald (2 adults); unspecified location, 12.IV.1986, unspecified collector (1 larva). Oktibbeha County, Sand Creek at MSU North Farms, 31.I.1987, T.L. Schiefer (30 larvae). Tishomingo County, Tishomingo State Park, Rock Quarry Branch, 7.III.1987, T.L. Schiefer (3 larvae); Tishomingo State Park, Bear Creek, 7.III.1987, T.L. Schiefer (2 larvae). Lowndes County, Tributary of Luxapilla Creek below beaver dam, 15.III.1987, T.L. Schiefer (28 larvae and 2 pupae). Grenada County, Tributary of Clark Creek, 15.IV.1987, T.L. Schiefer (6 larvae and 2 pupae). Lowndes County, Ellis Creek at intersection with East Nashville Ferry Road, 28.II.1988, T.L. Schiefer (63 larvae and 1 pupa). Choctaw County, sweep net at Choctaw Lake, 13.IV.1995, D.M. Pollock (1 larva).

Number of specimens examined – 152

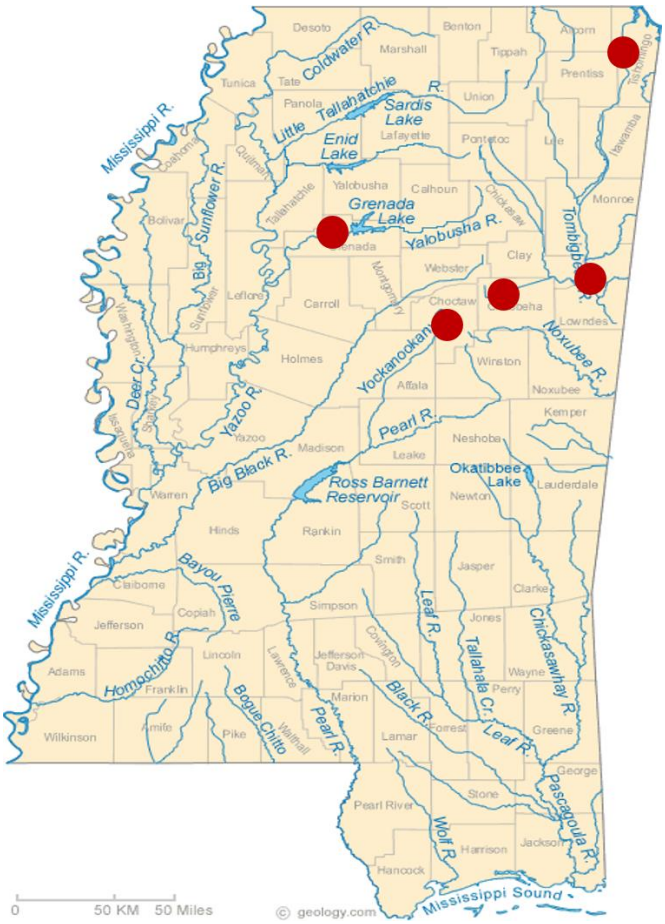

Figure 3.29 *Simulium venustum* Say distribution map

Mississippi counties where *S. venustum* have been collected or documented. Map credit: Geology.com.

***Simulium verecundum* (Stone and Jamnback)**

Adults of this species have been found only in Winston County in north-central Mississippi, while larvae and pupae have been collected in Oktibbeha, Noxubee, and Tishomingo counties. Larvae and pupae were recorded January through March, with adults only being collected in April.

Material examined – Mississippi: unspecified county, unspecified location, unspecified date, unspecified collector (1 larva). Oktibbeha County, Starkville, Sand Creek, 22.III.1930,

William J. Posey (1 larva); Starkville in a creek, 31.III.1982, S. Winters (1 larva). Tishomingo County, unspecified location, 12.IV.1986, unspecified collector (1 larva). Noxubee County, Noxubee National Wildlife Refuge, Bluff Lake outlet, 31.I.1987, T.L. Schifer (4 larvae and 1 pupa). Winston County, Tombigbee National Forest (33º11'50"N, 89º03'20"W), 5.IV.1999, J.A. MacGowan (1 adult); Tombigbee National Forest (33º15'18"N, 89º03'20"W), 12.IV.1999, D.M. Pollock (1 adult).

Number of specimens examined – 11

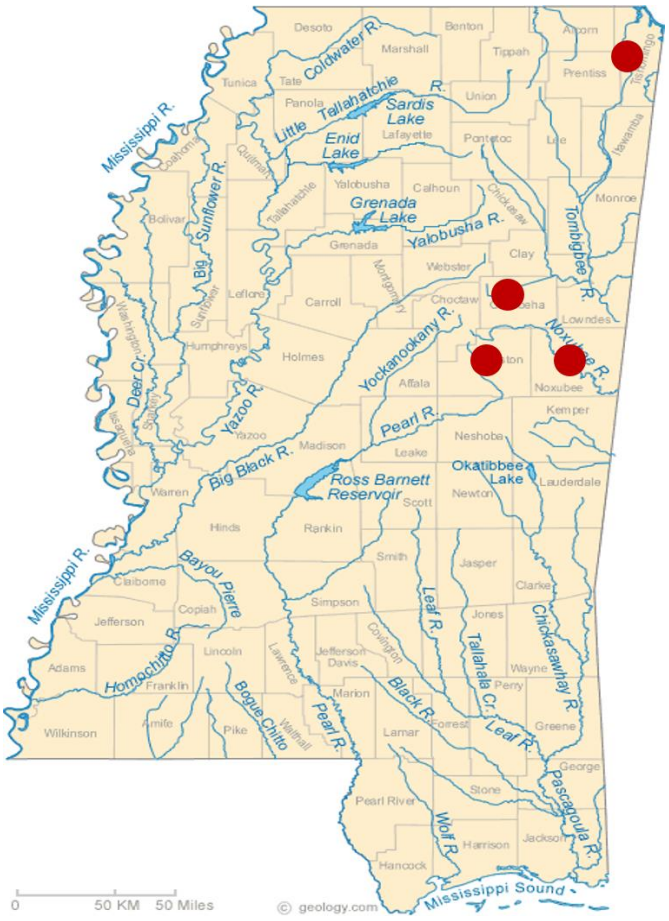

Figure 3.30 *Simulium verecundum* (Stone and Jamnback) distribution map

Mississippi counties where *S. verecundum* have been collected or documented. Map credit: Geology.com

Acknowledgements

We are especially grateful to Dr. Peter Adler (Clemson University) for confirming the identification of the black flies discussed in this paper.

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CHAPTER IV

THE PRIMARY PEST BLACK FLY SPECIES OCCURRING IN MISSISSIPPI

Abstract

Although there are 27 reported species of black flies in Mississippi, little is known about which of these are the primary pest species occurring in the state. During the 1930's, Dr. George Bradley described one primary pest species, *Cnephia pecuarum*, although it is likely that some of his specimens were actually *S. meridionale* (Dr. Peter Adler, personal communication). The resurgence of black flies in the state over the past decade has prompted renewed interest in these blood-sucking pests. Therefore, the purpose of this study was to determine the primary pest species occurring in Mississippi, their seasonality, distribution, and possible relationship of activity versus various meteorological conditions. Ten collection sites were selected across Mississippi based on previous research from Dr. George Bradley, reports from local MS extension agents, and complaints by local veterinarians and concerned citizens. Black fly adults were collected by hand netting at each of the ten sites for two years. Upon each visit, data was recorded including date, time, temperature, humidity, sky conditions, and wind speed. A total of 350 adult black flies were collected, returned to the lab, and identified. Subsamples were sent to Dr. P.H. Adler (Clemson University) for confirmation. The two main species collected were *Simulium jenningsi* group (248 specimens) and *Simulium meridionale* (98 specimens). Three other species were rarely collected (4 specimens): *S. parmatum* and *S. tribulatum* and *S. johannseni*. The two main pest species (named above) were predominantly active from March

through June each year. *Simulium jenningsi* was found mostly in central, south, and eastern Mississippi, while *S. meridionale* was found mostly in the northern Delta region. Preliminary statistical analysis using Pearson's chi-square test (IBM, SPSS Software, version 23) and stepwise multiple regression showed that temperature and humidity were significantly associated with black fly activity for both *S. jenningsi* and *S. meridionale*. This study indicates that there are at least two main pest species of black flies occurring statewide in Mississippi and these should be the ones targeted for control efforts.

Introduction

The first known documented description of black flies (most likely *Cnephia pecuarum*) in the southern United States occurred in 1818 in Choctaw "Country," what is now Mississippi. A much larger, well-documented outbreak occurred in 1859 in Greenville, MS and Clarendon, Arkansas (Webster 1904, Atwood and Meisch 2004) due to the pestiferous black fly species, *Simulium meridionale*. Subsequent sporadic outbreaks occurred in Arkansas, Indiana, Louisiana, Mississippi, and Tennessee from 1874 – 1884, and especially during the spring of 1882, when wild deer were pushed out of swamps by black flies and almost exterminated in Louisiana between the Ouachita and the Mississippi Rivers. Physicians reportedly verified several human deaths in Louisiana and Arkansas (Atwood and Meisch 2004). A total of 3,200 head of cattle was lost in a single week in Franklin Parish, Louisiana (Webster 1904). While working on plantations, farmers lost hundreds of horses and mules before they could be removed to areas of protection. Cities like Memphis, Tennessee and Vicksburg, Mississippi had horses and mules attacked on streets and in their stables. Reportedly, it was almost impossible to run a horse cart during spring planting, beginning in March and April 1882 (Webster 1904, Atwood and Meisch 2004).

Starting in the late 1920s and early 1930s, many reports and complaints were received from Mississippi county extension agents, local veterinarians, physicians, and local farmers about "gnat" attacks. These pests were reported as *Cnephia pecuarum* and *Simulium meridionale*, which prompted Dr. George Bradley's extensive research on black flies in the South, focusing primarily in the Mississippi Delta. Reports of black flies apparently disappeared from the state, until 2008 – 2009, when the Mississippi State Department of Health (MSDH) and the Mississippi State University Extension Service began receiving complaints from the public about increased human biting incidents and backyard poultry deaths resulting from black flies. Since so little is known about current black fly activity in Mississippi and a lack of statewide expertise on these pests, this study was initiated. In particular, determining which black fly species are the primary pests in Mississippi, clarify their seasonality and geographic distribution, and attempt to identify meteorological factors affecting their activity.

Methods

Adult black flies were collected from January 1, 2015 through December 31, 2016 by hand netting from ten locations around Mississippi, each located by a river or creek. Selection of sites was based on historical reports of black fly problems and a survey of Mississippi State University County Extension Agents (Table 4.1 & Figure D.1). Collections (n=180) were made twice per month in the peak of black fly activity, February – July, and only once per month, August – January. Black flies were placed in 70% ethanol and returned to the lab for identification using published keys (Stone & Snoddy 1969, Adler et al. 2004); subsamples of each species were sent to Dr. Peter Adler (Clemson University) for confirmation. Voucher specimens of each species, confirmed by Dr. Peter Adler, are deposited in the Mississippi State University Entomological Museum.

Statistical Analysis

Data analysis was performed using SPSS Statistical Software (IBM SPSS Statistical Software, version 23, Armonk, NY) and R package version O.5.2 focused on black fly activity (dependent variable) in relation to temperature, sky conditions, humidity, and wind speed (independent variables). Field notes of these meteorological parameters were made upon each site visit; however, more precise data were subsequently obtained online from the nearest National Weather Service station for each site. For definition of sky condition, categories previously defined by Weather Underground were used (The Weather Company, wunderground.com). Initially, Pearson chi-square was used to evaluate statistical significance, if any, of the meteorological factors: temperature, humidity, sky conditions, and wind speed, in relation to the two main species collected (*S. jenningsi* and *S. meridionale*). Each variable, except for species, was categorized based on the frequency of collection: temperature (28.9ºF – 67.8ºF, low temperature; 67.9ºF – 82.0ºF, intermediate temperature; 82.1ºF – 96.8ºF, high temperature), humidity (30% - 40%, low humidity; 41% - 70%, intermediate humidity; 71% - 100%, high humidity), wind speed (0 – 10, low wind speed; 11 – 16, intermediate humidity; +17, high wind speed); and sky conditions (no data, clear, drizzle, hazy, light rain, mostly cloudy, overcast, partly cloudy, rain, scattered showers, thunderstorm, and missing data). A second analysis using R statistical software was performed. In this case, a stepwise multiple regression was done in the "Olsrr" package (Hebbali, 2018).

Figure 4.1 Collection site #10, Clarke County, MS, Buckatunna Creek

All collection sites were based on Dr. George Bradley's prior research, complaints from local county extension offices, and/or local veterinarians (Figure D.10). Photo credit: Jerome Goddard.

Figure 4.2 Collection site #4, Leflore County, MS, Tallahatchie River

This location is one of Dr. George Bradley's research sites from the 1930s. He documented this river as one of the breeding locations for adult black flies (Table 4.1, Figure D.10). Photo credit: Tina Nations.

Table 4.1 Collection sites for the two-year survey

Collecting Site	County	Nearby body of water
1) Near Lula, MS	Tunica	Mississippi River
2) Near Sledge, MS	Quitman	Coldwater River
3) Near Webb, MS	Tallahatchie	Tallahatchie River
4) Near Money, MS	Leflore	Tallahatchie River
5) Greenville, MS	Washington	Mississippi River
6) Vicksburg, MS	Warren	Mississippi River
7) Jackson, MS	Hinds	Pearl River
8) Near Mendenhall, MS	Simpson	Strong River
9) Near Seminary, MS	Covington	Okatoma Creek
10) Near Shubuta, MS	Clarke	Buckatunna Creek

All ten collection sites were based on previous reports of black fly outbreaks in Mississippi; starting with Dr. George Bradley's research to current 2009 attacks (see Figures D.4 – D.13).

Results and Discussion

A total of 350 black flies were collected during the two-year survey comprised of five species listed in the table below (Table 4.2).

Table 4.2 Five species collected in the two-year survey.

# Collected	Species
248 specimens	*Simulium jenningsi* group
98 specimens	*Simulium meridionale*
2 specimens	*Simulium parmatum*
1 specimen	*Simulium johannseni*
1 specimen	*Simulium tribulatum*

Five species were collected during the two-year survey.

The most commonly collected species was *Simulium jenningsi* group (248/350, 71%), followed closely by *S. meridionale* (98/350, 28.0%). Three other species were only rarely collected – *S. parmatum* (2/350, 0.57%), *S. johannseni* (1/350, 0.29%), and *S. tribulatum* (1/350, 0.29%). Geographically, *S. meridionale* was mostly collected from sites in northwest and central Mississippi. *Simulium jenningsi* group was collected primarily from sites in central and eastern, and one location in northwest Mississippi, essentially year-round. *Simulium parmatum* was only

collected in southeast Mississippi during February and March; *S. tribulatum* was only collected in Warren County during 2015; and *S. johannseni* was only collected in Covington County during 2016.

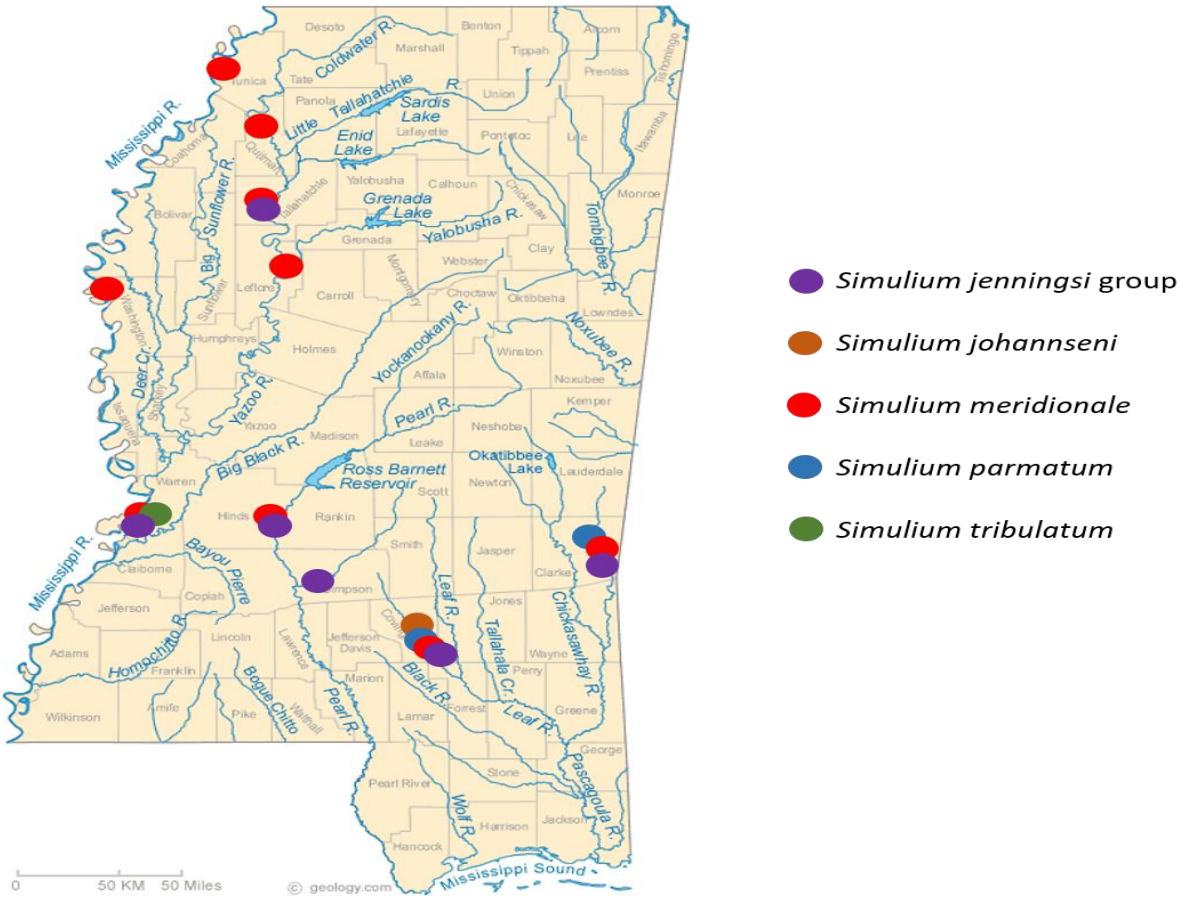

Figure 4.3 Distribution map of species collected in Mississippi

Distributions of adult black fly species collected during the project, 2015 – 2016.

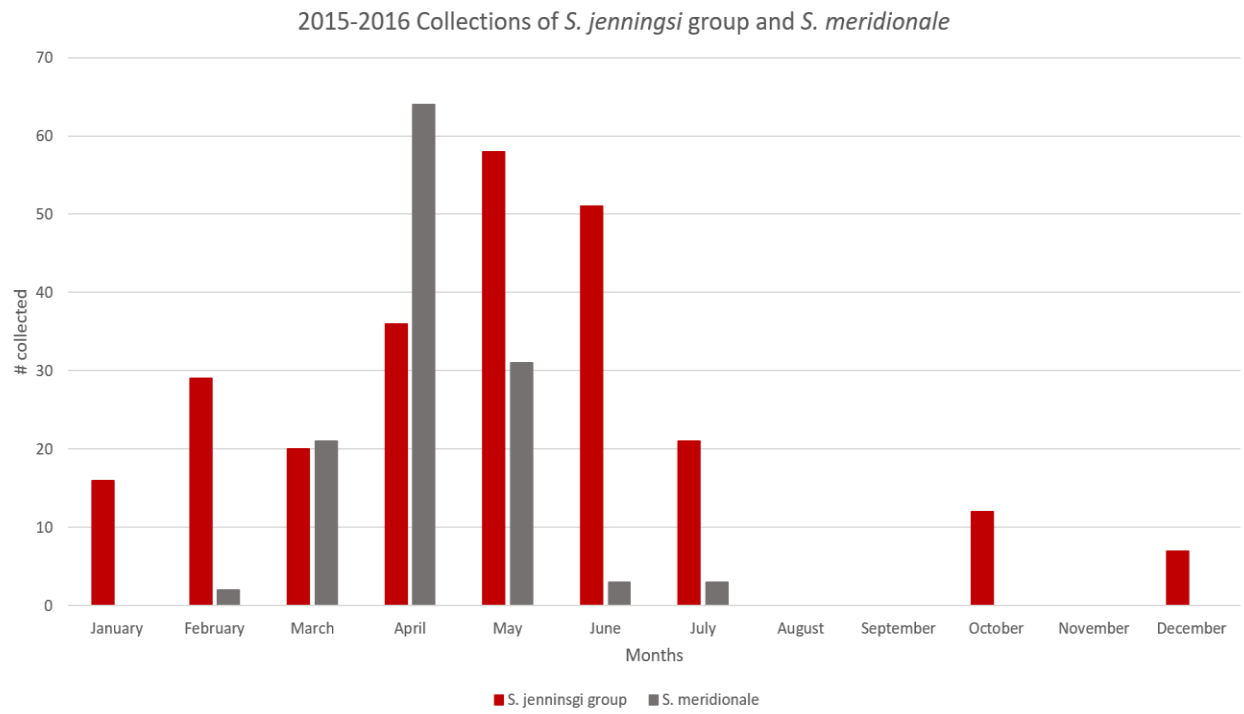

Figure 4.4 *Simulium jenningsi* group and *Simulium meridionale* seasonality for 2015 – 2016

This chart shows the seasonality for the two main species collected in this two year project, *S.jenningsi* group (27.78 ± 17.49) and *S. meridionale* (21.33 ± 23.79). The peak month for *S. jenningsi* group was in April, but for *S. meridionale*, it was May and June.

Analysis of Meteorological Parameters

A total of 363 trips were made to the ten collection sites, but only 361 could be analyzed (collection site #6 was not collected in 2015 due to a medical situation and collection site #7 has missing weather data March 2016) (Table 4.1). As for meteorological factors affecting black fly activity, previous research has shown that temperature has an effect of black fly emergence and activity (Bradley 1932, Colbo and Porter 1981, Lake and Burger 1983, McCreadie and Colbo 1991, Adler et al. 2017). Therefore, meteorological factors (humidity, sky conditions, temperature, and wind speed) were analyzed to determine, if any, effects on black fly activity. Due to the large numbers of "zeros" in the data set (days where nothing was collected), these two

statistical analyses should be considered preliminary. Further analysis is currently underway. Evaluation of four specific variables was performed using the chi-square (Pearson) test. The independent variables used was the two adult black fly species, *S. jenningsi* group and *S. meridionale*. The dependent variables were humidity, sky conditions, temperature, and wind speed. Analysis showed an association between temperature and *S. jenningsi* activity, but no other contributing factors (Table 4.3). Analysis of *S. meridionale* collection data revealed that temperature, humidity, and wind speed were significantly associated with activity (Table 4.3). In the second analysis using R the findings were similar. When analyzed for all sites combined where *S. jenningsi* group were collected, only temperature was significant ($p<0.0283$). Results for the all sites combined analysis for *S. meridionale* showed that only humidity was significant ($p<0.0479$). When analyzed by each location (site) for *S. jenningsi* group, humidity was significant for sites 8 and 10 ($p<0.0239$ and $p<0.011$ respectively), and for *S. meridionale*, humidity and sky conditions were significant for site 1 ($p<0.0290$).

Table 4.3 Pearson chi-square analysis for S. jenningsi group and S. meridionale

Species	Variables	df	*p* value (<0.05)
Simulium jenningsi group			
	Humidity	2	0.533
	Sky conditions	11	0.839
	Temperature	2	0.049
	Wind speed	2	0.102
Simulium meridionale			
	Humidity	2	0.003
	Sky conditions	11	0.219
	Temperature	2	0.013
	Wind speed	2	0.010

Pearson chi-square analysis of species and independent variables ($n=361$, $p<0.05$). Interactions of humidity x sky conditions x temperature x wind speed.

Conclusions

A total of 350 black flies were collected, returned to the lab, and identified. Subsamples were confirmed by Dr. P.H. Adler (Clemson University). The two main species I found were *Simulium jenningsi* group (248 specimens) and *Simulium meridionale* (98 specimens). Three other species were rarely collected (4 specimens): *Simulium parmatum*, *S. tribulatum*, and *S. johannseni*. Overall, the two main pest species (above) were predominantly active from April through June each year. Peak months of activity were May and June for *S. jenningsi* group and April and May for *Simulium meridionale*. *Simulium jenningsi* was found in central, south, and eastern Mississippi, while *Simulium meridionale* was found mostly in the northern Delta region. Analysis of meteorological parameters (temperature, humidity, wind speed, sky conditions) using chi-square analysis and stepwise multiple regression showed that temperature and humidity were common predictors of activity for both species. Other possible factors included wind speed and sky condition. These findings are not unexpected – most insects are susceptible to desiccation, so humidity is important and, of course, black flies are unable to fly during cold temperatures and high wind speeds. The role of sky conditions is not as clear but has been previously reported as a factor in black fly activity (Wolfe 1960, Alverson 1976, Martínez et al. 2009). This study confirms that there are at least two main pest species of black flies occurring in Mississippi; they are active primarily during spring and early; and temperature and humidity are the two main factors affecting their activity.

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CHAPTER V

THE MEDICAL, VETERINARY, AND ECONOMIC IMPACTS OF BLACK FLIES IN MISSISSIPPI

Abstract

Not much information is available concerning the economic impacts black flies have on humans and animals in Mississippi. The reemergence of black files in Mississippi has renewed interest in these pests to determine their economic impacts on humans and animals. For this chapter, the economic effects of black flies were utilized with a four-tiered approach: 1) a literature review search to compile recent reports concerning human health effects from black flies in Mississippi 2) a query of local mosquito personnel at the annual Mississippi Mosquito Vector Control Association meeting from 2015 – 2017 to find out if anyone had received complaints and/or sprayed for black flies in their local municipality 3) a request to the Mississippi State Department of Health for ICD-9 (International Classification of Diseases) data codes for outpatient hospital discharge data for patients in Mississippi diagnosed with insect bites/stings, both with and without infection. Data for the years 2010 – 11 were the only ones available. Lastly, 4) a survey (questionnaire) of back-yard poultry owners to assess the effects, if any, black flies had on their poultry flocks, livestock, and any other animals they owned.

Results of this four-tiered approach revealed four recent medical and popular press articles since 2011; one city (Greenville) reported aerial spraying for a black fly outbreak in June 2011 (see Appendix E1); Health Department ICD-9 data codes for the years 2010 – 11 revealed a

total of 13,760 outpatient discharge cases for bites/stings in counties across the state. Statistical analysis of biting incidence data from counties known to have black fly problems versus counties without black fly problems showed no significance or clear patterns. Lastly, the backyard poultry questionnaire yielded 33 responses out of 522 mailed. Poultry owners reported poultry and livestock attacks during known years of black fly outbreaks and also during known months of peak activity, with approximately 45 poultry reportedly killed by black flies and one "livestock" death.

Introduction

Black flies may cause a variety of medical, veterinary, and economic effects worldwide. Human and animal disease transmission (e.g., onchocerciasis and leucocytozoonosis) by black flies is well documented (Getting 1945, WHO 1971, Noblet 1975, Jones et al. 1977, Adler and McCreadie, 2009, Rohner 2000, Jones et al. 2014). Medical complications following black fly biting have also been reported (Stoke 1914, Borah et al. 2012, Goddard et al. 2018), and there are at least two human deaths attributed to black fly attacks (Atwood and Meisch 2004). Although, black flies have likely caused enormous losses of livestock since recorded history. It wasn't until 1927, in Yazoo City, Mississippi, that black flies were reported killing an estimated 100 farm animals. In 1928, in Charleston, MS, swarms of black flies killed an undocumented amount of livestock. Then in 1931, one of the worst reported outbreaks occurred in Mississippi and Arkansas with over 1,000 mules killed in Coahoma County, Mississippi and more than 500 mules killed in eastern Arkansas. These outbreaks triggered the USDA, Bureau of Entomology, to start investigating the importance of black flies to better understand their biology and possible control measures (Webster 1904, Atwood and Meisch 2004, Nations et al. 2015).

In Mississippi, black flies seemed to disappear from the state during the 1930s, with few or no reports of outbreaks until 2008 – 2009, when the Mississippi State Department of Health and the Mississippi State University Extension Service began receiving reports from the public about increased human biting incidents and backyard poultry deaths resulting from black flies. Outbreaks continued in the years 2011, 2012, 2014, and 2018. As a result, one limited study was performed to assess the impact of black flies on poultry (Jones et al. 2014). However, much is yet unknown about the current medical, veterinary, and economic impacts of black flies in Mississippi; therefore, this study was initiated to further document these impacts.

Methods

Search of Literature and Popular Press for Human Nuisance Biting by Black Flies

All information provided in blogs, newspapers, and databases concerning human health effects of black flies in Mississippi was surveyed to establish baseline data. For the literature search, we surveyed PubMed, Google Scholar, Science Direct, EBSCO Host, and the Armed Forces Pest Management Board's Literature Retrieval System using the terms or phrases, "Simuliidae," "Simulium," "Black Flies," "Black Fly," "Pests," "Black Flies in Mississippi," "Black Fly Outbreaks in Mississippi," "Black Fly Attacks in Mississippi," "Biting," "Nuisance," "Seasonality," "Biology," "Ecology," "Vectors," "Bite Reaction," "Black Fly Fever," "Recreational Activities," "Adverse Effects," "Medical Effects," "Health Effects," "Human Health Effects," "Livestock Health Effects," "Poultry Health Effects," "Veterinary Effects," "Human Pests," "Poultry Pests," "Livestock Pests," Hospital Discharge Data," "Insect Bites/Stings," "Outpatient Hospital Discharges," "Insect Bites/Stings with/without Infections," "Economic Impacts," "Economic Effects," "Epidemiology," "Control," "Treatments," "Control of Black Flies," "Treatments from Black Flies," "Pesticides," "Adulticides for Black Flies,"

"Insecticides," "Insecticides for Black Flies," "Prevention," "Prevention from Black Flies," and "Personal Protection."

Query of City and County Public Works Personnel

For two consecutive years (2015-16), attendees at the Mississippi Mosquito and Vector Control Association (MSMVCA) in Jackson, MS, were queried concerning black fly attacks or outbreaks in their local municipalities. If so, they were asked what surveillance and control measure(s) they used and approximate costs involved.

Human Clinical Data Search (ICD-9 Data)

With appropriate IRB approval, a request was made to the Mississippi State Department of Health (MSDH) as to outpatient hospital discharge data, and particularly, ICD-9 data codes for insect bites/stings, with and without infection, for the years 2009 through 2017 (Table 5.1) NOTE: these are *their* categories (bites/stings; with/without infection), not ours. Data were analyzed in a number of ways to try to ascertain if there were increased biting incidents (leading to hospital or clinic visits) in areas known to have black flies versus those without. Analysis included biting incidence per year, county, region, and comparisons of selected sites throughout the state.

Table 5.1 ICD-9 codes used for outpatient discharge(s) with superficial injuries with/without infection(s)

Code Definition	Location on the body (including/excluding)	Sub-Code	Sub-Code Definintion
910: Superficial injury of face, neck, and scalp, except eye(s)	Includes: cheek, ear, gum, lip, nose, throat Excludes: eye and adnexa	910.4	Insect bite, nonvenomous, without mention of infection

Table 5.1 (continued)

		910.5	Insect bite, nonvenomous, infected
911: Superficial injury to trunk	Includes: abdominal wall, anus, back, breast, buttock, chest wall, flank, groin, interscapular region, labium (major and minus), penis, perineum, scrotum, testis, vagina, vulva Excludes: hip and scapular region	911.4	Insect bite, nonvenomous, without mention of infection
		911.5	Insect bite, nonvenomous, infected
912: Superficial injury to shoulder and upper arm	Includes: axilla and scapular region Excludes: no other areas in this region	912.4	Insect bite, nonvenomous, without mention of infection
		912.5	Insect bite, nonvenomous bite
913: Superficial injury of elbow, forearm, and wrist	Includes: elbow, forearm, wrist Excludes: no other area in this region	913.4	Insect bite, nonvenomous, without mention of infection
		913.5	Insect bite, nonvenomous, infected
914: Superficial injury of hand(s) except finger(s) alone	Includes: hand(s) Excludes: no other area in this region	914.4	Insect bite, nonvenomous, without infection of infection
		914.5	Insect bite, nonvenomous, infected
915: Superficial injury of finger(s)	Includes: fingernail and thumb (nail) Excludes: no other area in this region	915.4	Insect bite, nonvenomous, without mention of infection
		915.5	Insect bite, nonvenomous, infected
916: Superficial injury of hip, thigh, leg, and ankle	Includes: hip, thigh, leg, ankle Excludes: no other area in this region	916.4	Insect bite, nonvenomous, without infection
		916.5	Insect bite, nonvenomous, infected

Table 5.1 (continued)

917: Superficial injury of foot and toe(s)	Includes: foot, toe(s) Excludes: no other area in this region	917.4	Insect bite, nonvenomous, without mention of infection
		917.5	Insect bite, nonvenomous, infected
919: Superficial injury of other, multiple, and unspecified site(s)	Includes: other areas of the body Excludes: no other areas	919.4	Insect bite, nonvenomous, without mention of infection
		919.5	Insect bite, nonvenomous, infected

ICD-9 collected data is for the years, 2010-11. This data excludes 918(.4)(.5) [the area of the eye(s) and adnexa].

Survey of Backyard Poultry Owners

A survey (questionnaire) was developed for backyard poultry owners in Mississippi to assess how much – and to what extent – the Mississippi poultry industry may be affected by black fly infestations. This questionnaire was prepared with assistance from Dr. Kristine T. Edwards, a local veterinarian, and included questions about 1) domestic animals on hand, 2) outbreak years, 3) attacks/deaths of birds or other livestock, 4) protection methods and costs, 5) treatments and costs, and 6) veterinary fees, if any. A total of 512 questionnaires were mailed to a list of backyard poultry owners developed by personnel at the Mississippi State University Poultry Diagnostics Lab in Pearl, MS.

Results and Discussion

Search of Literature and Popular Press for Human Nuisance Biting

Literature and popular press search revealed three online articles and papers referencing black fly nuisance biting in Mississippi; also, a fourth paper was published showing sensitivity to the reactions of a black fly bite in 2018. In June 2011, The Natchez Democrat published an article referencing the attack of black flies in the local community, Natchez, MS, and detailed several preventive measures that could be taken to protect the public. The article also showed a photograph of a young girl covering her eyes to protect herself from being attacked by black flies (Zema 2011).

During 2013, the MS Gunowners Blog posted a question asking if anyone else was having problems with black flies and could anyone recommend possible control measures for this pest (Anonymouns 2013). Also, in December 2013, a physician with the University of Mississippi School of Medicine, Department of Family Medicine, published an article about the clinical reaction of an older patient who was bitten by a black fly. He diagnosed sequelae from the black fly bite as a systemic reaction due to sensitivity or toxicity from the attack (Chen 2013.

Lastly, during the last week of March through the middle of April 2018, multiple reports of black fly outbreaks were reported in the tri-county Jackson, MS area and Natchez, MS. Local entomologists from the Mississippi State Department of Health (MSDH) were sent out to investigate this outbreak. The first complaint was located 0.55 miles from the Jackson Futbol Club (Jackson Country Club) and 1.68 miles from McLeod Elementary School (Figure 5.1). During this investigation, the Mississippi State Department of Health Medical Entomologist, Dr. Wendy Varnado, was attacked and severely bitten by these nuisance pests. The reaction to her

black fly bites lasted for two days causing intense itching and skin lesions and a clinical description of her case was published (Goddard et al. 2018).

Figure 5.1 Distance map depicting site of 2018 black fly outbreak

This map shows the distance from the Pearl River to Jackson Futbol Club (Jackson Country Club) and McLeod Elementary School in Jackson, MS. This location site was also a collection site in this project, 2015-16 (Collection site #7). Map credit: Google Maps (2018).

Query of City and County Public Works Personnel

During the Mississippi Mosquito Vector Control Association (MSMVCA) annual meeting, the local public works personnel were asked if they had any complaints or reports of black fly outbreaks or attacks during the years 2015-16. If they did, we asked what control measures were utilized to reduce effects of the black flies. The only municipality to respond was the City of Greenville which reported an outbreak in 2011. The local pest control operator, Vector Disease Control Incorported (VDCI), that handles their seasonal mosquito control,

reported that they conducted an aerial adulticide spray for the city on June 2, 2011 due to black flies. The City of Greenville had so many complaints from the local community that an immediate response was needed. Vector Disease Control Incorporated sprayed the city limits of Greenville with the adulticide, Dibrom, costing the City of Greenville $10,500 (Figure 5.2).

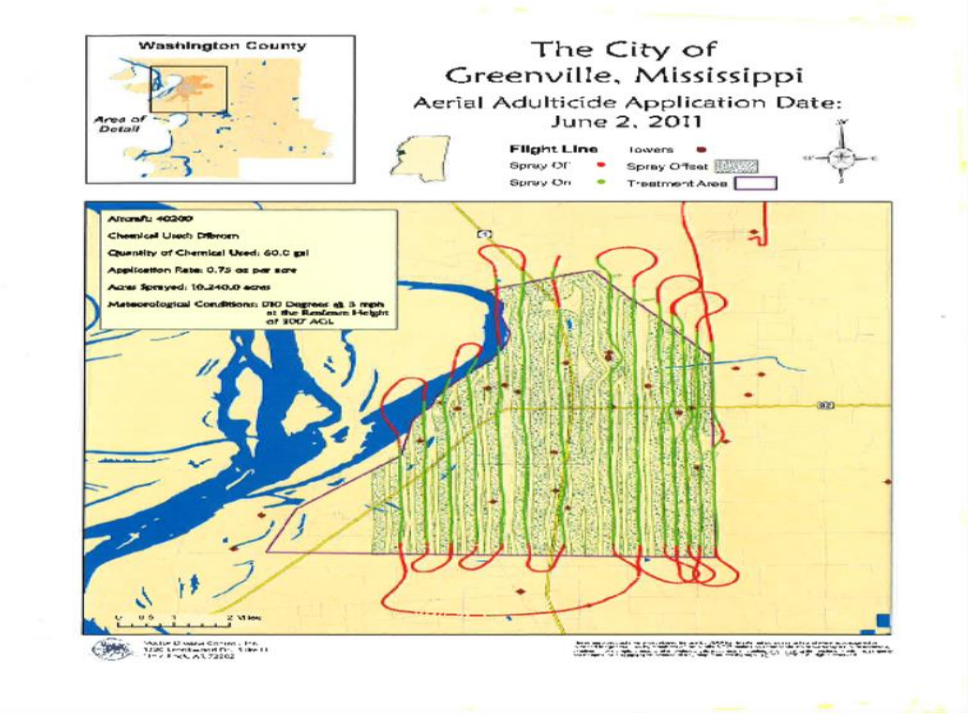

Figure 5.2 Aerial spray map of the City of Greenville

City of Greenville had to have their local pest control operator, VDCI, do an aerial adulticide treatment, Dibrom, for the local community due to a black fly outbreak. This aerial spraying cost the City of Greenville $10,500. Photo credit: Kris New with VDCI, June 2011.

Human Clinical Data Search (ICD9 Data)

ICD9 statewide hospital and clinic outpatient data for insect bites/stings with or without infection for the years 2009 – 2017 was requested, but only accessible data available was from the years 2010 – 11. Counties were separated and totaled for the two years which yielded 13,160 patients. Outpatient totals per county were calculated and divided by the 2010 U.S. Census Totals per county. The total was then multiplied by 10,000 to get the mean biting incidence per

county. These data were analyzed with SPSS Statistical Data Software (version 23, IBM, New York, NY). Three categories were created and analyzed: 1) the ten collection sites (known to have abundant black flies) versus 72 remaining Mississippi counties, 2) delta versus non-delta Mississippi counties, and 3) north, central, and south regions in Mississippi. Statistical analysis was used to determine if there was any significant difference in biting incidence in outbreak counties and non-outbreak counties, delta and non-delta counties, and the three regions in Mississippi. An independent t-test was used to analyze the categories: 1) the ten collection sites versus 72 remaining counties and 2) delta and non-delta counties; ANOVA was used to analyze biting incidence among north, central, and south regions of Mississippi. An independent t-test, using 95% CI ($p<0.05$), showed no significant differences between our ten collection sites versus the 72 remaining counties and delta versus non-delta counties. The *p*-value for our ten collection sites and the 72 remaining Mississippi counties was 0.801 with a standard error difference of 7.823 for equal variances assumed; not assumed equal variances has a standard error difference of 8.268. The *p*-value for delta versus non-delta Mississippi counties was 0.157, equal variances assumed has a standard error difference of 5.485, and equal variances not assumed has a standard error difference of 6.301. ANOVA results between north, central, and south regions in Mississippi showed no significance within groups. Post Hoc test results using Tukey HSD, Bonferroni, and Dunnett T3 yielded a similar result. Final significance had a *p*-value of 0.090 within these subsets.

Table 5.2 Independent t-test results: ten collection sites versus remaining MS counties

	t	df	*p*-value (95% CI)	Std. Error Difference	Lower	Upper
Equal variances assumed	0.055	80	0.956	7.823	-15.135	16.002
Equal variances not assumed	0.052	11.273	0.959	8.268	-17.711	18.579

Biting incidence in ten collection sites versus the 72 remaining MS counties were analyzed using an independent t-test to determine any significance between the groups. There was no significant difference between biting incidence our ten collection sites and the remaining Mississippi counties.

Table 5.3 Independent t-test results: delta versus non-delta MS counties

	t	df	*p*-value (95% CI)	Std. Error Difference	Lower	Upper
Equal variances assumed	-0.691	80	0.492	5.485	-14.706	7.126
Equal variances not assumed	-0.601	36.031	0.551	6.301	-16.569	8.989

Biting incidence in delta versus non-delta MS counties were analyzed using independent t-test to determine any significance between groups. There was no significant difference between biting incidence in delta and non-delta counties in Mississippi.

Table 5.4 ANOVA results: north, central, and south MS regions

			Std. Error	*p*-value (95% CI)	Lower	Upper
Tukey HSD						
	North	Central	6.022	0.285	-5.21	23.56
		South	6.022	0.079	-1.19	27.58
	Central	North	6.022	0.285	-23.56	5.21
		South	6.521	0.812	-11.56	19.60
	South	North	6.022	0.079	-27.58	1.19
		Central	6.521	0.812	-19.60	11.56
Bonferroni						
	North	Central	6.022	0.395	-5.55	23.91
		South	6.022	0.094	-1.53	27.93
	Central	North	6.022	0.395	-23.91	5.55
		South	6.521	1.000	-11.93	19.97
	South	North	6.022	0.094	-27.93	1.53
		Central	6.521	1.000	-19.97	11.93
Dunnett T3						
	North	Central	6.098	0.356	-5.82	24.17
		South	5.921	0.087	-1.38	27.77
	Central	North	6.098	0.356	-24.17	5.82
		South	4.911	0.798	-8.13	16.17
	South	North	5.921	0.087	-27.77	1.38
		Central	4.911	0.798	-16.17	8.13

All three regions in MS were analyzed using ANOVA to determine if there was any significance between groups. There was no significant difference in biting incidence between the tree regions in Mississippi.

Survey of Backyard Poultry Owners

Unfortunately, responses from the backyard poultry owners to the online survey were minimal. The survey was offered online through Survey Monkey and involved 21 questions concerning: types of animals, seasonality of attacks and/or outbreaks, mortality of animals, protection, control measures used, veterinary cost, insurance reimbursement, and money spent on protection and control measures used, if any. There were 512 survey cards mailed out backyard poultry owners in Mississippi. Of the 512 questionnaires, 18.2% (93 returned: (93/512) * 100 =

18.2%) of the survey cards were returned. And only 6.4% (33 responses: (33/512) * 100 = 6.4%) responses were filled out using Survey Monkey. Individual calls to property owners only resulted in three of the 33 responses. Also, we allowed for overlap of some responses (i.e. types of animals, months and years of attack/outbreak, animals affected, deaths of animals) to gather as much information as possible. With this limited information, we were able to conclude 51.5% (17/33*100 = 51.5%) of animals affected were backyard poultry (including chickens, quail, turkeys, guineas). Most of the respondents reported severe intermittent attacks (1 out of 3 years) to animals, 45.4% (15/33*100 = 45.4%); next was severe attacks every year (at least 10 black flies per animal), 18.1% (6/33*100 = 18.1%); and lastly, 12.1% (4/33* 100 = 12.1%) reported no problems at all. The reported months of attacks, outbreaks, and/or deaths were May – June, 57.5% (19/33*100 = 57.5%) and secondly, 42% (14/33*100 = 42.4%) reported March – April as severe months (Figure 5.3). Respondents were also asked "what years did animal attacks or deaths occur from black flies," which yielded these years: 1) 2011 and 2014 at 36.3% (12/33*100 = 36.3%), 2) 2009 and 2012 at 30.3% (10/33*100 = 30.3%), 3) 2015 and 2016 at 18.1% (6/33*100 = 18.1%) (Figure 5.4). Backyard poultry owners reported 45 (98%) poultry died from black fly attacks (chickens, turkeys, guineas, quail) and 1 "livestock" (2%).

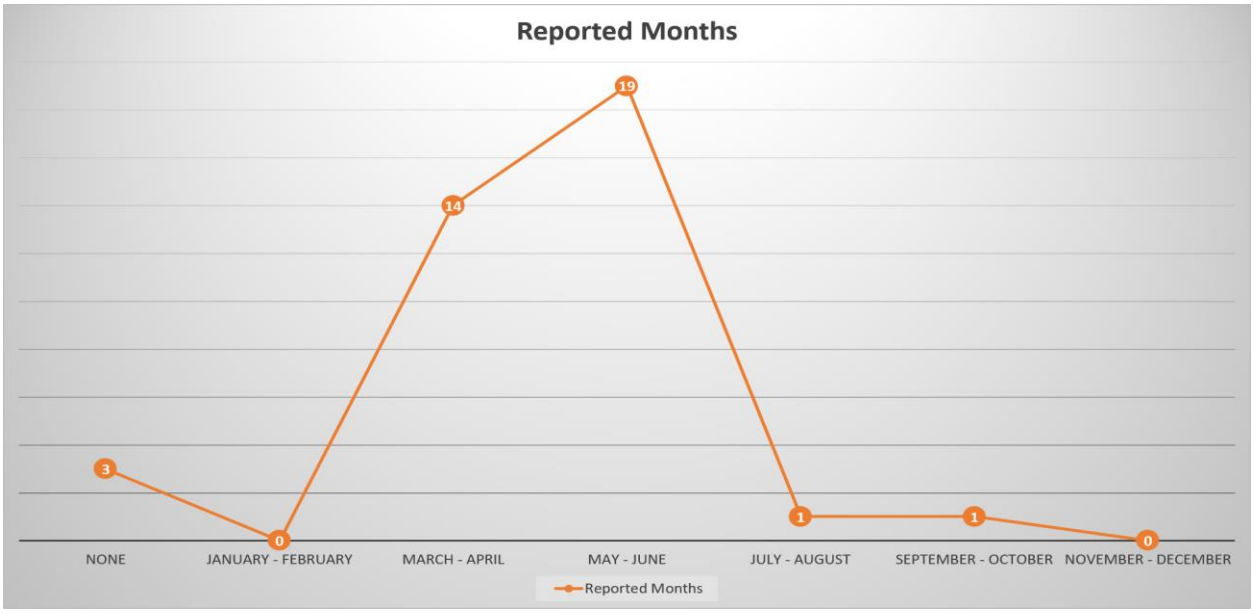

Figure 5.3 Reported months of attacks and/or outbreaks

The questionnaire asked the respondents "What month of the year did animal attacks or deaths occur from black flies?" This chart agrees with known months of attacks or outbreaks.

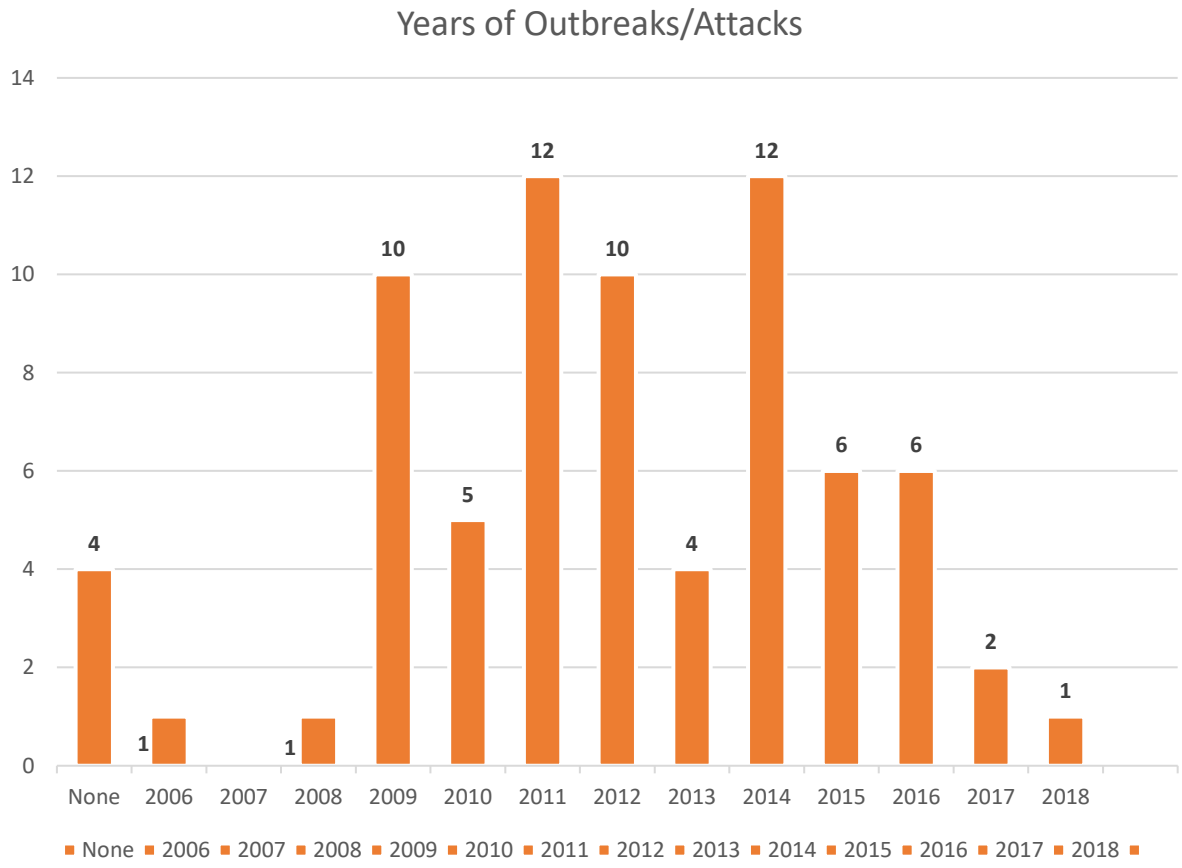

Figure 5.4 Reported years of outbreaks and/or attacks

The questionnaire asked respondents "Please check the years animal attacks or deaths occurred from black flies?" Most outbreaks, attacks, and/or deaths occurred in 2009, 2011, 2012, and 2014. Supporting known outbreaks and attacks in Mississippi.

When respondents were asked how much money was spent on pesticides, treatments, or repellents, 36.3% (12/33*100 = 36.3%) stated no treatments, pesticides, or repellents were used. And 30.3% (10/33*100 = 30.3%) stated they spent $51 - $100 on pesticides, repellents, and treatments for their backyard poultry, livestock, or other animals.

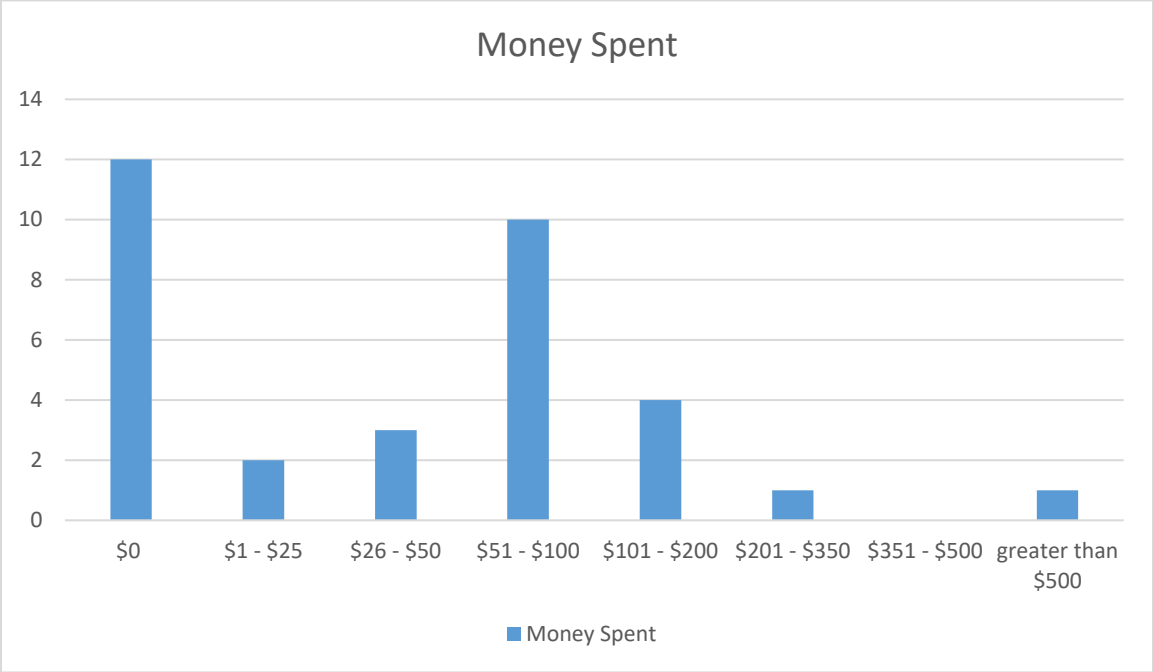

Figure 5.5 Reported money spent on pesticides, repellents, and treatments

Respondents were asked "If products used, approximately how much money was spent total?". Most reported using no protection for their animals and 30.3% reported spending $51 - $100 on control measures.

Conclusions

The literature search for reports of recent black fly outbreaks revealed recent published articles in 2009 and 2011 demonstrating black flies are currently nuisance pests in Mississippi and can affect outdoor activities. In city and county municipalities, very few pest control personnel reported using any control measures for black fly reduction during peak season(s). Also, statistical analysis of hospital discharge data showed there was no significance between insect biting incidence in counties with and without black fly outbreaks and/or attacks. Mississippi backyard poultry flocks (including chickens, turkeys, quail, guineas) are being affected by black flies – even causing deaths. Results from this questionnaire supported previous findings of known black fly outbreaks on backyard poultry in Mississippi (Jones et al. 2014).

Interestingly, respondents reported using little or no control measures to protect their animals from these nuisance pests.

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CHAPTER VI

OVERALL RESULTS AND DISCUSSION

The objectives for this project were met, and two chapters have already been published in scientific journals. After the literature search, there is now a better understanding of black fly outbreaks and attacks on humans and other animals in Mississippi. Historical research on the ecology, biology, seasonality, distribution, economic impacts, and control measures for the southern buffalo gnat, *Cnephia pecuarum*, in Mississippi have been re-typed, photos and figures scanned, and digital files stored on a server at the MEM, Mississippi State University for public awareness and education. A compiled annotated list of black fly species occurring in Mississippi (based on 14,471 specimens) was published, which revealed more species than originally recorded. Twenty-seven species were documented; as well as four new state records. Distribution of all 27 species is now better detailed, both seasonally and geographically.

In the two-year systematic collections at 10 sites around the state, a total of 350 black flies were collected. The two main species collected in Mississippi were *Simulium jenningsi* group (248 specimens) and *Simulium meridionale* (98 specimens). Three other species were also collected (4 specimens): *Simulium parmatum*, *S. tribulatum*, and *S. johannseni*. Overall, the two main pest species (above) were predominantly active from March through June each year. Peak months of activity were April and May for *S. meridionale* and May and June for *Simulium jenningsi* group. *Simulium jenningsi* was found in central, south, and eastern Mississippi, while *Simulium meridionale* was found mostly in the northern Delta region. Preliminary analysis of

meteorological parameters (temperature, humidity, wind speed, sky conditions) show that temperature and humidity are common predictors of activity for both species. Other possible factors include wind speed and sky conditions. Therefore, these two primary pest species are ones of public health importance and should be the targets for control.

The literature search for reports (media and science) of black fly outbreaks revealed recent published articles in 2009 and 2011 demonstrating that black flies are indeed currently nuisance pests in Mississippi and can affect outdoor activities. In city and county municipalities, very few pest control personnel reported using any control measures for black fly reduction during peak season(s), although one municipality did. Also, statistical analysis of hospital discharge data showed no significance between insect biting incidence in counties with and without black fly outbreaks and/or attacks. This was not surprising in light of the fact that we don't even know for sure if these reported "bites" were from black flies (or any other insect, for that matter). Results from a questionnaire to Mississippi backyard poultry owners (including chickens, turkeys, quail, guineas) revealed that poultry are being affected by black flies – sometimes even causing deaths. These results also support previous findings of known black fly seasonality in Mississippi. Interestingly, respondents used little or no control measures to protect their animals from these nuisance pests.

In conclusion, black flies are important medical and veterinary pests in the State of Mississippi. Contrary to what was once thought, the pests have not disappeared, but are, in fact, increasing in prevalence and frequency. Both humans and animals need protection from these pests and specific personal protection and control measures need to be developed.

APPENDIX A

SUPPLEMENTAL MATERIAL FOR CHAPTER I

Figure A.1 Black fly eggs

Black fly eggs, *Simulium vittatum* complex Zetterstedt, on vegetation. Females will lay their eggs on the surface water or local vegetation in well aerated, fast flowing rivers or streams. Estimated size of eggs, 0.20 – 0.50 mm. Photo credit and used with permission: @Dwight Kuhn.

Figure A.2 Black fly larvae

Shown here is *Simulium vittatum* complex Zetterstedt larvae floating in the water. Larvae attach to local rocks and other debris with a caudal sucker and "dangle" by a silk thread in the water. Estimated size of larvae, 5 – 15mm. Photo credit and used with permission: @Dwight Kuhn.

Figure A.3 Black fly pupae

Black fly larvae pupating in a cocoon. They remain attached to local vegetation until the adults emerge. Shown here is *Simulium vittatum* complex Zetterstedt. Estimated size of pupae, 5 – 15mm. Photo credit and used with permission: @Dwight Kuhn.

Figure A.4 Emergence of a black fly

Simulium vittatum complex Zetterstedt emerging from its cocooning period and rising to the water's surface. Estimated size of adult(s), 5 – 15mm. Photo credit and used with permission: @Dwight Kuhn.

Figure A.5 Adult black fly species

The species shown in this picture, *Simulium venustum* Say, has caused severe outbreaks in the northeastern part of the United States. Although previously collected by entomologists and Adler et al. (2004) in Mississippi, no adults were collected in this study. Estimated size of adult(s), 5 – 15mm. Photo credit and used with permission: @Dwight Kuhn.

Figure A.6 Adult black fly

Another pest species in the United States, *Stegopterna mutata* complex (Malloch). None were collected in this study but were collected by Adler et al. (2004) from Mississippi. Estimated size of adult(s), 5 – 15mm. Photo credit and used with permission: @Dwight Kuhn.

APPENDIX B

SUPPLEMENTAL MATERIAL FOR CHAPTER II

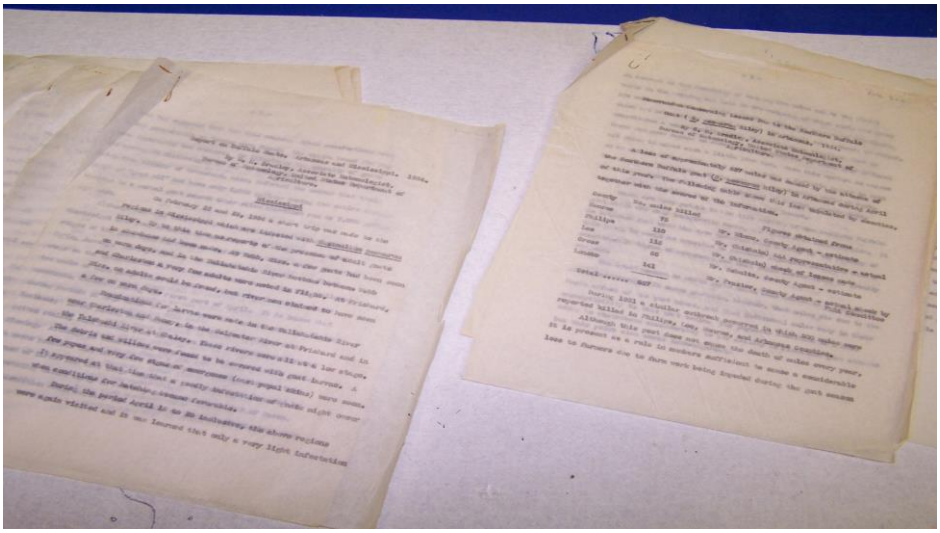


Figure B.1 Unpublished papers by Dr. George H. Bradley

Six original Dr. George H. Bradley papers were retyped and published in November 2015 in the journal, *Midsouth Entomologist* (see Chapter II), on the ecology, biology, seasonality, distribution, treatments, and control measures for black flies in the delta. Photo credit: Tina Nations.

Figure B.2 Smoke fires protecting cattle from black flies

Dr. George Bradley's research showed smoke fires and plumes protected livestock from black fly attacks. Photo credit: Dr. George Bradley, near Charleston, MS, 1932 (USDA).

Figure B.3 Farmer using a “smoker” on his plow to protect mule from black flies

Dr. Bradley studied protection measures that farmers used to protect their livestock from black fly attacks and/or outbreaks. Many would attach smoke plumes to plows and wagons. Photo credit: Dr. George Bradley, March 1932 (USDA).

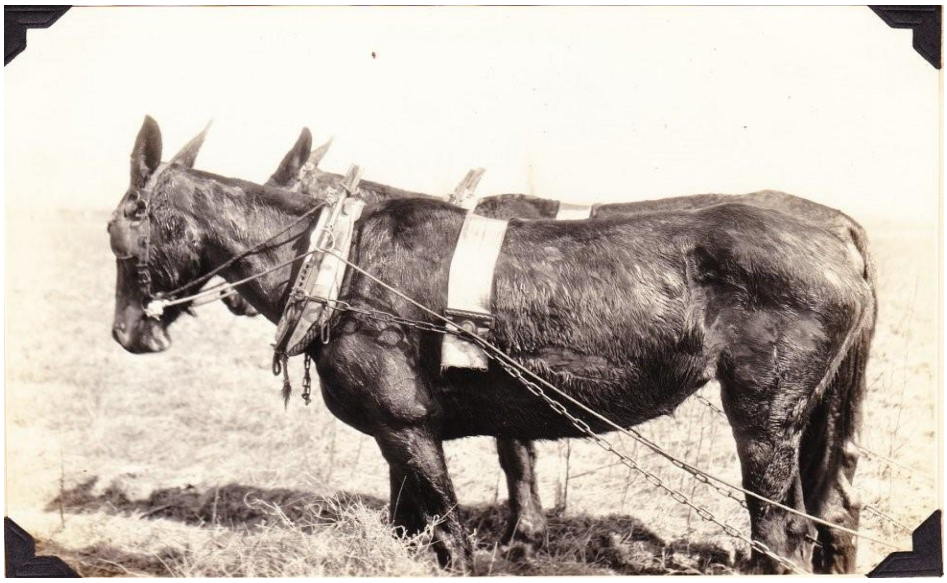


Figure B.4 Greased and tarred mules

Farmers would use grease and/or tar as another control measure to protect their livestock from black flies, which often burned or “skinned” the hides of mules. Photo credit: Dr. George Bradley, 1932 (USDA).

Figure B.5 Children carrying a smoke plume to go fishing

Adults and children carried smoke plumes while outdoors during black fly seasonal attacks and/or outbreaks to protect themselves from black flies. Photo credit: George Bradley, near Charlestion, MS, 1932 (USDA).

Figure B.6 Black flies, or buffalo gnats, on the udders of cow

Black flies were documented on the udders or teats of cattle. Also seen here, black flies on the legs and lower abdominal area of the cow. Photo credit: George Bradley, in Webb, MS, 1932 (USDA).

Figure B.7 Closer view of black flies, or buffalo gnats, on the udders of cow

Udders of this cow are infested with black flies. Photo credit: George Bradley, in Webb, MS, 1932 (USDA).

Figure B.8 Chickens eating black flies, or “gnats”, off cows

Another control measure used were chickens. They would pick and eat the nuisance pests off the cows. Photo credit: George Bradley, 1932 (USDA).

Figure B.9 Dried pupae and pupal skins on a tree branch pulled from the Tallahatchie River

Documentation of breeding habitats in Dr. George Bradley's research. This is one of his many photos recording massive amounts of larvae, pupae, and pupal skins of black flies in the Delta region. Photo credit: George Bradley, near Charleston, MS, 1932 (USDA).

Figure B.10 Dried pupal skins on a limb pulled from the Tallahatchie River

Pupal skins on a tree limb near Charleston, MS, in the Tallahatchie River. Another recorded location of the breeding habitats of black flies. Photo credit: George Bradley, 1932 (USDA).

Figure B.11 Tallahatchie River near Charleston, MS

Tallahatchie River was a great breeding habitat for black flies in northern MS in the Mississippi Delta Region. Tallahatchie River as one of the collection sites for this project. Photo credit: George Bradley, 1932 (USDA).

APPENDIX C

SUPPLEMENTAL MATERIAL FOR CHAPTER III

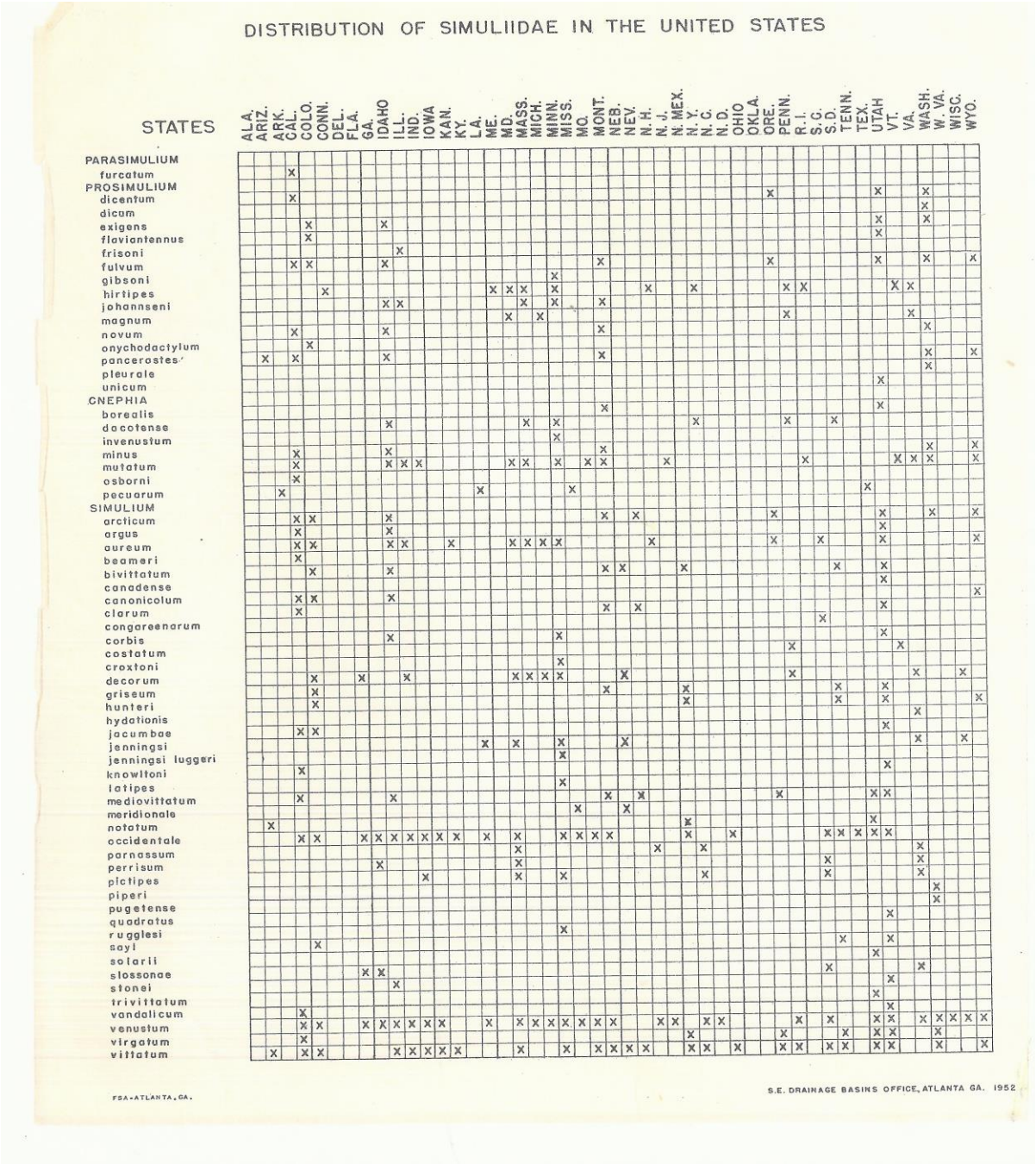


Figure C.1 Distribution map of black species in the United States from 1952

S.E. Drainage Basins Office reported on black fly species nationwide in 1952. Mississippi had only 5 species reported. Photo credit: F.S.A., Drainage Basins Office.

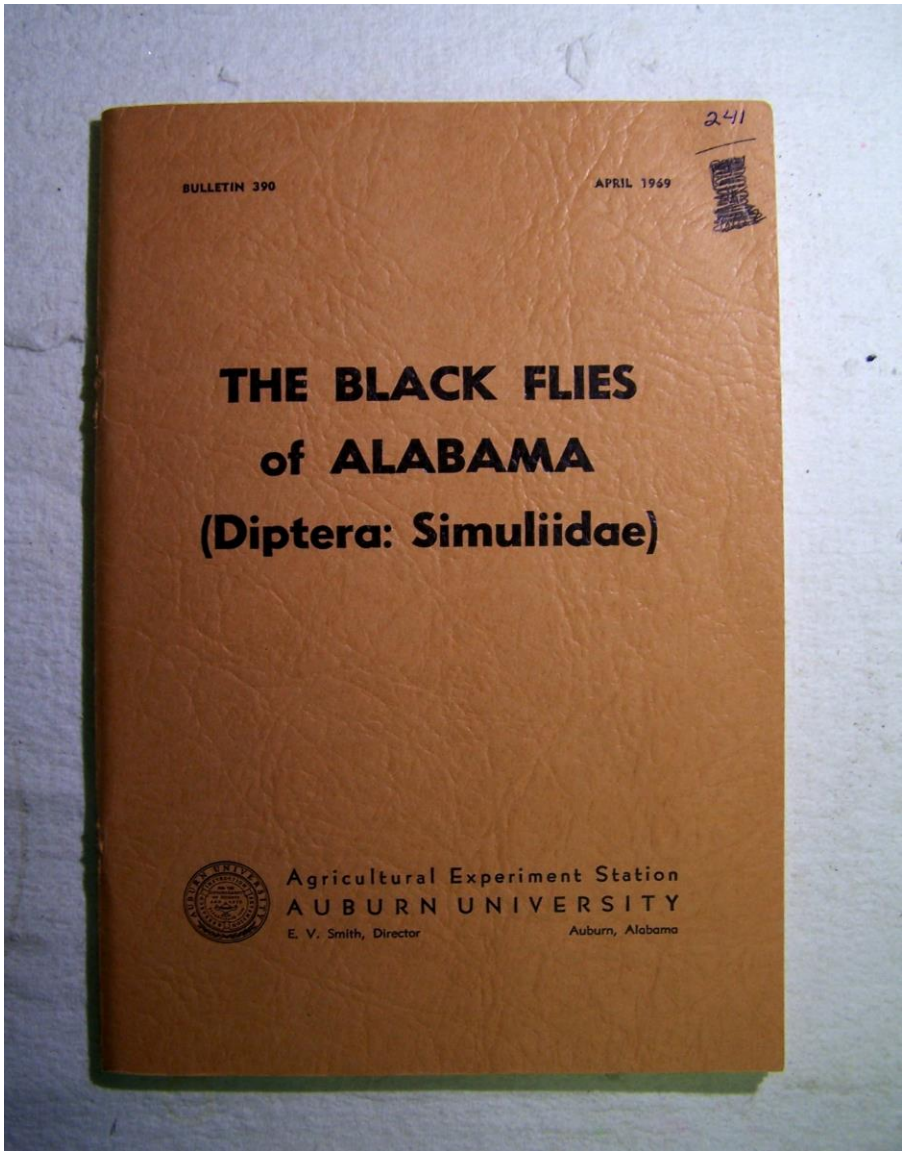


Figure C.2 The Black Flies of Alabama (Diptera: Simuliidae), Stone and Snoddy 1969

Seventeen years after the nationwide distribution list, Stone and Snoddy (1969) published a bulletin, Auburn University Agricultural Experiment Station, on the taxonomic, distribution, biology, and ecology of black flies in Alabama. This provided southeastern United States entomological professionals a better understanding of the newly described 28 species found in Alabama. Seventeen of these species have been documented in Mississippi (Nations et al., 2018). Photo credit: Tina Nations.

Figure C.3 CDC CO_2-baited light trap

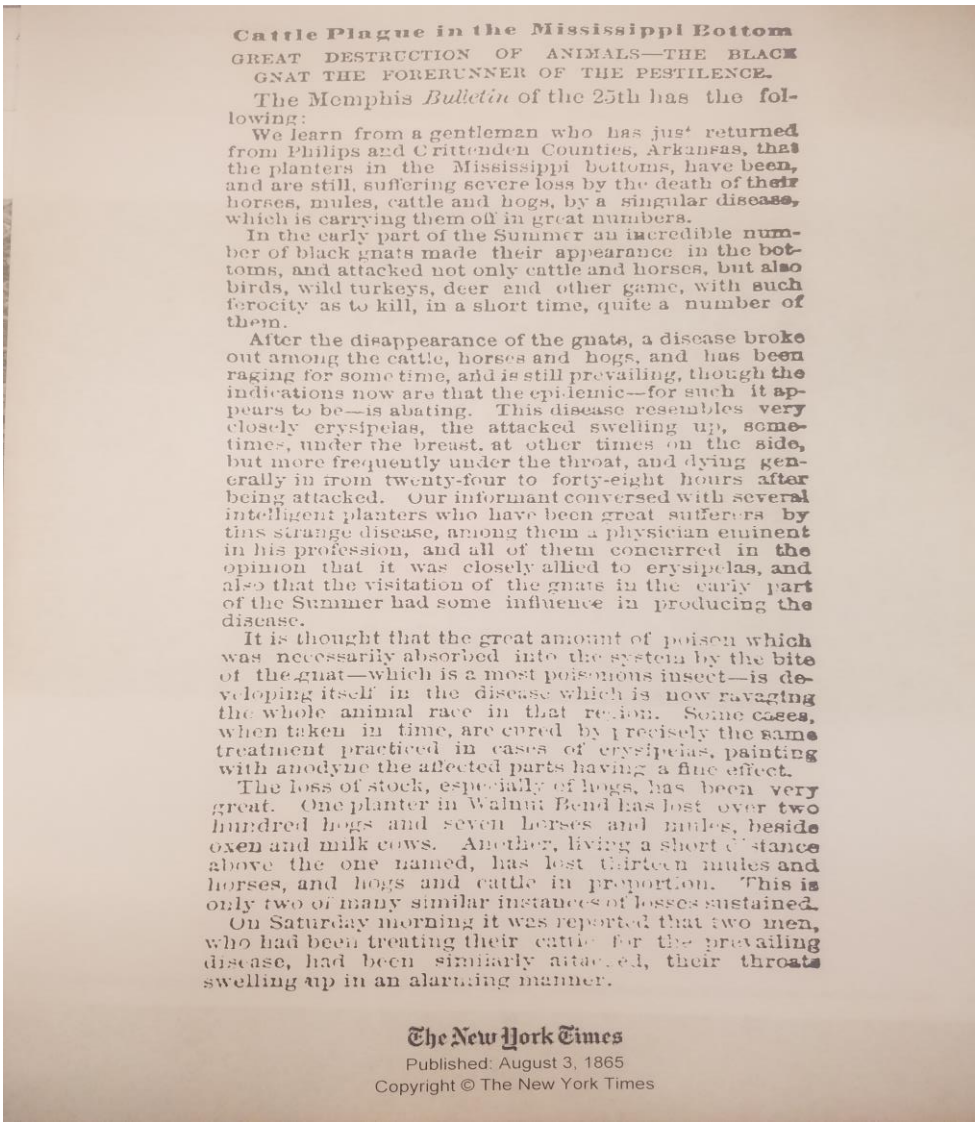
Over 8,000 black flies were collected in a 5-day period from the end of March to the beginning of April. Photo credit: Tina Nations, April 2018.

Figure C.4 Black flies from CDC CO_2-baited light trap in Jackson, MS

These nuisance pests, identified as *Simulium meridionale*, killed two backyard chickens in Jackson, MS during the last week of March 2018. Estimated size of adult(s), 1 – 5mm. Photo credit: Tina Nations, April 2018.

APPENDIX D

SUPPLEMENTAL MATERIAL FOR CHAPTER IV



Cattle Plague in the Mississippi Bottom

GREAT DESTRUCTION OF ANIMALS—THE BLACK GNAT THE FORERUNNER OF THE PESTILENCE.

The Memphis *Bulletin* of the 25th has the following:

We learn from a gentleman who has just returned from Philips and Crittenden Counties, Arkansas, that the planters in the Mississippi bottoms, have been, and are still, suffering severe loss by the death of their horses, mules, cattle and hogs, by a singular disease, which is carrying them off in great numbers.

In the early part of the Summer an incredible number of black gnats made their appearance in the bottoms, and attacked not only cattle and horses, but also birds, wild turkeys, deer and other game, with such ferocity as to kill, in a short time, quite a number of them.

After the disappearance of the gnats, a disease broke out among the cattle, horses and hogs, and has been raging for some time, and is still prevailing, though the indications now are that the epidemic—for such it appears to be—is abating. This disease resembles very closely erysipelas, the attacked swelling up, sometimes, under the breast, at other times on the side, but more frequently under the throat, and dying generally in from twenty-four to forty-eight hours after being attacked. Our informant conversed with several intelligent planters who have been great sufferers by this strange disease, among them a physician eminent in his profession, and all of them concurred in the opinion that it was closely allied to erysipelas, and also that the visitation of the gnats in the early part of the Summer had some influence in producing the disease.

It is thought that the great amount of poison which was necessarily absorbed into the system by the bite of the gnat—which is a most poisonous insect—is developing itself in the disease which is now ravaging the whole animal race in that region. Some cases, when taken in time, are cured by precisely the same treatment practiced in cases of erysipelas, painting with anodyne the affected parts having a fine effect.

The loss of stock, especially of hogs, has been very great. One planter in Walnut Bend has lost over two hundred hogs and seven horses and mules, beside oxen and milk cows. Another, living a short distance above the one named, has lost thirteen mules and horses, and hogs and cattle in proportion. This is only two of many similar instances of losses sustained.

On Saturday morning it was reported that two men, who had been treating their cattle for the prevailing disease, had been similarly attacked, their throats swelling up in an alarming manner.

The New York Times
Published: August 3, 1865

Figure D.1 Black fly article in The New York Times, 1865

Black flies were becoming such a nuisance in the 1860s that The New York Times published an article from the Memphis Bulletin making the public aware about the economic effects they had on the community. Photo credit: Tina Nations; copyright article: The New York Times, August 3, 1865.

Figure D.2 Historical Highwater Events sign in Greenville, Mississippi

According to the Greenville water gauge, there were historical highwater levels in 2008, 2009, and 2011 which corresponded with outbreaks in that area during those years. Photo credit: Jerome Goddard.

Historical site of black fly outbreak – Money, MS

Figure D.3 Money, MS

Dr. George Bradly documented Money, MS as one of the outbreak areas in his research, specifically noting, the Tallahatchie River as a breeding habitat. Photo credit: Jerome Goddard.

Figure D.4 Collection Site #1

Black fly collection site #1 in north MS, near Lula, along the Mississippi River (Table 4.1). Photo credit: Tina Nations.

Figure D.5 Collection Site #2

Coldwater River in north Mississippi, near Sledge, was the location for collection site #2 (Table 4.1). Photo credit: Tina Nations.

Figure D.6 Collection Site #3

This location is 6.5 miles east of Webb, MS, along the Tallahatchie River. Dr. George Bradley referenced this location in his works as a breeding habitat for black flies in the 1930s (Table 4.1). Photo credit: Tina Nations.

Figure D.7 Collection Site #4

Another referenced breeding location by Dr. George Bradley in the 1930s, near Money, MS, along the Tallahatchie River (Table 4.1). Photo credit: Tina Nations.

Figure D.8 Collection Site #5

The Mississippi River in Greenville, MS, 2019. Photo credit: Jerome Goddard.

Figure D.9 Collection Site #6

Vicksburg, MS, along the Mississippi River, has had several black fly outbreaks over the years. The local Public Works Department collected over 2,000 adult black flies, *Simulium meridionale* Riley, in June 2011 (Table 4.1) (see Chapter III for the distribution of *S. meridionale* in Mississippi). Photo credit: Tina Nations.

Figure D.10 Collection Site #7

Recent black fly outbreaks along the Pearl River, Jackson, MS, prompted trapping at this location to document the massive number of black flies that erupted. This site is less than 1 mile from the Jackson Futbol Club and 1.6 miles from McLeod Elementary School (Table 4.1) (Figure 5.1). Photo credit: Tina Nations.

Figure D.11 Collection Site #8

Collection Site #8 along the Strong River, near Mendenhall, MS, has had reported black fly outbreaks (especially in 2018 and 2019) with local backyard poultry owners (Table 4.1). Photo credit: Tina Nations.

Figure D.12 Collection Site #9

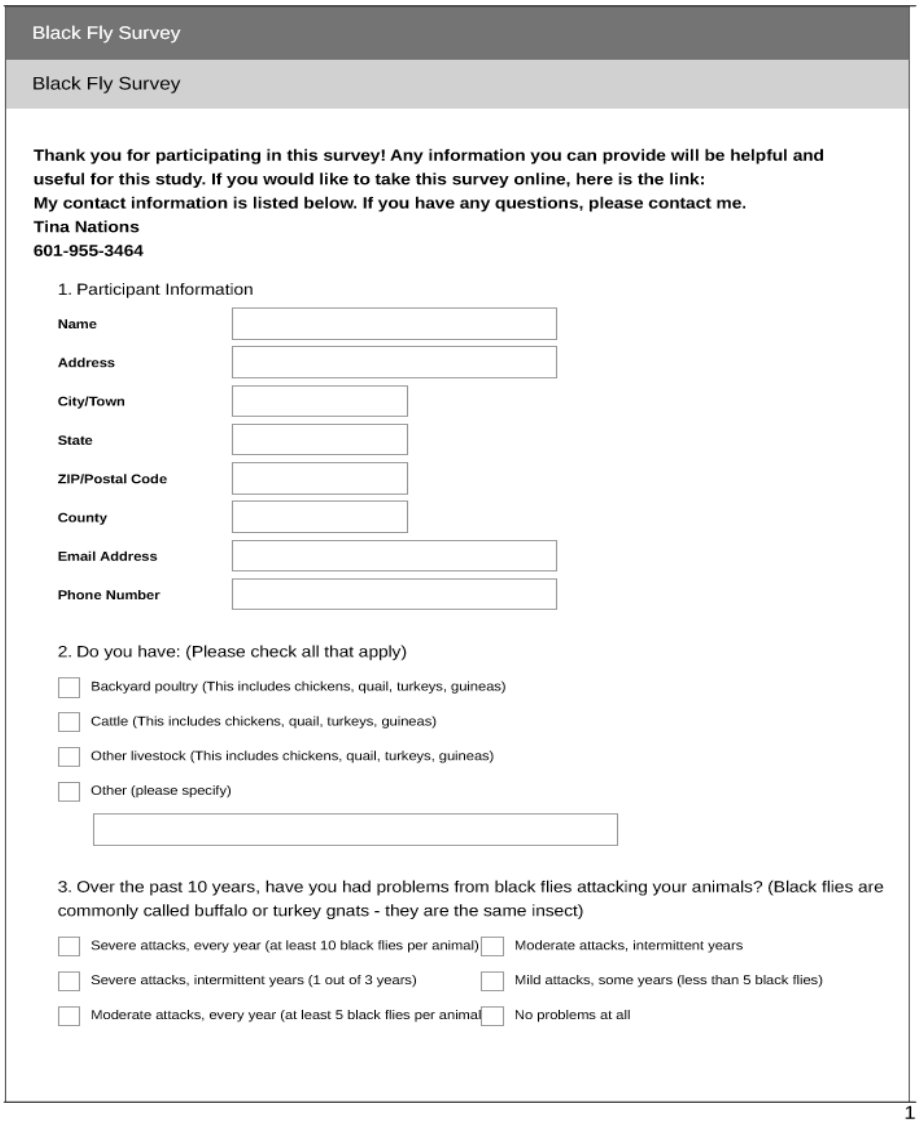
Both adult black fly pest species, *Simulium jenningsi* group Malloch and *Simulium meridionale* Riley, have been collected at this local outdoor recreational spot in Seminary, MS, at the Okatoma Creek (Table 4.1). Photo credit: Tina Nations.

Figure D.13 Collection Site #10

Collection site #10 has had an enormous abundance of *Simulium jenningsi* group Malloch erupt from the Buckatunna Creek. Dr. Jerome Goddard reported "dive-bombing" from black flies at this location (Table 4.1). Photo credit: Tina Nations.

APPENDIX E

SUPPLEMENTAL MATERIAL FOR CHAPTER V



Black Fly Survey

Black Fly Survey

Thank you for participating in this survey! Any information you can provide will be helpful and useful for this study. If you would like to take this survey online, here is the link:
My contact information is listed below. If you have any questions, please contact me.
Tina Nations
601-955-3464

1. Participant Information

Name
Address
City/Town
State
ZIP/Postal Code
County
Email Address
Phone Number

2. Do you have: (Please check all that apply)

- Backyard poultry (This includes chickens, quail, turkeys, guineas)
- Cattle (This includes chickens, quail, turkeys, guineas)
- Other livestock (This includes chickens, quail, turkeys, guineas)
- Other (please specify)

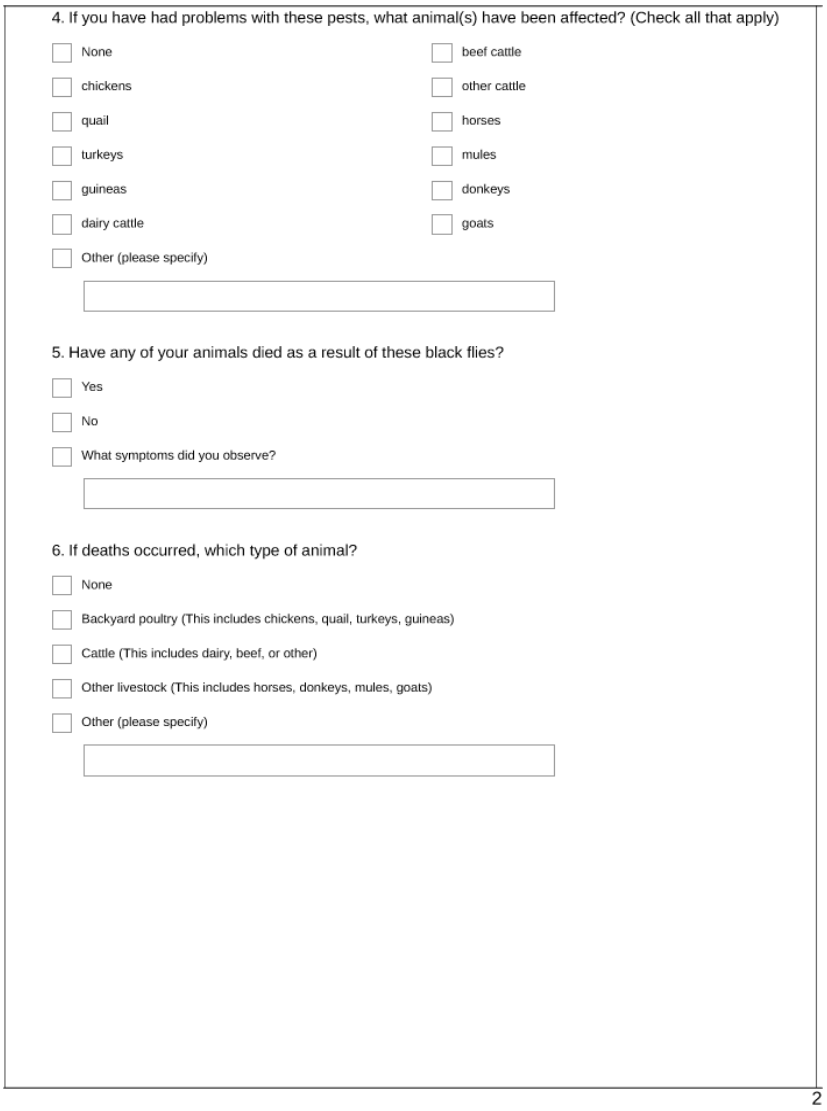
3. Over the past 10 years, have you had problems from black flies attacking your animals? (Black flies are commonly called buffalo or turkey gnats - they are the same insect)

- Severe attacks, every year (at least 10 black flies per animal)
- Severe attacks, intermittent years (1 out of 3 years)
- Moderate attacks, every year (at least 5 black flies per animal
- Moderate attacks, intermittent years
- Mild attacks, some years (less than 5 black flies)
- No problems at all

1

Figure E.1 Backyard poultry questionnaire, page 1

Page one of the backyard poultry questionnaire asked basic information from the participant. Credit: Survey Monkey.com.


4. If you have had problems with these pests, what animal(s) have been affected? (Check all that apply)

- None
- chickens
- quail
- turkeys
- guineas
- dairy cattle
- Other (please specify)
- beef cattle
- other cattle
- horses
- mules
- donkeys
- goats

5. Have any of your animals died as a result of these black flies?

- Yes
- No
- What symptoms did you observe?

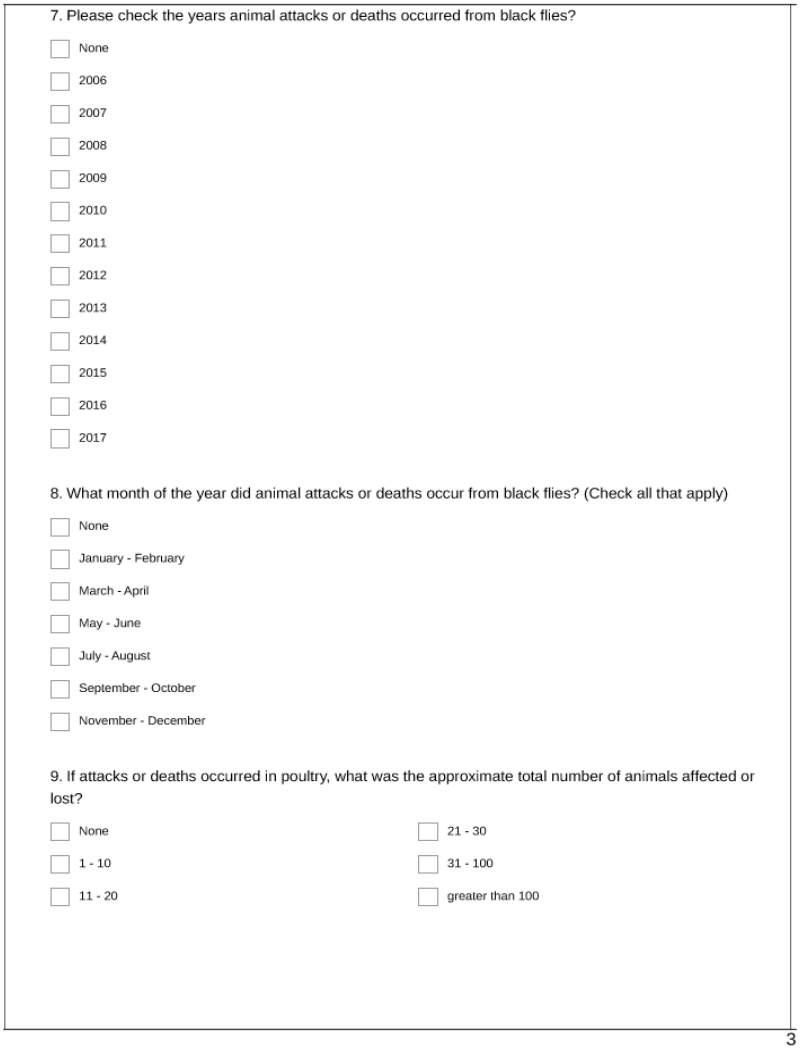
6. If deaths occurred, which type of animal?

- None
- Backyard poultry (This includes chickens, quail, turkeys, guineas)
- Cattle (This includes dairy, beef, or other)
- Other livestock (This includes horses, donkeys, mules, goats)
- Other (please specify)

2

Figure E.2 Backyard poultry questionnaire, page 2

Questions detailing the type of animal an owner had any documented symptoms from the animal(s). And, did the owner have any deaths. Credit: Survey Monkey.com.



7. Please check the years animal attacks or deaths occurred from black flies?

- None
- 2006
- 2007
- 2008
- 2009
- 2010
- 2011
- 2012
- 2013
- 2014
- 2015
- 2016
- 2017

8. What month of the year did animal attacks or deaths occur from black flies? (Check all that apply)

- None
- January - February
- March - April
- May - June
- July - August
- September - October
- November - December

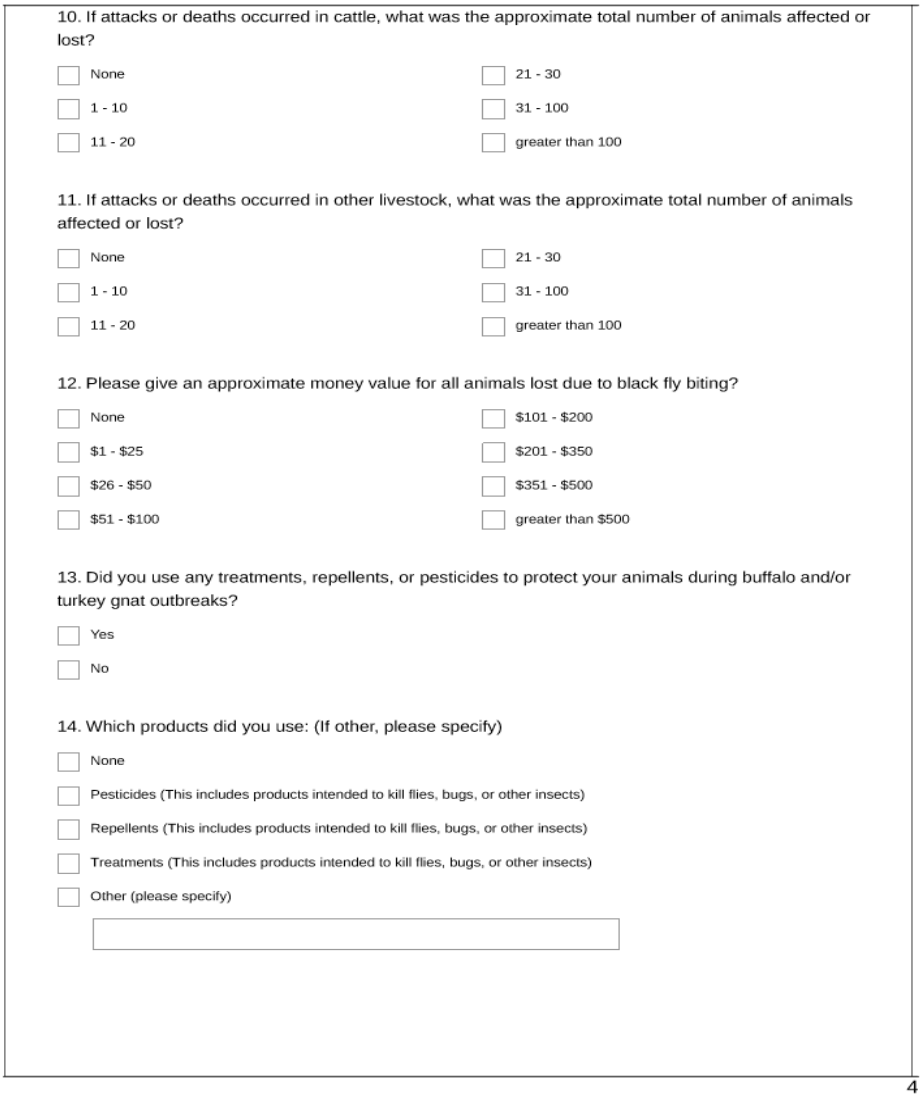
9. If attacks or deaths occurred in poultry, what was the approximate total number of animals affected or lost?

- None
- 1 - 10
- 11 - 20
- 21 - 30
- 31 - 100
- greater than 100

3

Figure E.3 Backyard poultry questionnaire, page 3

Determining months of outbreaks/attacks was an important factor for correlating seasonality. Credit: Survey Monkey.com.



10. If attacks or deaths occurred in cattle, what was the approximate total number of animals affected or lost?

- ☐ None
- ☐ 1 - 10
- ☐ 11 - 20
- ☐ 21 - 30
- ☐ 31 - 100
- ☐ greater than 100

11. If attacks or deaths occurred in other livestock, what was the approximate total number of animals affected or lost?

- ☐ None
- ☐ 1 - 10
- ☐ 11 - 20
- ☐ 21 - 30
- ☐ 31 - 100
- ☐ greater than 100

12. Please give an approximate money value for all animals lost due to black fly biting?

- ☐ None
- ☐ $1 - $25
- ☐ $26 - $50
- ☐ $51 - $100
- ☐ $101 - $200
- ☐ $201 - $350
- ☐ $351 - $500
- ☐ greater than $500

13. Did you use any treatments, repellents, or pesticides to protect your animals during buffalo and/or turkey gnat outbreaks?

- ☐ Yes
- ☐ No

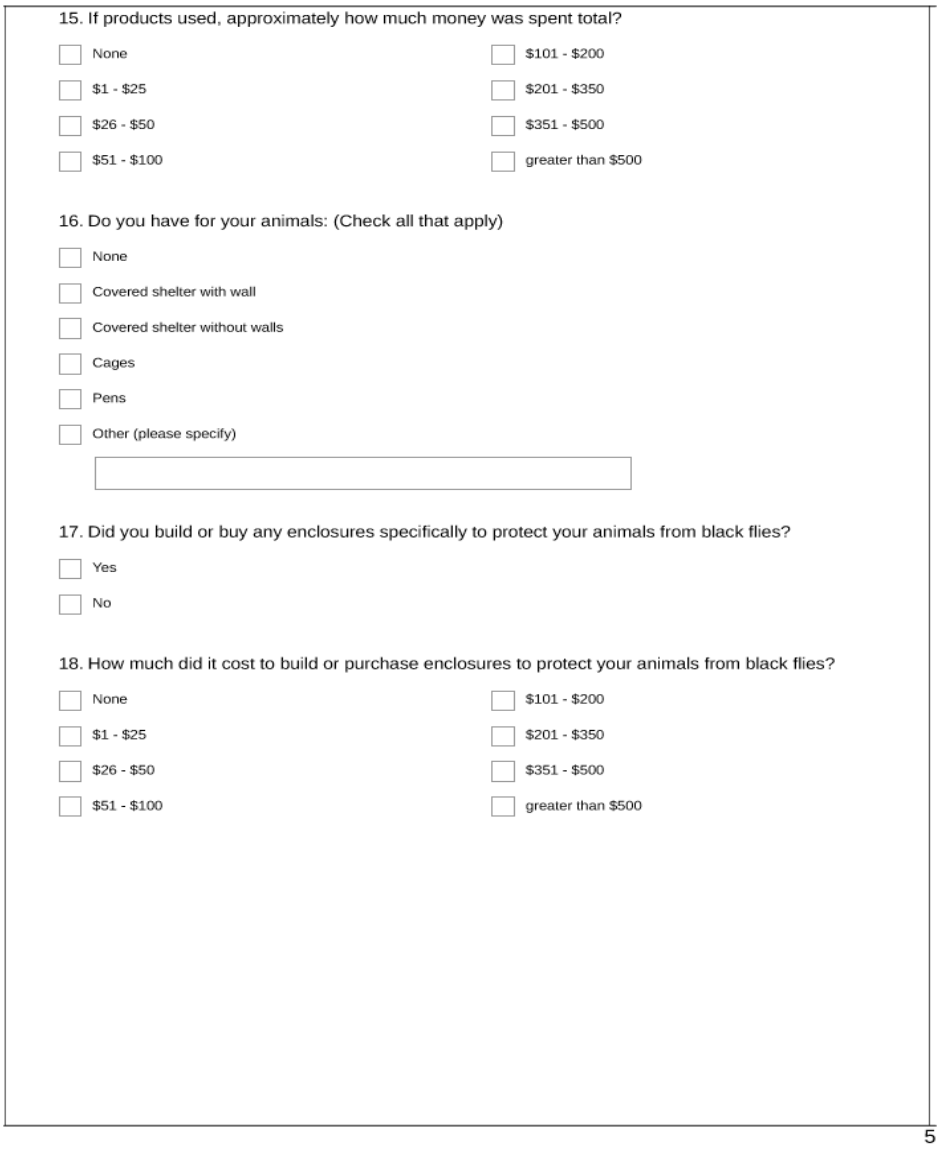
14. Which products did you use: (If other, please specify)

- ☐ None
- ☐ Pesticides (This includes products intended to kill flies, bugs, or other insects)
- ☐ Repellents (This includes products intended to kill flies, bugs, or other insects)
- ☐ Treatments (This includes products intended to kill flies, bugs, or other insects)
- ☐ Other (please specify)

4

Figure E.4 Backyard poultry questionnaire, page 4

Economic impacts were a major objective with this project. Credit: Survey Monkey.com.



15. If products used, approximately how much money was spent total?

- [ ] None
- [ ] $1 - $25
- [ ] $26 - $50
- [ ] $51 - $100
- [ ] $101 - $200
- [ ] $201 - $350
- [ ] $351 - $500
- [ ] greater than $500

16. Do you have for your animals: (Check all that apply)

- [ ] None
- [ ] Covered shelter with wall
- [ ] Covered shelter without walls
- [ ] Cages
- [ ] Pens
- [ ] Other (please specify)

17. Did you build or buy any enclosures specifically to protect your animals from black flies?

- [ ] Yes
- [ ] No

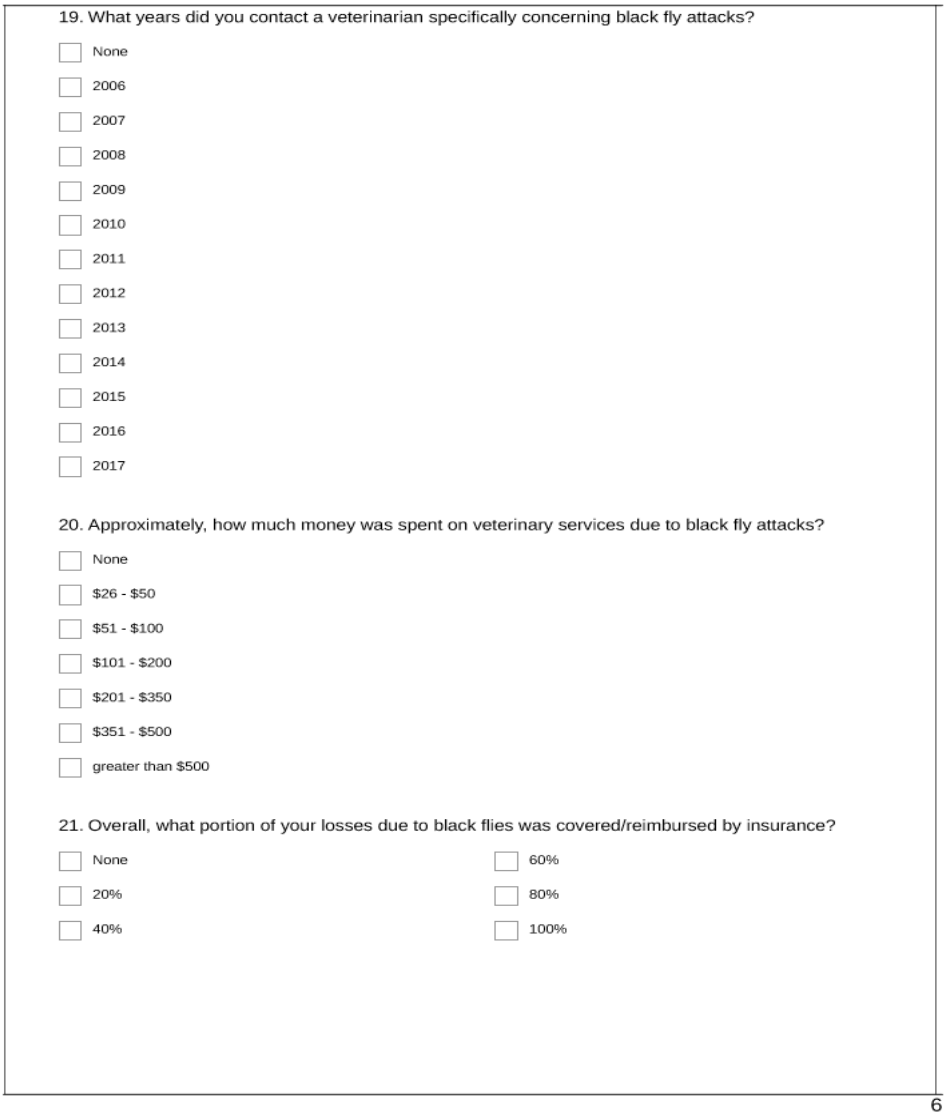
18. How much did it cost to build or purchase enclosures to protect your animals from black flies?

- [ ] None
- [ ] $1 - $25
- [ ] $26 - $50
- [ ] $51 - $100
- [ ] $101 - $200
- [ ] $201 - $350
- [ ] $351 - $500
- [ ] greater than $500

5

Figure E.5 Backyard poultry questionnaire, page 5

Monetary impacts were one of main objectives for this questionnaire. Credit: Survey Monkey.com.



19. What years did you contact a veterinarian specifically concerning black fly attacks?

- [ ] None
- [ ] 2006
- [ ] 2007
- [ ] 2008
- [ ] 2009
- [ ] 2010
- [ ] 2011
- [ ] 2012
- [ ] 2013
- [ ] 2014
- [ ] 2015
- [ ] 2016
- [ ] 2017

20. Approximately, how much money was spent on veterinary services due to black fly attacks?

- [ ] None
- [ ] $26 - $50
- [ ] $51 - $100
- [ ] $101 - $200
- [ ] $201 - $350
- [ ] $351 - $500
- [ ] greater than $500

21. Overall, what portion of your losses due to black flies was covered/reimbursed by insurance?

- [ ] None
- [ ] 20%
- [ ] 40%
- [ ] 60%
- [ ] 80%
- [ ] 100%

6

Figure E.6 Backyard poultry questionnaire, page 6

This questionnaire determined economic impacts black flies have on backyard poultry and other domestic animals. Credit: Survey Monkey.com.