

DETERMINANTS OF SPATIAL AND TEMPORAL VARIATION OF WEST NILE VIRUS
TRANSMISSION IN TEXAS

A Dissertation

by

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ABSTRACT

West Nile virus (WNV) is a zoonotic vector-borne virus that infects avian and mammal hosts. In Texas, WNV was first reported in 2002 in Harris County and has since been reported annually throughout the state. With variable funding available for mosquito surveillance in Texas, predictive modeling is an economical method for mosquito control, but has not been parameterized for major metropolitan areas of central and southeast Texas. Thus, this dissertation uses historical databases to create predictive models that are specifically tailored for major cities in Texas.

To investigate the 2012 WNV epidemic in Dallas County, TX, logistic regression models identified an index of urbanization (composed of greater population density, lower normalized difference vegetation index, higher coverage of urban land types, and more impervious surfaces), lower elevation, and older populations as key factors in predicting the risk of WNV in *Culex quinquefasciatus*. Our model was then extrapolated as a risk map, which highlighted north and central Dallas County as areas of high risk for WNV-positive mosquitoes.

A similar study for Harris County was conducted, where the best-fit model found that areas with higher elevation, more impervious surfaces, greater median income, and predominantly Hispanic populations will have higher vector indexes, which measure the average number of WNV-infected female *Culex* mosquitoes collected per trap night. The predictive map based on this model emphasized high-risk areas in central and north Harris County.

Harris County's long-term database was also used to investigate temporal patterns between vector abundance, WNV infection in *Cx. quinquefasciatus*, and weather patterns. A time-series analysis revealed correlations between abundance and environmental variability

measurements, following our hypothesis of Schmalhausen's law that states organisms are susceptible to mean (average) temperature and precipitation measurements as well as extreme or variability in weather. The infection rate model identified temperature with an 8-month lag as a significant covariate for WNV infection rates, highlighting the importance of overwintering temperatures preceding the WNV season.

These models (landscape, demographic, and meteorological conditions) can be used by local mosquito control agencies to predict WNV infection in *Cx. quinquefasciatus* for proactive and effective control efforts.

DEDICATION

This dissertation is dedicated to the memory of Dr. Marilyn O. Ruiz, whose work in Illinois was the inspiration for our work in mosquito and WNV surveillance in Texas. Dr. Ruiz was a true pioneer in the field and she will be greatly missed. May her legacy continue to inspire future epidemiologists and entomologists.

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This work was supervised by a dissertation committee consisting of Dr. Gabriel L. Hamer (advisor) of the Department of Entomology and Dr. Rudy Bueno, Jr. of the Department of Entomology, Dr. Daikwon Han of the Department of Epidemiology and Biostatistics, and Dr. Mikyoung Jun of the Department of Statistics.

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NOMENCLATURE

ACF	Autocorrelation function
AIC	Akaike's Information Criterion
CCF	Cross-correlation function
CDC	Centers for Disease Control and Prevention
DEM	Digital Elevation Model
GV	Gravid traps
HCPH	Harris County Public Health Mosquito and Vector Control
IR	Infection rate
JE	Japanese Encephalitis
MIR	Minimum infection rate
MLE	Maximum Likelihood Estimate
PACF	Partial autocorrelation function
<i>PCI</i>	Urbanization principal component group 1
SD	Standard deviation
SS	Storm sewer traps
ULV	Ultra-low volume adulticide spraying
VI	Vector index
WNF	West Nile Fever
WNND	West Nile Neuroinvasive Disease
WNV	West Nile virus
YFV	Yellow fever virus

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CHAPTER I

INTRODUCTION AND LITERATURE REVIEW

1.1 West Nile Virus Distribution And Epidemiology

West Nile Virus (WNV) was first discovered in 1937 in Uganda, spreading to Egypt, India, and Israel by the 1950's (Bernkopf et al. 1953, Work et al. 1953). The virus had reached Europe by the 1970's (Mouchet et al. 1970, Karabatsos 1985, Hayes and Monath 1989) and more recently, China, Japan, and South Korea found their first patients positive for WNV in 2004, 2005, and 2012, respectively (Takasaki 2007, Li et al. 2013, Hwang et al. 2015). Since its introduction to the New World in Queens, New York in 1999, WNV has spread throughout much of North America, including Canada in 2004 and is now spreading to Central and South America (Nasci et al. 2001b, Reisen and Brault 2007, Artsob et al. 2009). With one of the largest distributions, WNV has now been detected on every continent except for Antarctica (Chancey et al. 2015). The virus is now considered endemic in Europe, Asia, Africa, and North America, where the avian population is capable of maintaining the virus and human cases of WNV have been reported every year (Rappole et al. 2000, Peterson et al. 2004, Petersen et al. 2007, Pollock 2008, Paz and Semenza 2013).

Since its introduction into the United States (U.S.), the Centers for Disease Control and Prevention (CDC) has reported 46,086 cases of WNV, with 46.8% of the cases classified as the more severe form of WNV, West Nile neuroinvasive disease (WNND) (CDC 2016a). WNV has a case-fatality rate of 4% in the U.S. according to the cumulative data from the CDC, with 2,017 deaths reported since 1999 (CDC 2016a). Most cases of WNV are not clinically apparent and often go unrecognized, which may result in underreporting of the disease (Petersen et al. 2013a).

According to Petersen et al. (2013b), the actual numbers for illnesses and resulting WNV cases from 1999-2010 may be closer to 3 million and 780,000, respectively.

The U.S. witnessed the largest outbreak of WNV in 2012. In total, the U.S. and the District of Columbia reported 5,674 total cases of WNV with 2,873 of WNND cases and 286 deaths (CDC 2016b). The majority of cases and deaths during this epidemic occurred in Texas. Texas alone reported a total of 1,868 cases of WNV with 844 cases of WNND and 89 deaths throughout the state (Murray et al. 2013). During this epidemic, Texas accounted for 33% of the total cases, 29% of the WNND cases, and 31% of the total deaths reported in 2012.

The first human case of WNV in Texas occurred in 2002 in Harris County, TX and cases have consistently been reported annually throughout Texas since then (Nolan et al. 2013). Texas reported 5,277 cases of WNV to the CDC between 1999-2016 and over 60% of the cases were classified as the WNND form (CDC 2016a). Texas also has the second-highest number of total reported WNV cases in the continental U.S., following California which reported 6,031 cases in the same time period (CDC 2016a). In comparison, 2,150 WNV disease cases were reported from 46 states and the District of Columbia in 2016, and the majority of the cases (61%) were classified as the neuroinvasive form (CDC 2016b, Burakoff et al. 2018). Given that Texas has been the site of some of the largest numbers of WNF and WNND cases coupled with minimal prior work focusing solely on Texas, this dissertation will focus on WNV transmission dynamics and patterns occurring in Texas.

1.2 West Nile Virus Taxonomy

WNV is a flavivirus from family *Flaviviridae*, which includes four genera containing over 60 species. This family is most well-known for the type virus the yellow fever virus (YFV)

(Simmonds et al. 2017). The majority of the viruses in this family are arthropod-borne and the host range spans birds and mammals of all genera (Simmonds et al. 2017). WNV belongs to the *Flavivirus* genus, which also includes YFV, Dengue virus, and Zika virus (Simmonds et al. 2017). Specifically, WNV is part of the Japanese Encephalitis (JE) complex and serologically cross-reacts with the three other virus species in the complex including JE, St. Louis encephalitis (SLEV), and Murray Valley encephalitis viruses (Mackenzie et al. 2002). The members of the JE complex are responsible for causing many arthropod-borne diseases in humans (Schweitzer et al. 2009).

Previously, phylogenetic analyses have found two distinct lineages of WNV: Lineage 1 and Lineage 2. Lineage 1 has a diverse geographical distribution and can be found in Australia, the U.S., Africa, Europe, the Middle East, India, and Russia (Lanciotti et al. 2002). Whereas Lineage 2 is predominantly found in sub-Saharan Africa and Madagascar (Lanciotti et al. 2002). The original WNV strain in New York, the other strains found in the U.S., and strains from Israel form a unique clade within Lineage 1 (Lanciotti et al. 2002). Due to the high genomic similarities between the U.S. and Israel viruses, researchers hypothesized that WNV was likely introduced into the U.S. from the Middle East and more specifically Israel (Lanciotti et al. 1999).

Since the identification of the first two lineages, three more lineages have emerged. Lineage 3 is comprised of the Rabensburg virus which was isolated from mosquitoes found in South Moravia, Czech Republic (Bakonyi et al. 2005). Lineage 4 is comprised of a strain from a *Dermacentor spp.* tick originating from the Russian Caucasus region (Prilipov et al. 2002, Brault 2009). Lineage 5 encompasses a unique WNV lineage originally isolated from mosquitoes in India (Bondre et al. 2007). Curiously, while the strains were found in distinct environments around the world, the distribution of the lineages does not correlate with host preference or

geographical location, but instead with the patterns of migrating birds (Berthet et al. 1997, Brault 2009).

1.3 West Nile Virus Ecology And Transmission Cycle

WNV is a zoonotic disease involving avian hosts in the enzootic period of the virus (Figure 1). During the enzootic period of the transmission cycle, the mosquito infected with WNV feeds and transmits the virus to the avian host (Weaver and Reisen 2010). The virus amplifies within the avian host and the avian host then maintains a sufficient viremia to infect other mosquitoes, which can infect dead-end hosts, such as horses and humans. The species responsible for WNV transmission will differ depending on the predominant species in the area (Turell et al. 2005). Furthermore, characteristics of the vector such as ornithophilic feeding may make some vectors more efficient than others in maintaining the virus in nature (Turell et al. 2005). Some mosquitoes such as *Cx. quinquefasciatus* and *Cx. tarsalis* are considered opportunistic feeders and will change their host preference depending on the season, making them excellent maintenance and bridge vectors. Both mosquitoes are ornithophilic feeders in the early summer, which allow the virus to be “maintained” in the nature. These mosquitoes will then switch their feeding preferences to mammals later in the summer, allowing spillover of WNV into mammal populations (Tempelis et al. 1965, Molaei et al. 2007, Andreadis 2012).

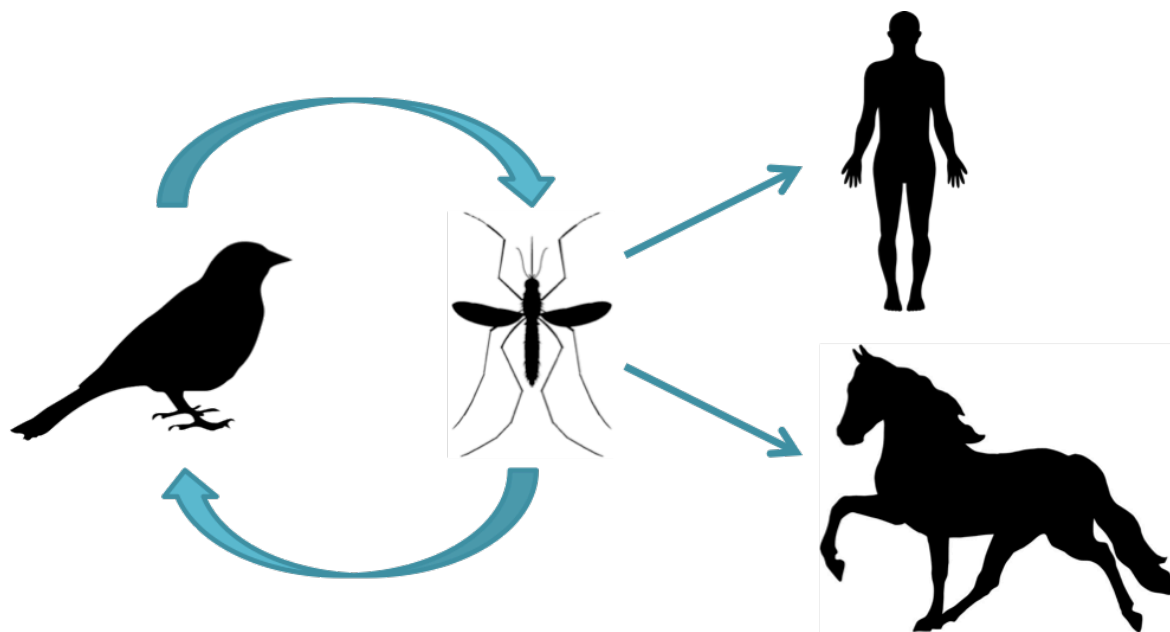


Figure 1 West Nile virus cycle. An infected mosquito feeds on an avian host, which then amplifies the virus and maintains a sufficient viremia to infect other mosquitoes. Infected mosquitoes can pass the virus to dead-end hosts such as humans and horses.

Vertical transmission of WNV is another possible mechanism for mosquito vectors to acquire the virus as studies have demonstrated effective vertical transmission of WNV from female mosquitoes to their offspring. Baqar et al. (1993) first reported vertical transmission of WNV in *Aedes albopictus*, *Ae. aegypti*, and *Cx. taeniorhynchus* F₁ offspring by parenterally infecting the parental strains in a laboratory setting. Another study in California looked at whether three wild-caught *Culex* species could vertically transmit WNV to their offspring. They found variability in *Cx. tarsalis*' ability to vertically transmit WNV (Goddard et al. 2003). *Cx. tarsalis* from Yolo County were able to transmit the virus to their offspring; however, the same species from Riverside County reported no positive mosquitoes among the F₁ progeny (Goddard et al. 2003). The authors reported positive progeny for *Cx. quinquefasciatus*, but not for *Cx. pipiens* (Goddard et al. 2003). Another group intrathoracically inoculated *Cx. pipiens* and *Ae.*

albopictus with WNV and found positive F₁ progeny from *Cx. pipiens* only (Dohm et al. 2002a). The ability of multiple species to overwinter may contribute to maintenance of WNV in nature during colder temperatures, specifically where females can enter diapause or quiescence (a period of low or absent reproductive activity) during the winter and then start vertical and horizontal transmission the following spring (Goddard et al. 2003, Unlu et al. 2010). Ultimately, while vertical transmission is possible, it occurs at very low levels (<1%) (Goddard et al. 2003).

Human and animal hosts are most commonly exposed to WNV through the bite of an infected mosquito, but Gould and Fikrig (2004) reviewed other ways humans may become infected with WNV. Although rare, humans can become exposed to WNV in an occupational setting (CDC 2002c, 2003), blood transfusion and organ donation (Iwamoto et al. 2003, Pealer et al. 2003, Desalvo et al. 2004, Kumar et al. 2004, Stanley et al. 2009), and *in utero* or breast feeding from mother to baby (CDC 2002a, b). While humans can potentially infect other humans through these alternative routes, humans cannot pass on the virus to an uninfected mosquito thus ending the WNV transmission cycle.

1.3.1 Vectors of WNV

Culex mosquitoes are the primary enzootic vectors of WNV (Turell et al. 2001, Turell et al. 2005, Weaver and Reisen 2010), as well as capable bridge vectors to humans due to their opportunistic and generalist feeding behaviors on avian and mammal hosts (Hribar et al. 2001, Turell et al. 2001, Kilpatrick et al. 2005b, Hamer et al. 2008b). More specifically, *Cx. pipiens*, *Cx. restuans*, *Cx. quinquefasciatus*, and *Cx. tarsalis* are the principal vectors for WNV in the U.S., but other species may be competent vectors with lower transmission rates (Turell et al. 2001). *Cx. pipiens* is the most important vector for the northeast, northcentral, and mid-Atlantic

U.S., but *Cx. restuans* may play a role in the WNV transmission cycle to a lesser extent (Apperson et al. 2002, Turell et al. 2002, Ebel et al. 2005, Kilpatrick et al. 2006, Kramer et al. 2008). Among the midwestern and western states, *Cx. tarsalis* is considered the main vector of WNV since it has the greatest abundance in those areas, can maintain and amplify similar viruses in the JE complex, and is an efficient laboratory vector of WNV (Goddard et al. 2002, Bell et al. 2005, Turell et al. 2005, DiMenna et al. 2006, Nielsen et al. 2008, Brault 2009). Furthermore, *Cx. pipiens* is considered the major WNV vector in the northeastern states based on their ornithophilic feeding preferences, predominant abundance in the northeast, and high infection rates associated with WNV (Turell et al. 2000, Bernard et al. 2001, Kilpatrick et al. 2005b).

In the southern U.S., including Texas, *Cx. quinquefasciatus* is the most relevant mosquito species involved in the transmission cycle, exhibiting an opportunistic feeding behavior and high abundance during the WNV transmission season (summer through early fall) (Gibbs et al. 2006, Molaei et al. 2007, Reisen et al. 2008b, Andreadis 2012). Molaei et al. (2007) reported feeding preference shifts for *Cx. quinquefasciatus* mosquitoes in Harris County, TX throughout the WNV season. The authors found a pronounced shift in the sources of blood meals, shifting from avian hosts to mammalian hosts later in the summer and early fall between September and November (Molaei et al. 2007). While feeding behaviors and general phenology of *Cx. quinquefasciatus* has been studied, but how the surrounding environment and conditions affect the mosquito are still largely unknown for Texas. Given the accepted role of *Cx. quinquefasciatus* as the main vector of WNV in Texas, this dissertation will mainly focus on the ecology of this mosquito species and how its life cycle can change based on external pressures.

1.3.2 Reservoirs and Hosts of WNV

Birds, humans, and horses are the most well-known hosts for WNV. Birds serve as major amplifying hosts of the virus. Some species of birds are capable of tolerating the virus and producing high enough viremic titers to infect mosquitoes that feed on them, but other species may die as a consequence of infection. In the U.S., over 300 species of dead birds have tested positive for WNV (CDC 2017). When WNV first arrived in North America, Canada geese (*Branta canadensis*), domestic geese (*Anser spp.*), chickens (*Gallus gallus*), house sparrows (*Passer domesticus*), and rock doves (*Columba livia*) had high seroprevalences (Komar et al. 2001). After the experimental infection of 25 bird species with WNV, the most competent species for WNV consisted of passerines: blue jays (*Cyanocitta cristata*), common grackles (*Quiscalus quiscula*), house finches (*Carpodacus mexicanus*), American crows (*Corvus brachyrhynchos*), and house sparrows (Komar et al. 2003). Passerine birds are competent amplifying hosts and will produce sufficient concentrations of the virus to efficiently infect mosquitoes feeding on them to continue the enzootic cycle. Members of the Corvidae family have also demonstrated severe illness and subsequent high mortality rates from WNV infection (Reed et al. 2003). While birds are typically infected through mosquito feeding, birds may also be infected through ingestion of an aqueous solution containing the virus or an infected mosquito (Komar et al. 2003). Oral ingestion of WNV may have important implications for scavenging birds of prey such as vultures, hawks, or eagles that may eat infected prey or carrion and warrants further research (Marra et al. 2004a).

Migratory birds may have played a significant role in bringing WNV to the New World and the consequent distribution of WNV throughout North America. For example, Rappole et al. (2000) suggested how the Eurasian Wigeon (*Anas Penelope*) and its international migration

patterns might have introduced WNV into North America. The bird is commonly found in Iceland but travels to temperate and tropical zones in the Old World for the winter, where it could come into contact with mosquitoes carrying WNV (Edgell 1984). There is also evidence that Eurasian Wigeons can also travel to the Eastern seaboard of North America, which could have served as an entry point for WNV into the Western hemisphere (Edgell 1984). The migratory patterns of various bird species may have also contributed to the spread of WNV throughout the country in a short amount of time. WNV expanded throughout North America along the Atlantic seaboard, which aligns with the Atlantic flyway migration route taken by many bird species (Rappole et al. 2000). After expanding to new regions, WNV can establish the zoonotic cycle in the new environment and remain active year-round, which is true for subtropical climates that are home to avian reservoirs of WNV (Reed et al. 2003). The rapid spread of WNV westward in North America could be contributed to the elliptical migration pattern of bird species such as the Connecticut warbler (*Oporornis agilis*) (Reed et al. 2003). Furthermore, simulations of the spread of WNV follows closely with mosquito and migratory bird movement, where migratory birds serve as critical long-distance transport agents (Peterson et al. 2003). The high viremic titers in migratory birds further support the hypothesis that migratory birds play a major role in WNV distribution (Roehrig et al. 2002).

Experimental and natural infection of WNV in various animals has demonstrated the virus' ability to adapt to a wide range of hosts. WNV antigen and antibodies can be found in various vertebrates including bats (Constantine 1970, Paul et al. 1970, Davis et al. 2005), domestic cats, dogs, and rabbits (Odelola and Oduye 1977, Komar 2000, Marfin et al. 2001, Austgen et al. 2004), peridomestic animals (cotton-tailed rabbits, raccoons, opossums, chipmunks, squirrels, skunks) (Marfin et al. 2001, Tiawsirisup et al. 2005, Bentler et al. 2007,

Platt et al. 2007, Blitvich et al. 2009), frogs (Kostyukov et al. 1985, Kostyukov et al. 1986, Klenk and Komar 2003), alligators (Miller et al. 2003), and crocodiles (Steinman et al. 2003, Machain-Williams et al. 2013). The movement of these widely-varied animals, whether through natural migration or human-mediated movement, could potentially introduce WNV into regions with previously no or low prevalence of the virus.

Finally, humans and horses also serve as a host for WNV, but to a much lesser extent compared to birds. Humans and horses are known as dead-end or incidental hosts, where they may exhibit symptoms and die, but are not able to produce a sufficient viremia to infect another mosquito. While humans cannot infect another mosquito, they can potentially infect other humans through blood-to-blood or blood-to-mucous membrane contact such as organ or blood donations, occupational/laboratory settings, or from mother to child (Gould and Fikrig 2004).

1.4 Drivers of WNV Transmission

WNV is considered the arbovirus with the largest geographic distribution in the world (Kramer et al. 2008). Although WNV has been introduced into diverse landscapes around the world, WNV does not act consistently in these different environments. The ability of WNV to become established in the enzootic cycle, amplify to high levels, and occasionally spillover to human hosts is highly variable in space over large geographic areas (Murray et al. 2010, Petersen et al. 2013a). Even at small spatial scales, there is substantial heterogeneity in mosquito, bird, and human infections (Ruiz et al. 2004). WNV is also highly variable temporally, with each season having variable degrees of transmission intensity and seasonal activity (Ruiz et al. 2010). In the last 15 years, there has been substantial progress in understanding the biotic and abiotic

drivers of WNV transmission (Randolph and Rogers 2010). In this section, we will review the major drivers of WNV transmission that have been identified thus far in the literature.

1.4.1 Temperature

Temperature has direct and indirect impacts on mosquito populations and the virus. Ambient temperature affects the rate of development and abundance of vector populations (Rueda et al. 1990, Paz and Albersheim 2008) and changes the seasonal phenology of mosquito and host populations (Reisen et al. 2010, Mirski et al. 2012). Furthermore, ambient temperatures may determine the spatial and temporal variation in human case incidence, mosquito acquisition of the virus, and reservoir presence (Reisen et al. 2006a, Mirski et al. 2012). Additionally, increasing temperatures decrease the extrinsic incubation period—the time interval between an infectious blood meal until the mosquito is capable of transmitting the virus—and accelerate the rate of virus evolution (Smith 1987, Kilpatrick et al. 2008). Ambient temperature has direct impacts on the WNV cycle. Warmer temperatures increase mosquito productivity, increase the growth rate of mosquito populations by shortening the time of development for *Culex* vectors, and decrease the gonotrophic interval by increasing biting rates (Meyer et al. 1990, Shaman et al. 2005, Kilpatrick et al. 2008, Paz and Albersheim 2008, Ruiz et al. 2010, Paz et al. 2013, Paz and Semenza 2013).

Ambient temperature is a leading risk factor for mosquito development and transmission efficiency. Experimentally, higher incubation temperatures of *Cx. pipiens* resulted in higher infection rates with disseminated infections reported as early as 4 days post-infection at 30°C (Dohm et al. 2002b, Reisen et al. 2008b). Furthermore, transmission accelerates nonlinearly with the extrinsic incubation temperature, rendering higher temperatures an important risk factor in

WNV transmission (Kilpatrick et al. 2008). Ruiz et al. (2010) reported a significant positive association between temperature on the timing and location of increased mosquito infection in Chicago, IL. As a result, increased temperature has a strong effect on increasing the minimum infection rate (MIR) within a week (Ruiz et al. 2010).

This strong positive association between temperature and WNV has also been found in other regions (Chuang et al. 2011, Chung et al. 2013, Degroote et al. 2014). In South Dakota, higher temperature in the current and two weeks prior to a trapping event in combination with higher precipitation 3-4 weeks prior shows a positive influence on *Cx. tarsalis* abundance (Chuang et al. 2011). Degroote et al. (2014) concluded that higher than average winter temperatures from the previous year and higher than average spring and early summer temperatures were associated with higher WNV ratings throughout various regions of the U.S. during the 2012 WNV epidemic.

More locally in Texas, a study investigating factors that led to the 2012 WNV outbreak in Dallas looked at previous winter temperatures and discovered an inverse association between the number of WNND cases and the number of freezing temperature days below 28°C (Chung et al. 2013). Furthermore, the same study found that another large outbreak in Dallas took place in 2006 and experienced a similarly large number of outliers in regards to temperature, rainfall, and summer heat (Chung et al. 2013). The group concluded that higher temperatures combined with an earlier spring increased amplification of WNV activity within the enzootic cycle and, thusly, increased transmission in humans (Chung et al. 2013).

While temperature's positive relationship with mosquito abundance and WNV occurrence is currently accepted, higher temperatures can also have a negative effect on these outcomes. For example, Brault (2009) concluded that temperature negatively correlates with the

longevity of mosquitoes, but higher temperatures may increase transmission efficiency. Likewise, mosquitoes begin to slow their activity at extremely high temperatures (Reisen 1995, Reisen et al. 2006a). Regardless, temperature remains to be a significant risk factor for mosquito survival, successful virus transmission, and the maintenance of WNV in reservoirs and vectors as concluded by the various aforementioned studies.

1.4.2 Precipitation

Precipitation is known to have important consequences on mosquito production and abundance by providing habitats for the larval and pupal stages of mosquitoes, which also has an impact on rates of WNV transmission. However, the influence of prior precipitation on WNV transmission is complex and no clear patterns have emerged from multiple studies. Major reasons for differences over large geographic areas could be due to the different predominant mosquito vectors of WNV in certain areas and their specific ecologies.

Above-average precipitation and the resulting pools of standing water can increase the abundance of mosquitoes and, therefore, lead to increased incidences of mosquito-borne diseases such as WNV (Takeda et al. 2003, Soverow et al. 2009). However, other studies have demonstrated that heavy rainfall can disturb *Culex* larval habitats by washing away the ditches and drainage channels these mosquitoes prefer, with the greater degrees of washouts determined by longer rain exposure (Shaman et al. 2002b, Koenraadt and Harrington 2008). Storm sewer catch basins, ditches, and organic waste sites are ideal habitats for *Cx. quinquefasciatus* since these locations are polluted with a high density of organic material. Therefore, we would expect a decrease in the abundance of this species if the larval habitats are constantly washed away (Molaei et al. 2007).

While rainfall can be a driver of WNV activity, the lack of rain may also be just as important. Drought conditions disrupt the aquatic ecosystem of predators and competitors that serve to limit mosquito larval activity, allowing larvae to fully develop and emerge as adults (Chase and Knight 2003). In southern Florida, droughts can cause fragmentation in surface pools and the remaining stagnant water will become enriched with organic materials, as well as draw avian hosts and mosquitoes closer to limited water sources, increasing the interactions between the reservoir and vector to facilitate the enzootic cycling and amplification of WNV (Shaman et al. 2002b, Shaman et al. 2005). SLEV, which has a similar transmission cycle to WNV and has since been outcompeted by WNV, follows similar conditions for increased risk of SLEV transmission to humans where birds and *Culex* mosquitoes congregate together during droughts (Shaman et al. 2002a). In Dallas, TX, the 2012 WNV epidemic was attributed to drought conditions that reduced water movement, created stagnant water pools, and accumulated high organic content ideal for *Culex* and other vector mosquitoes to breed (Roehr 2012, Chung et al. 2013), but perhaps increased bird and *Culex* mosquito congregation played a role as well.

The timing and location of precipitation is also important, given its interactions with other abiotic factors such as temperature, slope, and the landscape of the local area. A study in Chicago, IL investigated the role of temperature, precipitation, and landscape features on MIR, where Ruiz et al. (2010) concluded that lower precipitation in the spring followed by increased precipitation preceded increases in MIR, but this was not true for all years. Overall, lower precipitation was considered the leading predictor for high MIR's (Ruiz et al. 2010). Furthermore, the authors also suggested that lower elevations and more impervious surfaces will allow water to collect at low-lying elevations and produce resources needed for mosquito survival (Ruiz et al. 2010).

Rainfall was a significant factor in prediction models for the 2012 WNV epidemic throughout the U.S., as shown by Degroote et al. (2014), where large amounts of precipitation early in the year and drought conditions later in the year were associated with higher WNV activity. Winter to spring precipitation had a positive association with higher WNV activity in the following summer, but summer precipitation was inversely associated with WNV activity (Degroote et al. 2014). This suggests that early winter and spring precipitation will accumulate water on the surface for mosquitoes to breed and coupled with high temperatures, will facilitate high mosquito abundances early in the season (Degroote et al. 2014). The standing water will further become enriched the longer it stays in the environment and provide excellent larval habitats for *Cx. quinquefasciatus* (Degroote et al. 2014). Precipitation from the previous year may be a significant driver of virus transmission in subtropical areas, but this is limited by the geographic heterogeneity and may not be true outside of subtropical areas (Uejio et al. 2012). Landesman et al. (2007) showed that there are inconsistent precipitation patterns when looking at different regions of the U.S. due to the inherent geographic heterogeneity. The authors found precipitation to have a positive effect on human incidence of WNV on the East coast of the U.S. and a negative effect along the west coast (Landesman et al. 2007). The opposite relationships in the pattern of human WNV patterns along the eastern and western U.S. could be due to the ecological differences of the predominant WNV vectors in their corresponding region, *Cx. pipiens* and *Cx. tarsalis*, respectively (Landesman et al. 2007).

The mechanisms involving precipitation and its effects on WNV transmission dynamics are complex and have resulted in highly variable results. Regardless, precipitation is an important factor to consider due to its indirect roles in the WNV cycle and its interactions with abiotic and biotic conditions. While precipitation may not directly affect the WNV cycle, rainfall

interacts with other factors such as the landscape and temperature to create favorable conditions for mosquito productivity and can thusly mediate WNV transmission.

1.4.3 Landscape

In addition to weather influencing the spatial and temporal patterns of WNV transmission, many other physical features of the landscape are known to have significant associations with WNV transmission. Specifically, the landscape can determine the available microhabitats required for the survival of immature mosquitoes. The interactions between the landscape and climate allow localized increases in the abundance of certain species of mosquitoes that emerge as the predominant vectors of WNV in addition to their competency for amplifying the virus. For example, *Cx. quinquefasciatus* prefer water habitats with high organic content that is characteristic of human-modified urban and peridomestic areas, but *Cx. tarsalis* is most abundant in rural landscapes dominated by grasslands, pasture, and hay (Hribar et al. 2001, Bolling et al. 2005, Gibbs et al. 2006, Molaei et al. 2007, Nielsen et al. 2008, Reisen et al. 2008c, Winters et al. 2008, Reisen et al. 2009, Eisen et al. 2010, Chuang et al. 2011, Hahn et al. 2015). The most common features of the environment that have been studied and are capable of influencing mosquito and bird communities include vegetation/Normalized Difference Vegetation Index (NDVI), elevation, land type/land use, development/urbanization, and impervious surfaces (Ruiz et al. 2004, Bian et al. 2006, Cooke et al. 2006, Gibbs et al. 2006, Ozdenerol et al. 2008, Chuang et al. 2011, DeGroote and Sugumaran 2012, Degroote et al. 2014). Given their potential roles in the WNV transmission cycle based on the following studies, these factors will be considered in the dissertation.

Vegetation provides potential habitats for the mosquito and avian populations and is commonly measured by NDVI. NDVI is a normalized ratio of red and near infrared wavelengths to produce a metric of vegetation density (Lillesand et al. 2014). Brownstein et al. (2002) and Ruiz et al. (2004) concluded that the presence or increased coverage with vegetation (higher NDVI) are important risk factors for the incidence of WNV cases in which greater amounts of vegetation is required for avian host and mosquito habitats. Similarly, NDVI may be used to predict foci of WNV transmission to horses (Ward et al. 2005). However, a study in Iowa identified NDVI as a negative predictor of human WNV cases (DeGroote et al. 2008). Another study focused on Amherst, NY found a negative relationship between NDVI and *Cx. pipiens-restuans* abundance, but NDVI was not a significant predictor of *Aedes vexans* abundance (Trawinski and Mackay 2010). The differences in results from the various studies could be attributed to the varying preferences in habitats for each mosquito species and the ability for vegetation to mediate climatic factors based on the amount of vegetation available.

In combination with other physiographic elements, higher elevation may limit WNV transmission. Higher elevations will have lower temperatures, subsequent smaller mosquito abundances, and diversity in avian species composition compared to counterparts at lower elevations (Gibbs et al. 2006). Ruiz et al. (2010) found that elevation was a moderate predictor in determining the MIR of WNV in *Cx. pipiens* in Chicago, IL and suggested that elevation could mediate the effect of precipitation by collecting water at lower elevations to enrich with organic materials needed for mosquito population survival. Similarly, a study in North Dakota found an inverse relationship between the incidence of WNV in horses and elevation (Mongoh et al. 2007). In South Dakota, elevation had a negative relationship with human WNV risk, citing the

main vector's (*Cx. tarsalis*) inability to survive colder temperatures found at higher elevations as the reasoning for the decreased odds of human WNV disease (Chuang et al. 2012).

The land surface types and amount of urbanization or development may also mediate the mosquito population and human cases. Even though the literature examining the effect of land type and urbanization on *Cx. quinquefasciatus* development and WNV has been limited, extensive work has been conducted to identify the landscape factors involved in the development of *Cx. pipiens*, the northern WNV vector, and a species that can produce hybrid offspring with *Cx. quinquefasciatus* due to their similar biologies and ecologies (Farajollahi et al. 2011). In the northeastern U.S., WNV activity in mosquitoes and humans typically occurs in urban areas (Andreadis et al. 2004). The land types that impact the mosquito life cycle and transmission of the virus is dependent on the preference of the prominent mosquito species in the region. The *Cx. pipiens* complex dominates in the northeastern and southern U.S., while *Cx. tarsalis* is the suggested WNV vector in the western U.S., which could explain why human disease incidence of WNV is positively correlated with urban areas in the northeast and agricultural land in the west (Bowden et al. 2011). Furthermore, *Cx. quinquefasciatus* breed in urban areas, particularly near human habitations, urban catch basins, and storm drains (Molaei et al. 2007, Andreadis 2012). However, *Cx. tarsalis* prefer agricultural and rural landscapes (Reisen and Reeves 1990, Chuang et al. 2011, Andreadis 2012). The differences in the preferred habitats for multiple species of mosquitoes could also be due to the presence of unique primary hosts that are preferred by various species of mosquitoes, different survival rates against competitors, oviposition preferences for certain attractants that indicate the kind of organic material in aquatic habitats, or predilections for certain climates that allow a species to survive and become the dominant WNV vector in its region (Du and Millar 1999, Walton et al. 2009).

The association between urban landscapes and human cases indicates land use as a capable environment for enzootic and bridge transmission. Furthermore, some mosquitoes such as those in the *Cx. pipiens* complex have adapted to human-altered environments, thereby increasing exposure and transmission of medically-relevant pathogens to human and peridomestic animal populations (Farajollahi et al. 2011). Urbanization is a risk factor for human WNV incidence in the northeastern U.S. since case rates are significantly higher in urban classifications (Ruiz et al. 2007, Brown et al. 2008b), and it is likely this is also true in the southern U.S. given the similarities in habitat preferences between the main vectors of WNV in the northeastern (*Cx. pipiens*) and southern U.S. (*Cx. quinquefasciatus*) (Molaei et al. 2007, Andreadis 2012). In a comparison study between Detroit, MI and Chicago, IL, the rate of human WNV cases was higher in urban areas with moderate population density (Ruiz et al. 2007). Elsewhere, increased urbanization has been linked to increased WNV activity such as in California (Harrigan et al. 2010). Other measures of urbanization related to greater WNV human risk may include greater road density, less forested areas, or more built-up area/impervious surfaces (Brown et al. 2008b, LaBeaud et al. 2008).

In addition to urban land cover as a risk factor for WNV activity, another measurement of anthropogenic activity is impervious surfaces. Impervious surfaces represent the amount of human-made materials, which can be used to estimate the degree of artificial structures and urban land use (Arnold Jr and Gibbons 1996). A study in Wisconsin found that impervious surfaces may contribute to the collection of water in catch basins since these surfaces cannot absorb water, which may lead to greater abundances of mosquito larval habitats (Kronenwetter-Koepel et al. 2005). Two studies in Chicago, IL found conflicting results regarding impervious surfaces. Ruiz et al. (2010) reported impervious surfaces as a moderate predictor of WNV MIR,

but Messina et al. (2011) did not find a significant relationship between impervious surfaces and human cases of WNV for the same area. The differences between these two studies for the same area could be attributed to the differences in their outcome variables. The former used the mosquito MIR as the outcome measurement, while the latter study used human cases. Therefore, the differences in the conclusions from both of these studies could be due to the uncoupling between mosquito infection and the appearance of human infection with WNV.

Other landscape factors with a positive association with WNV cases include: soil water availability/poor soil drainage, pastures, surface water, slow-moving water sources, urban catch basins, roadside ditches, sewage treatment lagoons, the presence of manmade containers around the house, rural areas with wetlands, woodlands and arid shrubland, and availability of open water (Epstein 2001, Anderson et al. 2006, Ruiz et al. 2007, DeGroote and Sugumaran 2012, Nolan et al. 2012, Degroote et al. 2014). These factors play a role in the *Culex* mosquito development cycle or serve to attract avian hosts for resting and nesting periods, which establishes the enzootic cycle and amplifies viremic activity (Marra et al. 2004b).

1.4.4 Demographics

Social demographic predictors of human infection include socioeconomic status/education level, median age of the population, and race (Brownstein et al. 2002, Ruiz et al. 2004, Diuk-Wasser et al. 2006, Rios et al. 2006, Reisen et al. 2008a, Bisanzio et al. 2011, Rochlin et al. 2011). Demographic and social factors may indirectly affect the mosquito life cycle due to environmental factors or certain behaviors related to specific populations of people that may put them at higher risk for exposure (Ruiz et al. 2004).

Socioeconomic status (SES) is a single measurement that combines income, education, and occupation to compare the relative economic and social position of one person or family. Socioeconomic status and the individual components of SES are considered risk factors for different measures of WNV. For example, low income may affect someone's ability to get diagnosed and treated for WNV or poor housing may create unintentional habitats for *Culex* mosquitoes. The condition of the neighborhood is indicative of SES and may be an indicator of WNV prevalence. That is, low-income areas may have less upkeep of properties, which may create microhabitats for vectors and hosts to accumulate, or have different human behavioral responses to WNV due to different education levels compared to higher-income areas (Harrigan et al. 2010). In Tennessee, areas with low income, a high percentage of rental occupations, and more vacant housing were identified as social drivers of WNV transmission (Ozdenerol et al. 2008). When observing mosquito abundances, a lower SES was not a significant predictor (Dowling et al. 2013). However, the potential for larval mosquito habitats exists in areas with lower SES since there are greater numbers of containers capable of forming mosquito habitats and these homes are less likely to be maintained by homeowners (i.e., remove or apply treatments to standing water) (Dowling et al. 2013).

While low SES may be associated with higher WNV prevalence, not all areas have reported similar results. During the 2002 WNV outbreak in Chicago IL, higher median incomes were more likely to be found in tracts inside clusters of human WNV cases (Ruiz et al. 2004). Nearby in Ohio, higher-income areas were also associated with greater risk of human infection (LaBeaud et al. 2008). In a national study looking at the 2012 WNV outbreak, median income was positively associated with their binary county WNV rating (higher/lower rate in the county compared to the regional 2012 rate) in the northcentral and northeastern regions of the U.S., but

the percentage of those in poverty (another type of income measurement) was negatively associated with the WNV ratings in the southeast region that encompasses major metropolitan areas such as Dallas, Harris, Bexar, and Travis Counties (Degroote et al. 2014). The difference in significant predictors in various regions could be due to landscape heterogeneity and the variations in the ecology of the predominant mosquito vector.

Changes in the economy should also be taken into account when looking at the status of high- and low-income areas in relation to WNV levels. Census tracts with higher income and a greater number of unoccupied houses were associated with greater WNV occurrence during the 2012 Dallas County outbreak (Chung et al. 2013). Unoccupied houses or foreclosed houses as a result of the economic downturn in the 2000's may have contributed to unmaintained swimming pools that could form potential habitats for *Culex* mosquitoes by collecting stagnant rainwater (Reisen et al. 2008a, Chung et al. 2013).

Education is positively correlated with income and can measure another aspect of SES (Milligan et al. 2004). Those with higher education levels are more likely to be more politically engaged and involved in community issues (Milligan et al. 2004). Demanding mosquito control, maintaining homes to prevent mosquito habitation, or having the willingness to educate themselves on mosquito control and prevention may reduce mosquito abundance and WNV infection in homes (Harrigan et al. 2010). However, the negative correlation between WNV activity and education, income, or SES is not consistent everywhere. Education level as a measurement of SES has also been positively linked to WNV human risk in New York, where higher education levels were associated with greater WNV human risk (Rochlin et al. 2011). These populations of educated people may be more apt to act on their symptoms or have more access to medical services, both of which may have biased the number of reported cases towards

those with higher education levels (Rochlin et al. 2011). Other obstacles in preventing and controlling WNV in areas with higher education, income, or SES include active protesting of mosquito control efforts or living in gated communities that may not allow proper mosquito spraying at night.

In Houston, TX, there were no significant relationships between arbovirus activity and various SES measurements such as education level, median household income, and families living below the poverty level during the WNV outbreak in 2002 (Rios et al. 2006). Yet, when spatially comparing communities that did and did not have WNV activity, the authors concluded that 73% of the census tracts with arbovirus activity were located in areas with lower SES levels measured by lower educational attainment and greater poverty levels (Rios et al. 2006). While the study did not find significant relationships between the presence of WNV and SES, it should be noted that the authors focused on activity within the I-610 highway loop, which is heavily populated and was the focus of mosquito surveillance when WNV first appeared in the city.

Few studies have delved into the relationship between median age of the human population and WNV occurrence measured by the presence in mosquito populations, but clinically, WNV is known to affect older populations. In 2016, WNV affected older populations with a median age of 57 among the cases reported to the CDC (Burakoff et al. 2018). During the 2002 outbreak of WNV in Chicago, IL, a study found that older ages were associated with clusters of WNV incidence in humans and dead birds (Ruiz et al. 2004). Specific age groups may have higher incidences of WNV depending on certain behaviors that might put these groups at higher risk. For example, practices that older people may participate in such as yard work or constructing yard structures that collect water may contribute to greater mosquito abundances and bring competent hosts together to circulate WNV. Outdoor activities that are frequent among

older populations such as gardening will more likely expose them to mosquitoes and WNV (Blaine et al. 2010). Alternatively, another study looked at WNV on a broader scope and investigated relationships between WNV incidence rates throughout the U.S. and found inconsistent results depending on the region (DeGroote and Sugumaran 2012). They found a significant positive relationship between median age and WNV incidence in the Great Plains region and a significant negative relationship in the northwest region (DeGroote and Sugumaran 2012). In the southeast region, which includes Harris County, TX, median age also had a significant negative relationship, where younger populations reported higher incidence rates (DeGroote and Sugumaran 2012). For the remaining regions, median age did not have a significant relationship with WNV incidence rates (DeGroote and Sugumaran 2012).

Race and ethnicity is yet another demographic variable related to the occurrence of WNV. In Chicago, IL and Shelby, TN, greater percentages of black populations were associated with greater MIR and human risk of WNV, respectively (Ozdenerol et al. 2008, Ruiz et al. 2010). Other studies from Chicago, IL have found that higher risk of human WNV incidence rather than mosquito infection rates were associated with greater percentages of whites (Ruiz et al. 2004, Messina et al. 2011). Like other demographic factors, the associated culture and behaviors, as well as the environmental context mediate the associations between race and ethnicity with WNV occurrence.

White and black populations have been used as the reference population in many studies, but studies focused on Texas also need to account for the large percentage of Hispanics given the state's close proximity to Central and South America. A national study investigated the role of landscape and demography on incidence of human WNV cases between 2002-2009 (DeGroote and Sugumaran 2012). Their study concluded that the percentage of Hispanics was a significant

factor in WNV human incidence in the northwest and southeast regions, but these regions had positive and negative associations, respectively (DeGroote and Sugumaran 2012). While few studies have analyzed the relationship between Hispanic populations and WNV occurrence, studies investigating the association between Hispanics and Dengue have contributed to the understanding of the Hispanic population's role in the transmission of mosquito-borne viruses. A study in north Texas found that Hispanic populations were associated with higher numbers of containers, which can provide artificial habitats for Dengue vectors *Ae. aegypti* and *Ae. albopictus* (Lee et al. 2009). *Cx. quinquefasciatus* may also use similar habitats if stagnant rainwater collect and enrich in the containers. Furthermore, avian populations may seek this collected water as a source of hydration. The resulting interactions between avian hosts and mosquitoes at man-made water sources can circulate WNV in a localized area. Previously in Houston, TX, a study found that the greater number of artificial containers that can collect water was only suggestive of mosquito density, but was a significant predictor for WNV and SLE virus activity within virus-positive census tracts (Rios et al. 2006).

Much research has already been conducted on the abiotic factors involved in the transmission cycle of WNV but many of these attempts have not been focused on Texas or the main vector of WNV in the southern U.S. After the 2012 WNV epidemic with Dallas as the epicenter of this event, it is crucial for Texas to follow the steps of other states and utilize quantitative metrics to define when and where to initiate mosquito control tactics.

1.5 Quantitative Surveillance Measurements of West Nile Virus in Mosquitoes

Studies investigating determinants of WNV transmission often use human incidence as the measure of infection (Ruiz et al. 2004, DeGroote et al. 2008, DeGroote and Sugumaran 2012,

Chung et al. 2013, Murray et al. 2013, Degroote et al. 2014). As a reportable disease in the U.S., WNV human case data is readily available and organized by the CDC on ArboNET and other servers. However, transmission of arboviruses in the enzootic cycle between mosquitoes and wild amplification hosts does not always correlate with human disease (Eldridge, 1995). In this regard, there are likely diverse mechanisms for the uncoupling of human disease from the enzootic cycle. For example, reporting biases may be a non-biological factor with different demographics having a variable propensity to get tested (Bustamante and Lord 2010, Ruiz et al. 2010). Changes in case definition and reporting practices, limitations in finding the location of exposure, and the absence of information about the enzootic cycle are other challenges that should be considered when using epidemiological data for WNV (Eisen and Eisen 2008). Given that a component of arbovirus management aims to conduct mosquito abatement to minimize vector populations and dampen virus transmission in the enzootic cycle, it is critical to have an in-depth understanding of how biotic and abiotic factors relate to the enzootic cycle of WNV, in which the abundance of the vector, mosquito infection rate, and vector index are examples of such metrics. In addition, virus activity in mosquitoes and birds precedes human exposure (Hamer et al. 2008b), thereby offering an early warning system to allow intervention strategies to protect public health (Andreadis et al. 2004). Therefore, mosquito data becomes a useful and more neutral measure in determining the risk of WNV infection in humans (Brownstein et al. 2002, Ruiz et al. 2010). Several measurements estimating mosquito species abundance and infection status exist and are recommended by the CDC. Specifically, our studies will focus on three main measurements: abundance of vectors, infection rates, and the vector index.

Vector abundance refers to the number of mosquitoes collected over the number of nights used for trapping and is expressed as the number of mosquitoes per trap night (CDC 2013).

Vector abundances can be determined by the following:

$$N_i = \frac{m_i}{n_i} \quad (1.1)$$

where N is the abundance of species i , m is the number of mosquitoes trapped, and n is the number of successful trap nights. Trap malfunctions are not included in the denominator of this calculation. A large vector abundance can act as a threshold to begin vector control and monitor vector control efforts. However, high abundances of mosquitoes may occur without the presence of virus amplification, indicating a lag between the time WNV first appears in the mosquito population and a detectable level of WNV. The virus first needs to circulate through the avian population to create a high enough viremia to infect subsequent mosquitoes. Several studies have utilized the vector abundance as an outcome variable to identify likely mosquito habitats, relate landscape and demographic variables to vector abundance, and predict where mosquito populations may appear in a neighboring district (Diuk-Wasser et al. 2006, Reiter and LaPointe 2007, Walsh et al. 2007, Deichmeister and Telang 2010, Trawinski and Mackay 2010, Chuang et al. 2011, Karki et al. 2016). Calculating the vector abundance is useful to relate mosquito presence in an area based on a set of criteria, but the abundance does not consider WNV presence. The abundance is therefore limited in the breadth of conclusions regarding the WNV presence because the abundance of vectors is not always equivalent to the amount of WNV circulating in a population of mosquitoes.

Infection rates estimate the number of WNV-positive mosquitoes in a population (CDC 2013). The infection rate can be measured in two different ways: MIR or maximum likelihood estimate (MLE). The MIR is calculated as the following:

$$P_{MIR} = \frac{X}{y} \times 1,000 \quad (1.2)$$

where X represents the number of positive pools and y represents the total number of mosquitoes tested (Condotta et al. 2004). This measurement assumes that only one mosquito is positive in a positive pool (CDC 2013). This assumption is 99% likely when MIRs < 3 for mosquito pools of 50, but MIRs > 3 may result in underestimates of the true prevalence of WNV in mosquito pools (Bernard et al. 2001, Kilpatrick et al. 2005a). Conversely, the MLE does not make the assumption of one positive mosquito per positive pool and is the preferred measurement during outbreaks (Gu et al. 2008, CDC 2013). The MLE is calculated as:

$$P_{MLE} = 1 - \left(p - \frac{X}{p}\right)^{\frac{1}{q}} \quad (1.3)$$

where p represents the number of pools tested, X is the number of positive pools, and q is the pool size (Condotta et al. 2004). Regardless of which measurement is used, the infection rate provides a quantitative basis for comparing WNV outcomes across time, space, and population size. Unlike the vector abundance, the infection rate can be used to relate vector infection to human risk (Nasci et al. 2001b, Bell et al. 2005). Ruiz et al. (2010) used the MIR to measure the presence of WNV in Chicago, IL because of possible demographic biases related to human case reporting. They were also interested in climate, environmental, and demographic patterns that

relate to the ecology of *Cx. pipiens* and the temporal heterogeneity of WNV (Ruiz et al. 2010). The infection rate becomes valuable when sampling data are incomplete or spatially unequal since the calculations only require data about pooled mosquitoes instead of the complete population collected during surveillance activities. Consequently, conclusions cannot be inferred regarding the abundance of a vector if they only had laboratory/testing data and the calculated infection rates. The infection rate may also be over- or under-estimated for some species of mosquitoes if multiple species are combined into one pool, which is problematic when calculating infection rates for certain *Culex* species (Nasci et al. 2001b). Some species of *Culex* mosquitoes such as *Cx. restuans* or those in the *Cx. pipiens* complex are morphologically indistinguishable and have similar distributions in the U.S., making separation of the species into different pools difficult and therefore producing inaccurate infection rates (Harrington and Poulson 2008). Depending on the species, the calculated infection rates may underestimate the true infection rate if the species is commonly found in the study area or vice versa.

The vector index (VI) is another measurement of WNV relating the vector to the presence of WNV. Simply, the VI takes into account species composition, population density, and infection rate to form a single measurement of the amount of infected mosquitoes that are collected per trap night (Nasci et al. 2005, Gujral et al. 2007). The infection rate and the abundance of vectors are then multiplied to form the VI for each species and then the VI's are summed for all species present to form a single estimate of the abundance of infected vectors, making it especially useful when there is more than one dominant vector for WNV in an area (CDC 2013). The VI can be expressed as the following:

$$VI = \sum_i N_i P_i \quad (1.4)$$

where N is the abundance of mosquito species i (calculated from equation 1.1), and P represents the estimated IR for a mosquito species. The VI has previously been used to relate human incidence of WNV and has demonstrated greater predictive ability than the individual parts of the index (Bolling et al. 2009a, Bolling et al. 2009b, Jones et al. 2011, Kwan et al. 2012, Colborn et al. 2013). Furthermore, this measurement is currently used in public health and vector control programs such as the 2018 Arboviral Response Plan in Colorado (Colorado Department of Public Health and Environment 2018). To use the VI, the infection rate and vector abundance must be available. However, if the data from either of these measurements were compromised or a level of uncertainty exists in the data quality, then the VI should not be used. Regardless, the VI will determine the level of WNV infection in the population of mosquitoes instead of being restricted to the population of mosquitoes that were tested in the laboratory like the infection rate.

Differences in the type of outcome variable make it difficult to compare studies across different regions of the U.S., including studies that were mentioned and compared throughout this review. Interpretation and comparison of the conclusions from multiple studies should proceed with caution regarding the outcome measurement. For example, using the mosquito abundance as the outcome variable is a valid method; however, one cannot interpret high abundances as an equivalent for increased numbers of cases of WNV.

Nevertheless, mosquito surveillance data can easily be transformed into quantitative measures of WNV activity. Choosing the appropriate quantitative measurement is based on the quality of the dataset itself, how the data were collected, and the purposes for using these values. Each of the quantifiable measurements of WNV in mosquitoes have unique weaknesses;

however, the overall utility of these surveillance tools allow vector control programs to fully utilize their surveillance data in order to develop quantifiable thresholds and aid in administrative decisions for proactive and effective vector control.

1.6 Project Rationale

Without an available vaccine for humans, efficient mosquito control and public awareness campaigns are currently the most effective ways to prevent and/or minimize WNV human cases. Understanding the ecology of WNV dynamics with the mosquito, climate, landscape, and social demographics plays a key role in effective intervention campaigns. Furthermore, predicting when and where WNV may occur will provide an early warning system and the opportunity to control mosquitoes before bridge transmission to humans, as well as alert the public with the appropriate messages to reduce the risk of exposure.

Given the progress of understanding the relationships between meteorological, environmental, and demographic features of the landscape and WNV transmission, several studies have utilized these variables into surveillance strategies to predict future transmission potential. Predictive models are powerful tools because they can inform management decisions to be as efficient as possible with limited resources. Furthermore, predictive models are most cost-effective in comparison to blanket or prophylactic mosquito control measures, making predictive models a key component of surveillance and prevention programs (Reisen 2010). Several states have surveillance systems using a combination of meteorological and surveillance data to guide quantitative decision thresholds (Brown 2012, Geographic Information Science and Spatial Epidemiology 2015).

Although surveillance systems based on quantitative models to establish decision thresholds exist in certain regions of the U.S., these predictive models need to be parameterized with historic data from the same geographic region. While Texas has several mosquito and mosquito-borne disease surveillance and control programs, the data generated have not been optimally utilized to fully understand the abiotic and biotic factors influencing the spatial and temporal variation in WNV transmission in Texas (Chung et al. 2013). Furthermore, the predictors of WNV transmission have not been developed into predictive models catered for Texas. The goal of this dissertation is to fill this void and utilize historic mosquito and mosquito WNV infection data to build a predictive model relevant to Texas based on climate, landscape, and demographic features.

Once these significant factors are identified, local mosquito control agencies in Texas can integrate these models into their surveillance program or develop their own models based on the procedures described in this dissertation. Texas has fallen behind in mosquito surveillance and control compared to other states such as California and Illinois, which currently use models and quantitative thresholds as the foundation for decisions regarding mosquito control. Furthermore, Texas is a potential hotspot for new arboviruses due to the adjacency of Latin and South America, where many vector-borne diseases may be more prevalent. Because of the lack of research on WNV modeling in Texas and the potential risk for new arboviruses to establish in Texas, developing the models relevant to major metropolitan areas in Texas will put the state closer to the forefront of mosquito management and prepare the state against new threats.

1.7 Objectives

We collaborated with county and municipal public health agencies to utilize historic mosquito trapping and infection data to investigate spatial and temporal patterns of WNV infection as well as the meteorological, environmental, and demographic determinants of WNV.

This dissertation has three objectives:

1. Describe the landscape and demographic determinants affecting WNV dynamics during the 2012 WNV epidemic in Dallas County, Texas.
2. Investigate socioeconomic and environmental landscapes affecting WNV spatial patterns in Harris County, Texas.
3. Identify associations between prior weather and WNV infection in *Cx. quinquefasciatus* mosquitoes in Harris County, Texas.

We identified significant factors related to how WNV spatially and temporally differs from year to year in Texas. The predictive models created from this project will ideally estimate and anticipate the risk of WNV transmission in real-time.

CHAPTER II

LANDSCAPE AND DEMOGRAPHIC DETERMINANTS OF *CULEX* (DIPTERA:
CULICIDAE) INFECTION WITH WEST NILE VIRUS DURING THE 2012 EPIDEMIC IN
DALLAS COUNTY, TEXAS

2.1 Synopsis

In 2012, the U.S. experienced the largest outbreak of West Nile virus (WNV), with the majority of cases and deaths occurring in Texas. Texas reported 1,024 cases of WNV fever, 844 cases of WNV neuroinvasive disease, and 89 deaths throughout the state, with the majority of incidents occurring in Dallas, TX and surrounding areas. Previous studies explored relationships between human cases of WNV and demographic and landscape variables; however, the infection of mosquitoes may better reflect spatial variation in transmission intensity than human cases. In this study, we identified associations between features of the landscape and human population and *Culex quinquefasciatus* infection with WNV during the 2012 WNV epidemic in Dallas County. Using logistic linear mixed models, we modeled the infection rate of WNV in *Cx. quinquefasciatus* based on environmental variables and social demographics. During this epidemic, 25,917 female mosquitoes were organized into pools of ≤ 50 mosquitoes each and tested for WNV, of which 22,156 *Cx. quinquefasciatus* mosquitoes were identified. Out of 1,634 pools containing at least one *Cx. quinquefasciatus* mosquito, 256 pools (15.7%) tested positive for WNV. *Cx. quinquefasciatus* pools accounted for 96% of the positive pools in 2012. Major mosquito and WNV activity occurred between May and September, with a peak in the infection rate during the third week of July (47.7 per 1,000). We found increased probabilities for WNV-positive mosquitoes in north and central Dallas County. Based on the best-fit model, the most

significant predictors of the presence of WNV in *Cx. quinquefasciatus* pools were increased urbanization (based on an index composed of greater population density, lower normalized difference vegetation index, higher coverage of urban land types, and more impervious surfaces), lower elevation, and older populations. The best-fit model identifies key environmental and demographic factors that play a role in the mosquito's life cycle and the ability to obtain the virus during the 2012 WNV epidemic in Dallas County, TX. These relationships between the landscape and risk of enzootic transmission help to identify spatial regions of the landscape with highest risk of spillover to human disease.

2.2 Introduction

West Nile virus (WNV) is a zoonotic disease that circulates in an enzootic cycle between avian hosts and mosquitoes (Weaver and Reisen 2010). The virus amplifies within the avian host that maintains a sufficient viremia to infect other mosquitoes. Occasionally, WNV is transmitted outside this enzootic cycle to dead-end hosts like horses and humans. *Culex* mosquitoes are the primary enzootic vectors for WNV in North America (Turell et al. 2005, Colton and Nasci 2006, Weaver and Reisen 2010), as well as capable bridge vectors to humans (Hribar et al. 2001, Kilpatrick et al. 2005b, Hamer et al. 2008b). In the southeast U.S., including Texas, *Culex quinquefasciatus* is the major mosquito vector in the transmission cycle (Lillibridge et al. 2004, Molaei et al. 2007, Andreadis 2012). *Cx. quinquefasciatus* is an opportunistic feeder that primarily feeds on birds, but also takes frequent blood meals from humans and other mammals and has a high abundance during the WNV transmission season (summer through early fall) (Savage and Miller 1995, Gibbs et al. 2006, Molaei et al. 2007, Richards et al. 2007, Reisen et al. 2008b). *Cx. quinquefasciatus* prefer water habitats with high organic content, which is

characteristic of human-modified urban and peridomestic areas (Reisen et al. 1992, Hribar et al. 2001, Bolling et al. 2005, Gibbs et al. 2006, Reiter and LaPointe 2007, Reisen et al. 2008c, Hongoh et al. 2009, Hahn et al. 2015).

The occurrence and distribution of WNV depends on the presence of competent mosquito vectors, available susceptible avian hosts, and their potential interactions, all of which are influenced by various aspects in the landscape and anthropogenic behaviors (Epstein and Defilippo 2001, Ruiz et al. 2004, Kuhn et al. 2005, DeGroot and Sugumaran 2012, Degroot et al. 2014). The features of the environment capable of influencing mosquito and bird communities include elevation, vegetation (Normalized Difference Vegetation Index [NDVI]), and impervious surfaces. Physical features of the landscape mediate climate events to provide potential habitats for immature development of *Cx. quinquefasciatus*. For example, water may pool at lower elevations following rain events where impervious surfaces are present and consequently water cannot be absorbed (Ruiz et al. 2010). Furthermore, runoff may collect in underground storm drains after a rain event, leading to greater numbers of *Culex* populations in underground storm drains, which are common in urban areas such as Orange County, CA (Su et al. 2003). Urbanization can alter the climate within cities to form “heat islands,” where temperatures increase in areas with higher urbanization leading to enrichment of standing water to promote the life cycle of the mosquito (Reisen 2010). Social demographic predictors of human WNV infection include median age of the population, race, socioeconomic status, and age of the neighborhood (Brownstein et al. 2002, Ruiz et al. 2004, Diuk-Wasser et al. 2006, Bisanzio et al. 2011). These factors and behaviors associated with certain populations may additionally contribute to potential habitats for mosquitoes and avian hosts.

The magnitude of the 2012 WNV epidemic was unexpected given that the original introduction of WNV into the region occurred a decade previously and WNV activity was generally low leading up to the epidemic. In 2012, the U.S. reported 5,674 human cases, 627 equine cases, and 286 human deaths (CDC 2014, United States Department of Agriculture Animal and Plant Health Inspection Service 2018). Texas reported a total of 1,868 human cases, 120 equine cases, and 89 human deaths (CDC 2014, United States Department of Agriculture Animal and Plant Health Inspection Service 2018). The epidemic was centered in Dallas County, which reported 396 human cases, 3 equine cases, and 19 deaths (CDC 2014, United States Department of Agriculture Animal and Plant Health Inspection Service 2018). The total economic cost was approximately \$47.6 million for vector control and hospital management in Texas (Chung et al. 2013, Murray et al. 2013). Dallas County was an outlier in 2012, reporting the greatest number of cases in one of the largest epidemics of WNV after an extended period of moderate activity for reported human WNV cases.

Previous studies have explored the relationships between demographic and environmental variables with human WNV cases (DeGroote et al. 2008, Chuang et al. 2012, Degroote et al. 2014), but relatively few studies have explored these factors as they relate to WNV infection in *Culex* mosquitoes, especially in the southern U.S. Virus infection in mosquitoes is more reflective of the WNV enzootic cycle and is inherently less biased than human case data given artifacts from reporting and diagnosis biases (Eisen and Eisen 2008). Accordingly, understanding spatial patterns of mosquito infection provides an opportunity to identify predictors of spatial variation in the amount of virus in the WNV enzootic cycle (Eisen and Eisen 2008). While spatial heterogeneity of WNV infection in mosquitoes has been studied previously in other states such as Illinois and California, this topic remains unexplored in the

state of Texas, especially with regard to the local epidemic in 2012 (Ruiz et al. 2004, Reisen et al. 2008a).

Given the progress of understanding the relationships between environmental and demographic features of the landscape and WNV transmission, several studies have incorporated these variables into surveillance strategies to predict future transmission potential (Ruiz et al. 2004, Ruiz et al. 2007, Reisen et al. 2008a, Degroote et al. 2014, Shand et al. 2016). Predictive models are powerful tools because they can inform management decisions to be as efficient as possible with limited resources. Furthermore, predictive models are most cost-effective in comparison to mosquito control measures, making predictive models a key component of surveillance and prevention programs (Reisen 2010). Several states have surveillance systems using a combination of meteorological, environmental, and surveillance data to guide quantitative decision thresholds (Brown 2012, Geographic Information Science and Spatial Epidemiology 2015).

Although surveillance systems based on quantitative models to establish decision thresholds exist in certain regions of the U.S., these predictive models need to be parameterized with historic data from the same geographic region. While Texas has several mosquito and mosquito-borne disease surveillance and control programs, the data generated have not been utilized to fully understand the abiotic and biotic factors influencing the spatial and temporal variation in WNV transmission in Texas. Additionally, the predictors of WNV transmission have not been developed into predictive models catered for Texas. Studies that have focused on the 2012 WNV epidemic have not focused on Dallas County, TX as the center of their research. For example, Degroote et al. (2014) investigated associations between human incidence of WNV during the 2012 outbreak and factors of the landscape, climate, and human population across the

coterminous U.S. to understand outbreak dynamics across varying ecological environments. The authors divided the U.S. into regions based on the distributions of the suspected major WNV vectors in their respective region (Degroote et al. 2014). They found significant associations between human incidence and the risk factors, but these associations differed depending on the region and the ecologies of the dominant vector in the region (Degroote et al. 2014). Because the study undertook the task of conducting a national study, many of the findings were generalized for the encompassed region, which may underestimate the true association for counties that have higher incidences and have surrounding counties with low incidences.

Alternatively, other studies have concentrated on the clinical aspect of WNV rather than the features that might affect the enzootic cycle prior to human infection. Chung et al. (2013) focused on the epidemiology of human cases during the epidemic and how mosquito surveillance data (the vector index) could be used to predict the number of West Nile neuroinvasive disease (WNND). Clusters of WNND were located in areas with high housing density and the vector index followed a similar geospatial pattern (Chung et al. 2013). Furthermore, a hot-spot analysis on the WNND cases suggested that there were major hot-spots in the northern half of the county, a similar pattern that was seen during the 2002 and 2006 WNV outbreaks (Chung et al. 2013). This study investigated the vector index, a measurement derived from mosquito data, but the objective for using the vector index was to measure its predictive capability for human cases instead of identifying predictors for the vector index.

These two studies contributed to the general understanding of the 2012 WNV epidemic, but our objective in this study is to better understand the static landscape and demographic factors that might have played a role in the ecology of the mosquito vector during this epidemic in Dallas County, TX. Therefore, this study aims to describe the landscape and demographic

determinants affecting WNV dynamics during the 2012 WNV epidemic in Dallas County, Texas with a focus on the WNV enzootic vector *Cx. quinquefasciatus*. The models developed in this study can be integrated into mosquito surveillance programs as a cost-effective strategy for more directed control of WNV vectors and improved predictive capabilities of WNV outbreaks in Dallas County.

2.3 Methods

2.3.1 Mosquito Data

We compiled data from multiple entities in Dallas County (Figure 2). Mosquito trapping occurred weekly throughout Dallas County, using one gravid trap per location. A total of 506 unique trap locations were set throughout Dallas County in 2012 (Figure 2). Some trap locations had data for only one trap night and other trap locations were visited repeatedly up to 34 total visits between May and December. After the traps were collected, the mosquitoes were identified according to species and sex and then sorted into pools of ≤ 50 mosquitoes. Dallas County Health and Human Services and the Texas Department of State Health Services (DSHS) tested female mosquito pools for WNV using quantitative reverse-transcriptase polymerase chain reaction (qRT-PCR) and/or cell culture isolation. Dallas County performed qRT-PCR tests only. DSHS conducted qRT-PCR to detect WNV in the pool with a confirmation test using cell culture isolation.

To determine the temporal distribution of WNV throughout the epidemic, the infection rate (IR) per week was calculated using the Biggerstaff Pooled Infection Rate Excel add-in developed by the Centers for Disease Control and Prevention (Biggerstaff 2009). To investigate

the spatial distribution of the WNV IR for each trap location throughout the county, the IR per unique trap identification number was calculated using the same Excel add-in.

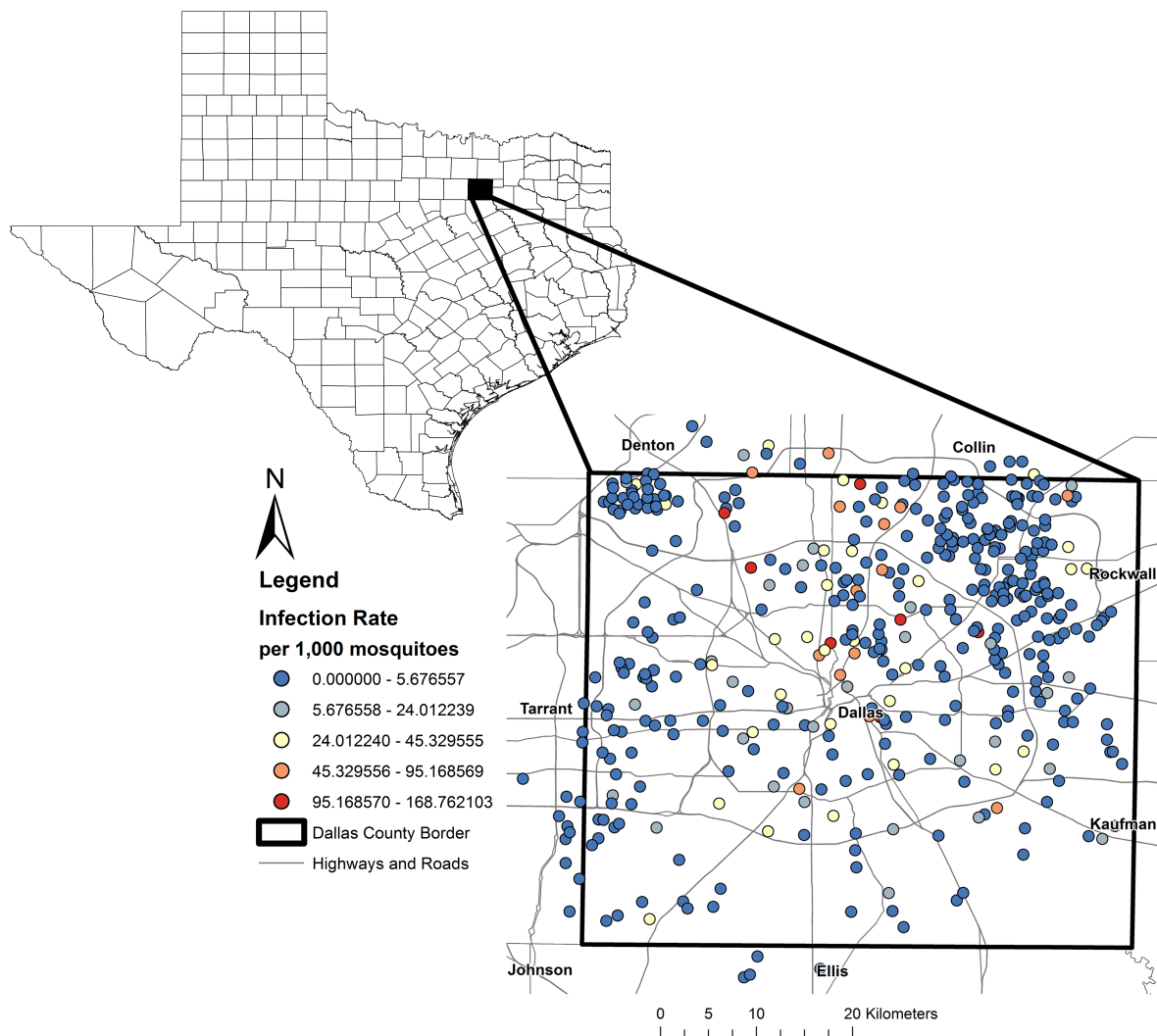


Figure 2 Map of Dallas County, TX and gravid trap locations within Dallas County. Point locations of the 506 trap locations in 2012 are shown with an infection rate (IR) estimated for each trap. The infection rate was summarized per trap location for 2012. Higher infection rates are located throughout central and north Dallas County.

2.3.2 Layer Processing

Mosquito trap data and landscape layers were organized and processed using ArcMap 10.2.2 (Esri, Redlands, CA). All layers were projected to NAD 1983 State Plane Texas North Central FIPS 4202 Feet. The raster layers were resampled to a resolution size of 250 m x 250 m.

Predictors from national and local databases were utilized to describe the environmental and socioeconomic landscapes (Table 1). These landscape and demographic variables have been previously investigated in prior studies (Ruiz et al. 2004, Cooke et al. 2006, Mongoh et al. 2007, Ruiz et al. 2007, DeGroote et al. 2008, Reisen et al. 2008a, Deichmeister and Telang 2010, Chuang et al. 2012, Chuang and Wimberly 2012, DeGroote and Sugumaran 2012, Degroote et al. 2014).

Landscape variables of interest included elevation, impervious surfaces, land cover, NDVI, and distance to water bodies. The land cover datasets were obtained from the Multi-Resolution Land Characteristics Consortium and the land cover classes were reclassified into developed land (urban, classes 21-24) versus non-developed (non-urban) land cover types. From this reclassification, a percentage of urban land cover within the buffer was used to represent the level of developed land around each trap. The dataset describing the water bodies in Dallas County, TX was gathered from the North Central Texas Council of Governments. Distance to water bodies describes the distance between center of the trap locations and the nearest water body. Demographic variables from the 2012 American Community Survey were measured at the census block group level. Demographic layers included median age, median income, and population density (Table 1).

A 750-m buffer was applied around each trap location to capture the physical and socioeconomic landscape around each trap. This buffer represents the approximate flight range

of *Cx. quinquefasciatus* mosquitoes (Medeiros et al. 2017). Information describing the environment and demographics within the buffer were extracted using ArcGIS.

Table 1 Predictor variables used in the model creation steps. We used variables in our model that describe the landscape and demographic of the population in Dallas County. The data for each variable were collected from their respective sources listed on the right.

Variables	Source
Digital elevation model (DEM)	National Elevation Database
Impervious surfaces Land cover	Multi-Resolution Land Characteristics Consortium (MRLC) (2011)
Normalized difference vegetation index (NDVI)	eMODIS (2012)
Distance to water bodies	North Central Texas Council of Governments
Demographic (census block groups) Median age Median income Population density	American Community Survey (2012)

2.3.3 Model Creation

A principal component analysis (PCA) was conducted due to an *a priori* prediction that many of the variables in Table 1 would be autocorrelated. To mitigate the risk of collinearity, a PCA approach was used to ensure that the different variables in the model were orthogonal. We created a PCA group called “urbanization” (*PCI*) that included variables related to the urban landscape: land cover, impervious surfaces, NDVI, and population density. The variables that comprise this PCA have individually been investigated as risk factors for WNV and *Culex* abundance (Cooke et al. 2006, DeGroote et al. 2008). Median population age and income were additionally included as demographic variables in the models, which have been previously

explored in relation to WNV (Ruiz et al. 2004, Degroote et al. 2014). Variables about the physical landscape such as distance to water bodies and elevation were included in the models as independent variables (Eisen et al. 2008, Nolan et al. 2012).

Logistic mixed models were used to model the probability of infection of mosquito pools. Trap week was included as a random effect to control for seasonal effects across the data. To correct for spatial autocorrelation, we used a nested random effect structure. Smaller blocks (4.60 km x 5.07 km per block) were nested within larger blocks (18.40 km x 20.29 km per block) across the study area ($\sim 3364.74 \text{ km}^2$), such that there were 4x4 small blocks in each large block and 3x3 large blocks encompassed the study area. Each trap location was given a unique group number within the large block and another unique group number representing the small block nested in the large block. This nested random effect structure is a blocking scheme that assumes the data points within the small blocks in the same large block are more similar to one another overall and that individuals from two different large blocks are less similar from one another, independent of the fixed effects across the landscape. Since the pools had a variable pool size of 1-50, an offset function was added to the model to control for the variance in pool size. The outcome variable for our models was the probability of a positive mosquito pool with a negative pool designated as 0 and a positive pool designated as 1. The dataset includes data during the peak season for *Cx. quinquefasciatus* mosquito and WNV activities in 2012, which corresponds to weeks 20-37 (Figure 3).

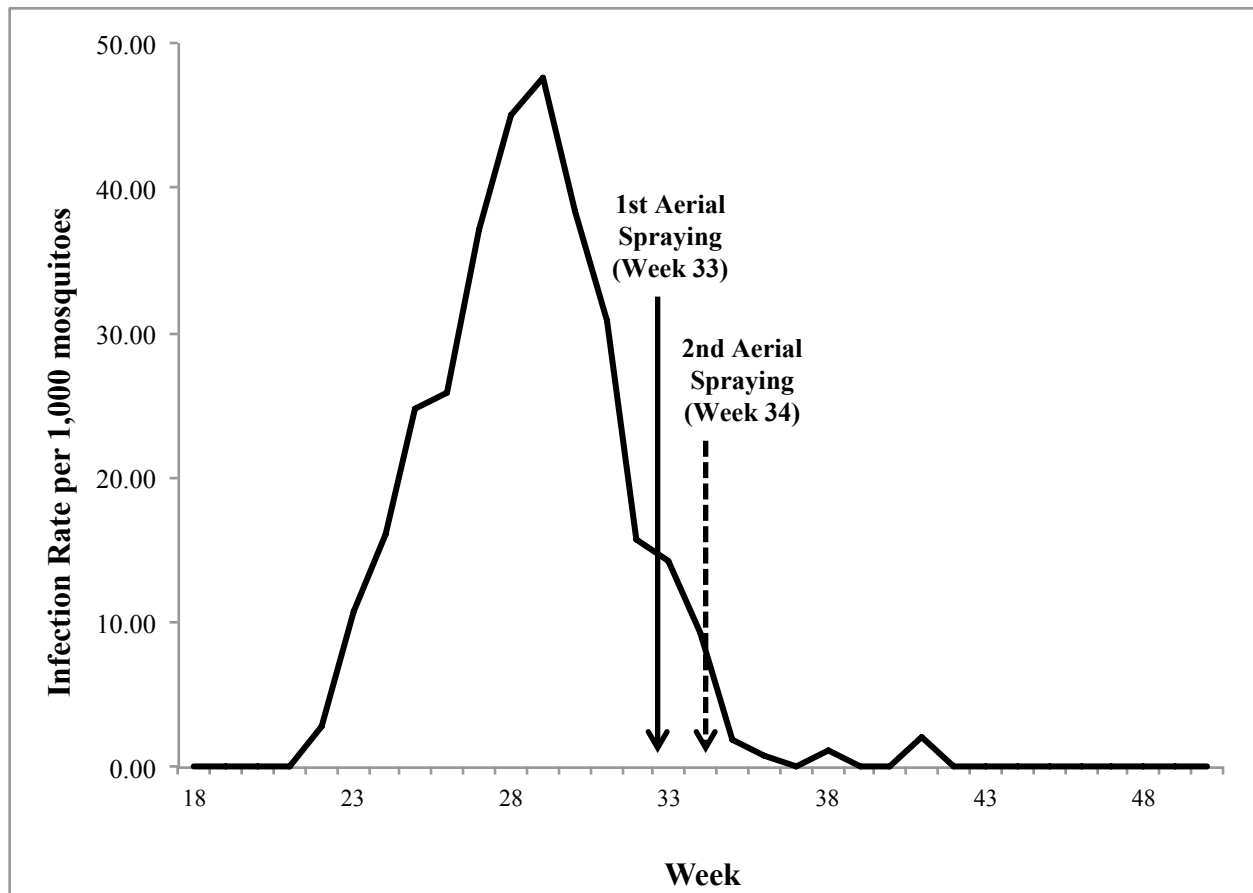


Figure 3 WNV infection rate in Dallas County, TX in 2012. Weeks 20-37 (May – September 2012) represent the time period with the majority of high WNV activity. The peak infection rate occurred in week 29 (July 16-22) with an infection rate of 47.66. The solid arrow represents the first aerial spraying for mosquitoes during week 33 (August 16-20) followed by a second spraying event (dashed arrow) in week 34 (August 22-23) in Dallas County.

Akaike's Information Criterion (AIC) corrected for small sample size and was used to distinguish a best-fit model from other candidate models, with the smallest AIC value representing a model with better fit. The AIC measures the relative quality of models for a given set of data to determine which model(s) fit the data best (Bozdogan 1987). Models were created using the *lme4* package in program R (Bates et al. 2015). A bootstrapping method was used to calculate 95% confidence intervals for each predictor estimate in the best-fitting model.

To generate a risk map describing the probability of finding a WNV-positive pool based on the coefficients of the best-fit model, the log link functions were entered into ArcGIS. We

derived the log-odds from the logistic regression using the following logit link (Diuk-Wasser et al. 2006, Peper et al. 2018):

$$\ln(\mu) = \beta_0 + \beta_1 x_1 + \beta_2 x_2 + \cdots + \beta_k x_k \quad (2.1)$$

where μ represents whether a pool tested positive ($\mu = 1$) or negative ($\mu = 0$), β_0 represents the intercept (or when $\ln(x) = 0$), $\beta_1 \dots \beta_k$ are the coefficients for each predictor variable, $x_1 \dots x_k$, where a unit increase in x results in the multiplicative effect on the probability of a positive mosquito pool (μ).

The final model was transformed into the probability of a mosquito pool testing positive for WNV by the binomial function:

$$p = \frac{\exp(\mu)}{1 + \exp(\mu)} \quad (2.2)$$

where μ represents the log-link function from equation 2.1.

2.4 Results

In 2012, 25,917 total individual female mosquitoes were organized into pools of ≤ 50 mosquitoes and tested for WNV. The majority of the mosquitoes (88%) that were pooled and tested were identified as *Cx. quinquefasciatus* (22,156 mosquitoes). A total of 2,642 pools were tested and 267 pools (10.1%) tested positive for WNV. 1,634 pools containing at least 1 *Cx. quinquefasciatus* mosquito were tested for WNV and 256 of these pools (15.7%) tested positive for WNV. *Cx. quinquefasciatus* pools accounted for the majority of the WNV-positive pools in

2012 (96%). During the 2012 season, 175 unique trap locations had at least one mosquito pool test positive (Figure 2).

Spatially, the highest infection rates occurred in north and central Dallas County (Figure 2). WNV activity was highest between May and September between weeks 20-37 (Figure 3). The county experienced the highest infection rate during the week of July 16-22 (week 29), with a peak of 47.66 for the infection rate. The first major aerial spraying across the county occurred during the week of August 16-20 (Week 33), which had an infection rate of 14.28 (Figure 3, solid arrow) (Ruktanonchai et al. 2014). The second major aerial spraying in Dallas County occurred during the week of August 22-23 (week 34), which had an infection rate of 9.36 (Figure 3, dashed arrow).

To represent the amount of development or urbanization, loadings from *PCI* were chosen as they explained 66% of the variance (Table 2). *PCI* consisted of positive loadings of land cover, impervious surfaces, and population density and a negative loading of NDVI.

Table 2 Principal Component Analysis (PCA) loadings for variables listed in the urbanization group (*PCI*). *PCI* explained the greatest amount of variance (66%), therefore, the loadings from *PCI* were chosen to represent the amount of urbanization around a trap location.

Variables	Loadings_{PCI}
Land cover	0.514
Impervious surfaces	0.573
NDVI	-0.465
Population density	0.438

The best-fit model included urbanization (*PCI* score), median age, and elevation (Table 3). Urbanization (Estimate: 0.436, 95% CI (0.232, 0.659)) and median age (Estimate: 0.272, 95%

CI (0.089, 0.508)) had positive associations with the probability of a pool being infected with WNV, while elevation (Estimate: -0.220, 95% CI (-0.438, -0.019)) had a negative association. The AIC of the best-fit model is fairly distinguishable in fit compared to the next-best models, where the AIC is ≥ 2.0 (Table 4).

Table 3 Best-fit model estimates with 95% confidence intervals (CI). Urbanization, median age, and elevation were significant predictors of the 2012 WNV epidemic in Dallas County, TX. Urbanization and median age have positive associations with the probability of finding a positive mosquito pool while elevation had a negative relationship with the outcome.

Variable	Estimate	95% CI
Urbanization	0.436	(0.232, 0.659)
Median age	0.272	(0.089, 0.508)
Elevation	-0.220	(-0.438, -0.019)
Intercept	-4.463	(-5.297, -3.713)

Table 4 Model comparisons between the best-fit model (bold) and similar models. The AIC of the best-fit model is fairly distinguishable in fit compared to the next-best model (≥ 2.0). A total of 64 models were compared in this study. This table does not display an exhaustive list of all the models that were compared.

Fixed Effects	AIC	Weight
Urbanization, median age, DEM	0.0	0.316
Urbanization, median age, median income, DEM	2.0	0.118
Urbanization, median age, distance to water, DEM	2.0	0.115
Urbanization, median age	2.7	0.083
Intercept	15.1	<0.001

The following log-link function based on equation 2.1 is derived from the logistic regression using the estimates from the best-fit model, corrected for the offsets, and scaled by the mean divided by the variates:

$$\ln(\mu) = -2.19 + 0.43 \times \frac{PC1}{1.62} + 0.30 \times \frac{Med\ Age - 36.88}{6.45} + (-0.20) \times \frac{DEM - 161.34}{21.58} + 0.94 \times \frac{15 - 15.48}{15.37} \quad (2.3)$$

where *PC1* represents the urbanization principal component group, *MedAge* represents the median age of the population, and *DEM* represents elevation. The final equation that was used to produce the map was transformed into the binomial function using equation 2.2.

The final risk map highlights areas that had greater probabilities of detecting a WNV-positive mosquito pool based on the significant factors identified from our models (Figure 4). The risk of WNV-positive *Cx. quinquefasciatus* ranged from 0.28% to 85.3%, with the greatest risk detected around the city of Carrollton in north Dallas County and within the city of Dallas. The southern region of Dallas County had lower probabilities compared to the northern counterpart. Based on these results, we can draw conclusions about the landscape and population demographics that were associated with WNV-infected mosquitoes during the 2012 epidemic.

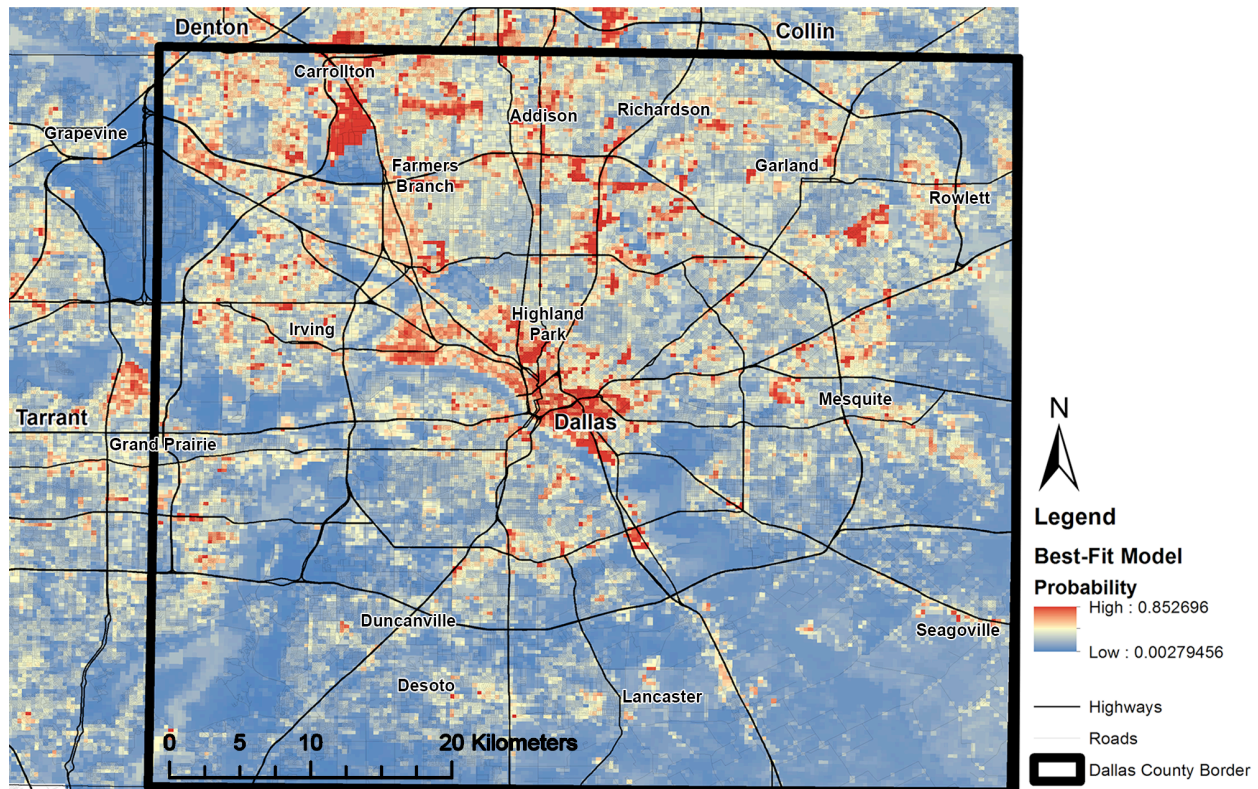


Figure 4 The probability of finding WNV-positive *Cx. quinquefasciatus* during the 2012 epidemic of WNV in Dallas, TX. The darker red colors represent areas of higher risk based on greater levels of urbanization, larger populations of elderly individuals, and lower elevations.

2.5 Discussion

This study investigated landscape and demographic factors related to the probability of finding a WNV-positive mosquito pool during the 2012 WNV epidemic in Dallas County, TX. Our study found an increased probability of WNV-positive mosquito pools in areas with higher urbanization, older populations, and lower elevations. Each of the factors from our model play an important role in the life cycle of the mosquito as well as the interactions between the vector and avian hosts that allow WNV to circulate in the enzootic cycle.

The urbanization index (*PCI*) had the largest effect on the probability of finding a positive mosquito pool, with the amount of urbanized land cover having the greatest influence within the principal component. The variables in *PCI* included positive loadings for land cover,

impervious surfaces, and population density and negative loadings for NDVI. NDVI measures the amount of vegetation present, therefore with increasing impervious surfaces and developed land and higher population density, we expected NDVI to decrease. These components directly affect the life cycle of *Cx. quinquefasciatus* because this species of mosquito prefers breeding habitats in urban areas particularly near human habitations, urban catch basins, and storm drains (Molaei et al. 2007, Andreadis 2012). Urban and peridomestic environments may form habitats containing water with high organic content, which are characteristic breeding grounds for *Cx. quinquefasciatus*. Furthermore, urban catch basins and storm drains provide shelter for *Cx. quinquefasciatus* for daytime resting and overwintering in quiescence during cold periods since this species does not enter diapause (Reisen et al. 1986, Su et al. 2003, Tesh et al. 2004a, Molaei et al. 2007). Other studies have yielded similar results, concluding that WNV and its vectors are found in urban environments (Hribar et al. 2001, Bolling et al. 2005, Cooke et al. 2006, Gibbs et al. 2006, Bowden et al. 2011). *Cx. pipiens*, the main WNV vector in the northeast U.S. and a close relative of *Cx. quinquefasciatus*, has also been positively linked to urbanization and the amount of development in Connecticut (Andreadis et al. 2004).

Avian reservoir populations are also affected by urbanization. Urbanization and development cause fragmentations in the landscape and create adjacent ecotones, which can also alter bird populations and thusly, the transmission dynamics and dispersal of WNV across the landscape (Wiens 1995, Coppedge et al. 2001, Marzluff 2001, Reisen 2010). As the landscape becomes more fragmented, adjacent bird populations may congregate in the same area because of increased habitat loss and overall reduce WNV transmission if these birds have low reservoir competence or alternatively increase WNV transmission if the bird populations are capable of producing high viremias of WNV (Allan et al. 2009). The overuse of certain avian hosts in urban

areas such as the American robin (*Turdus migratorius*), which is considered a superspreader of WNV, and concurrent underuse of other species with lower reservoir competence such as the great-tailed grackle (*Quiscalus mexicanus*) or European starling (*Sturnus vulgaris*) also alter the transmission dynamics of WNV (Hamer et al. 2009, Komar et al. 2018). The effects of urbanized cities on the avian community structure and resulting WNV transmission dynamics during the 2012 epidemic warrant further investigation.

Similarly, median age of the population had a positive correlation with the probability of finding a WNV-positive pool. Overall, the relationship between the median age of the population and *Cx. quinquefasciatus* populations has not been studied thoroughly in Texas. Certain behaviors associated with age can mediate *Cx. quinquefasciatus* habitats, the resulting abundances, and their interactions with avian hosts. These interactions will lead to greater amplification of WNV in the enzootic cycle and eventual spillover to human populations. Another study examining the 2002 outbreak of WNV in Chicago, IL found that older ages were associated within clusters of WNV incidence in humans and dead birds (Ruiz et al. 2004). Behaviors and activities of the older population may put them at higher risk for WNV infection if they contribute to certain aspects of WNV ecology, such as the interface at which mosquitoes and birds come into contact for enzootic transmission of the virus. Outdoor activities common among older populations, such as yard work or development of yard structures that collect water or food, can contribute to mosquito abundance and bring competent hosts together (Ruiz et al. 2004, Blaine et al. 2010). A nationwide study investigating various landscape, demographic, and climatic associations with high/low WNV ratings during the 2012 epidemic did not find a significant relationship between median age and the WNV ratings in the southeast region, which included Dallas County, Texas (Degroote et al. 2014). The southeast region included counties

that had low WNV ratings, which may have diluted the relationships between some variables and WNV activity present in this region (Degroote et al. 2014).

The best-fit model suggested a negative association between elevation and *Culex spp.* WNV infection rate, which is consistent with observations in prior studies (Mongoh et al. 2007, Ruiz et al. 2010, Bisanzio et al. 2011, Chuang et al. 2012). In combination with other physiographic elements, elevation may limit WNV transmission in Georgia where higher elevations have lower temperatures, subsequent smaller mosquito abundances, and diversity in avian species composition (Gibbs et al. 2006). Similarly, Ruiz et al. (2010) found that elevation also negatively affected minimum infection rates of WNV in *Cx. pipiens* in Chicago, IL and suggested that elevation could mediate the effect of precipitation by collecting water at lower elevations to enrich with organic materials needed for mosquito population survival. Further west, two more studies arrived at the same conclusions regarding elevation. A study in North Dakota found an inverse relationship between the incidence of WNV in horses and elevation (Mongoh et al. 2007). In South Dakota, elevation had a negative relationship with human WNV risk, citing the main vector's (*Cx. tarsalis*) inability to survive at colder temperatures found at higher elevations as the reasoning for the decreased odds of human WNV disease (Chuang et al. 2012). Elevation by itself may not play a significant role in Dallas and surrounding areas since there is a ≤ 100 m difference between the highest and lowest points. Instead, lower elevation and increased urbanization are likely mediating the effects of precipitation, similar to what was found in Chicago, IL.

Aerial spraying occurred late in the season starting at week 33, which corresponds to the time period when the infection rate was already decreasing (Figure 3). The peak in infection rate occurred in week 29 (July 16-July 22). The first aerial spraying occurred during the decline in

the infection rate at week 33 (August 16-20) and covered northern and central Dallas County (Ruktanonchai et al. 2014). A second spraying occurred the following week (week 34, August 22-23) for eastern and southern Dallas County (Ruktanonchai et al. 2014). Each of the spraying events cost about \$1 million, and the total costs for both spraying events in Dallas County likely exceeded \$1.6 million (Roehr 2012, Chung et al. 2013). The WNV season in mosquitoes had already dramatically decreased by the time spraying events occurred, making it difficult to discern if the aerial spraying was truly effective in reducing the infection rate of WNV or if the infection rate decreased due to seasonality of *Cx. quinquefasciatus*. However, spraying events later in the season highlight the significance of this unique dataset in that the results recorded during this period of limited vector control reduces the risk of introducing artifacts into the analysis due to control activities (Figure 3).

Studies on the effects of aerial spraying on *Cx. quinquefasciatus* abundance is limited for this event, but one study analyzed the effects of aerial adulticide application on human WNV incidence (Ruktanonchai et al. 2014). Here, aerial spraying in the county was associated with an overall reduction in WNND in the county, but disease incidence decreased in both treated and untreated areas with a greater difference seen in aerial-sprayed areas (Ruktanonchai et al. 2014). The number of cases during this time was expected to decrease given how late the aerial application of adulticides was conducted (Ruktanonchai et al. 2014).

Without an available vaccine for humans, mosquito control and public awareness campaigns are currently the most effective way to prevent and/or minimize WNV human cases. Understanding the ecology of WNV dynamics with the mosquito, environment, and social demographics plays a key role in effective intervention campaigns. Furthermore, predicting when and where WNV in mosquitoes occurs may provide an early warning system giving an

opportunity to control mosquitoes before bridge transmission to humans and to alert the public with the appropriate messages to reduce the risk of exposure. This study provides one of the first efforts to model *Culex* infection during the 2012 WNV epidemic for Dallas County, TX. Future efforts should concentrate on establishing long-term mosquito surveillance databases, modeling WNV for early detection in other major metropolitan areas of Texas, and utilizing quantitative measurements to identify a threshold for early control protocols. While this study only focused on one year, these findings provide the first steps to understanding the ecology of WNV in *Cx. quinquefasciatus* mosquitoes during the 2012 epidemic in the hopes of preventing another devastating event in a major city such as Dallas, TX.

CHAPTER III

THE EFFECTS OF LANDSCAPE AND DEMOGRAPHIC FACTORS ON WEST NILE
VIRUS INFECTION IN *CULEX QUINQUEFASCIATUS* SAY (DIPTERA: CULICIDAE) IN
HARRIS COUNTY, TEXAS

3.1 Synopsis

West Nile virus (WNV) was first introduced in the U.S. in 1999 and is now considered endemic with outbreaks reported in every state in the contiguous U.S. Previous work has revealed that environmental and landscape determinants of WNV transmission are heterogeneous across the U.S. Texas has experienced several WNV epidemics, and now that circulation has occurred for nearly two decades, we aim to exploit the natural spatio-temporal variation in WNV to identify factors driving these patterns. We analyzed Harris County's surveillance data from 2005-2015, focusing on the primary mosquito vector, *Culex quinquefasciatus* Say, to identify landscape and demographic predictors related to a high WNV vector index (VI), which estimates the average number of WNV-infected female *Culex* mosquitoes collected per trap night. During this period, 4,326,487 *Cx. quinquefasciatus* mosquitoes were identified, pooled, and tested for WNV. Out of 135,692 pools of mosquitoes, 4,717 pools tested positive for WNV (3.5%). Using linear mixed models, the best-fit model that explained higher VI's included higher elevation, more impervious surfaces, higher median income, and a predominantly Hispanic population. The resulting model was used to create a predictive map that highlights high-risk areas of Harris County for WNV-infected *Culex* mosquitoes, with the highest predicted VI's located in central and north Harris County. Our study provides one of the first modeling attempts to delineate the

relationship between WNV and the landscape in Texas to build a more efficient early warning system for WNV.

3.2 Introduction

West Nile virus (WNV) was first introduced to the U.S. in 1999 and has since spread throughout the continental U.S. Since its introduction, 46,086 cases of WNV have been reported, and 2,017 deaths have been recorded as of 2016 (CDC 2016b). In 2012, the U.S. witnessed the largest outbreak of WNV with the majority of the cases and deaths occurring in Texas (Murray et al. 2013). Texas alone reported a total of 1,868 cases of WNV, with 844 cases of West Nile virus neuroinvasive disease (WNND) and 89 deaths throughout the state from the 2012 mosquito season, with the majority of the incidents in Dallas, TX or the surrounding area (Murray et al. 2013).

WNV is a zoonotic disease with an enzootic cycle involving avian amplification hosts and mosquito vectors (Weaver and Reisen 2010). Once the avian host amplifies the virus and is fed on by mosquitoes, bridge transmission (e.g. spillover) occurs to other “dead-end hosts,” such as horses and humans. *Culex* mosquitoes are the primary enzootic vectors (Turell et al. 2005, Weaver and Reisen 2010) and serve as bridge vectors to humans (Kilpatrick et al. 2005a, Hamer et al. 2008b). In central and southeast Texas, *Culex quinquefasciatus* is the most relevant mosquito species involved in the transmission cycle (Strickman and Lang 1986, Lillibridge et al. 2004, Molaei et al. 2007).

Differences in breeding habitats and host-seeking behaviors of various mosquito species have been linked to the geographic distribution of land cover types. *Cx. quinquefasciatus* abundance has been linked to characteristics associated with urban landscapes, such as combined

sewage overflows, water storm drains, and water sources rich in organic matter (Beehler et al. 1993, Bolling et al. 2005, Calhoun et al. 2007, Chaves et al. 2009, Gleiser and Zalazar 2010). Furthermore, *Cx. quinquefasciatus* takes advantage of manmade water sources for breeding sites including roadside ditches, sewage treatment sites, household containers, and low places with poor drainage (Hribar et al. 2001, Ruiz et al. 2004, Reisen et al. 2008c). Anthropogenic modification of habitats can cause increased transmission of WNV through the creation of adequate habitats for vectors, which may increase avian-vector contact (Hamer et al. 2008b).

Environmental and demographic features of the landscape are known to have significant associations with WNV transmission (Epstein and Defilippo 2001, Kuhn et al. 2005, DeGroote and Sugumaran 2012, Degroote et al. 2014). The features of the environment capable of influencing mosquito and bird communities include elevation, water sources, and impervious surfaces. Social demographic predictors of human infection include median age of the population, race, median income, and age and status of the neighborhood (Brownstein et al. 2002, Ruiz et al. 2004, Diuk-Wasser et al. 2006, Reisen et al. 2008a, Bisanzio et al. 2011).

Given that a component of arbovirus management aims to conduct mosquito abatement to minimize vector populations and dampen virus transmission in the enzootic cycle, it is critical to have an in-depth understanding of how biotic and abiotic factors relate to the enzootic cycle of WNV, in which the vector index is one such metric. In addition, virus activity in mosquitoes and birds precede human exposure (Hamer et al. 2008b), thereby offering an early warning system to allow intervention strategies to protect public health (Andreadis et al. 2004). Therefore, mosquito infection data is a valuable parameter in determining the risk of WNV infection to humans (Brownstein et al. 2002, Eisen and Eisen 2008, Ruiz et al. 2010).

Currently, WNV has one of the largest geographic distributions of any zoonotic mosquito-borne virus due to its global expansion to all continents except for Antarctica (Kramer et al. 2008). Accordingly, WNV has adapted to many landscapes, mosquito vectors, and avian amplification hosts. This plasticity has resulted in many discrepancies in how the landscape and demographic features relate to WNV transmission intensity (Ruiz et al. 2004, Ruiz et al. 2007, DeGroote et al. 2008, Degroote et al. 2014) and emphasizes how results from one geographic region are unlikely to inform a different geographic region. This study focuses on Harris County, TX and the extensive mosquito surveillance program focused on *Culex spp.* mosquitoes that began due to St. Louis Encephalitis (SLE) in 1965 (Lillibridge et al. 2004). We evaluate the spatial variation of WNV infection in *Cx. quinquefasciatus* in Harris County and identify landscape (elevation, impervious surfaces, and distances to water sources) and demographic (median age of the population, race/ethnicity, gender, median income, and population density) determinants of the variation. Furthermore, we aim to develop a predictive map that may be used to predict locations of *Cx. quinquefasciatus* infection with WNV based on environmental and socioeconomic differences in Harris County, TX.

3.3 Methods

3.3.1 Mosquito Data

Annually, Harris County Public Health Mosquito and Vector Control (HCPH) places 489 mosquito traps throughout 268 operational areas (Figure 5) of the county (Nava and Debboun 2016, Randle et al. 2016). HCPH conducted mosquito surveillance on a weekly basis between 2002-2015. For this study, we used data starting in 2005, which was when consistent trapping throughout the county was established. During this 11-year period, HCPH conducted 99,066 trap

surveys/visits. Trap locations were visited at least once a year with an average of 29 weekly visits per trap per year.

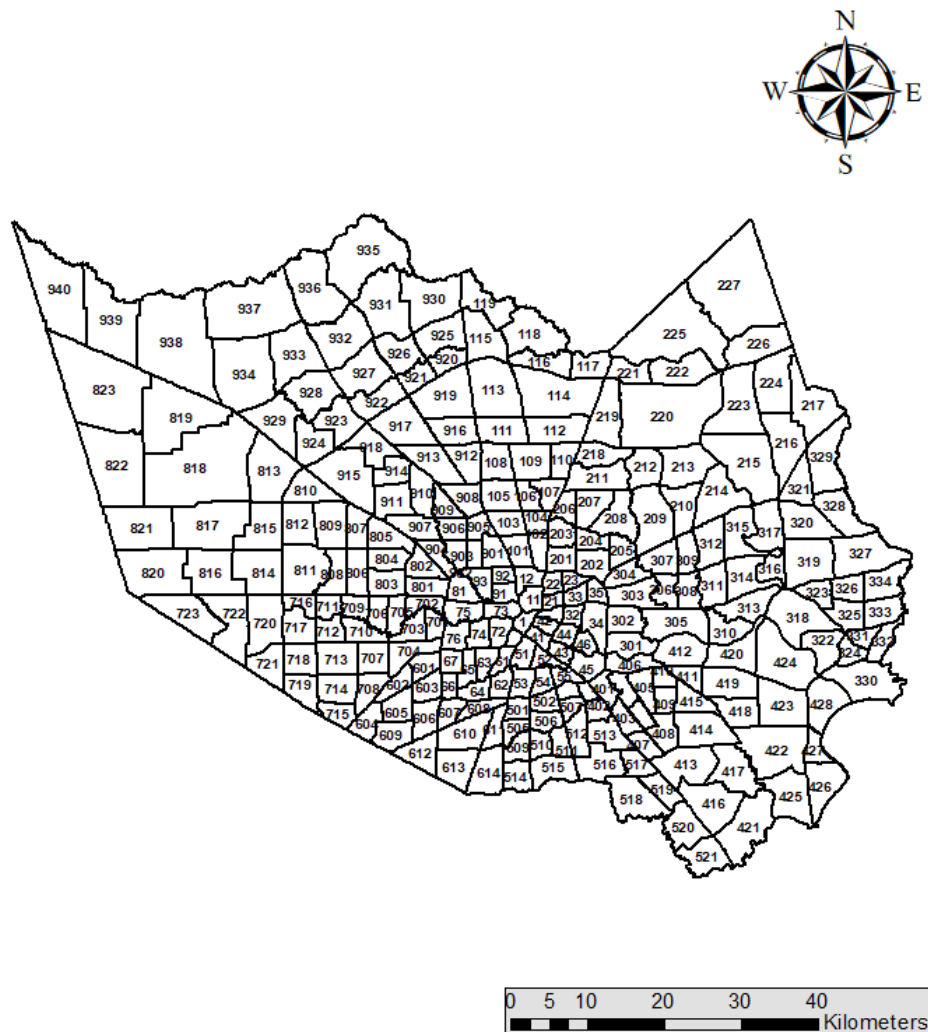


Figure 5 Operational areas in Harris County, TX. Harris County divides the county into “operational areas” for surveillance, inspection, surveying, and control purposes.

HCPH used a combination of storm sewer (SS), gravid (GV), under house, and BG traps, but only SS and GV traps will be considered in this study because of their ubiquitous usage

throughout the county during the entire study period (Figure 6). SS traps are modified CDC Light Traps that are baited with dry ice and attached to man hole covers underground. GV traps are baited with hay infusion water and placed in residential yards, usually under vegetation. The hay infusion is composed of mixing 1.3 kg of Coastal bermudagrass, *Cynodon dactylon* (L.), with 42 gallons of water and then aged for 10-14 days (Dennett et al. 2007a). SS traps capture females underground in storm sewers, while GV traps are used to trap gravid females above ground (Dennett et al. 2007a, Curtis et al. 2014).

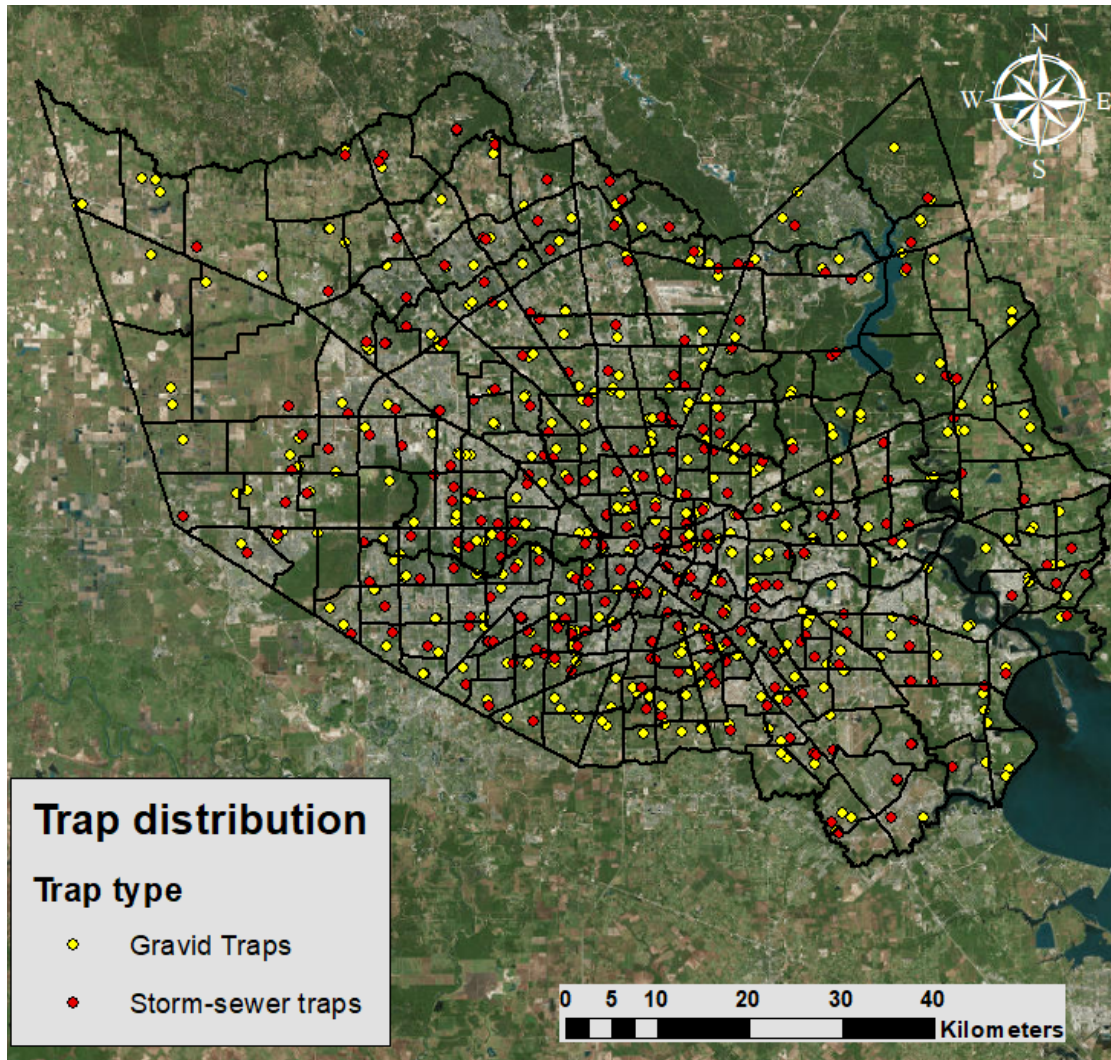


Figure 6 Gravid (GV) and storm-sewer (SS) trap locations in operational areas throughout Harris County between 2005-2015. GV traps are placed in residential yards with permission from homeowners. They are baited hay-infused water to attract female mosquitoes. SS traps are attached to man-hole covers and suspended into storm sewer systems. These traps are baited with carbon dioxide to attract female mosquitoes.

The mosquito collection protocol from Harris County has been previously described (Molaei et al. 2007, Curtis et al. 2014). In summary, traps are placed in the afternoon between 1:30 PM-5:00 PM and then collected the following morning between 7:30 AM-10:30 AM. Traps are placed into “operational areas,” lines that divide the county for surveillance, inspection, surveying, and control purposes (Figure 5) (Hunt and Hacker 1984). The 268 operational areas

are based on municipal, district, and zip code lines. After the traps were visited, mosquitoes were identified by species and sex on a chill table to preserve the presence of the virus and then sorted into pools of ≤ 50 mosquitoes, with a maximum of 3 pools per trap. Harris County tested for WNV antigen in mosquito pools using an enzyme-linked immunosorbent assay (ELISA) and positive results were confirmed with a Rapid Analyte Measurement Platform (RAMP) test (Lillibridge et al. 2004, Randle et al. 2016). To be considered a positive pool, the mosquito pool must test positive on both the ELISA and RAMP test. A positive pool is a mosquito pool that contains at least one *Cx. quinquefasciatus* mosquito positive for WNV.

3.3.2 Landscape and Demographic Variables

We used a variety of landscape and demographic variables for this study (Table 5). To calculate the shortest distance of traps from water bodies, we gathered water body data from the National Hydrography Dataset for the state of Texas, which is part of the U.S. Geological Survey (USGS) National Map Program (United States Geological Survey 2018b). This dataset includes information about rivers and streams, ponds and lakes, springs, canals, wetlands, and shorelines. In ArcGIS 10.4 (Redlands, CA), we calculated the shortest distance (in meters, m) between the center of a trap and the nearest water body.

Table 5 Predictor variables used in the model creation steps. We used variables in our model that describe the landscape and demographic of the population in Harris County. The data for each variable were collected from their respective sources listed on the right.

Variables	Sources
Distance to water bodies	U.S. Geological Survey
Impervious surface	National Land Cover Database

Table 5 (continued)

Variables	Sources
Elevation	National Elevation Database
Demographic (census tracts)	U.S. Census Bureau (2000 and 2010 Census)
Median income	
Median age	
Percentage of males	
Percentage of Hispanics	
Percentage of African-Americans	
Percentage of Caucasians	
Population density	

Demographic variables measured at the census tract level were obtained from the U.S. Census Bureau (United States Census Bureau 2018) for years 2005-2015. The demographic variables were represented as estimates of median income, median age, percentage of males, percentage of black residents, percentage of white residents, and percentage of Hispanic residents per census tract. We used both the official 2000 and 2010 decennial censuses to capture changes in the population during our study period. The 2000 Census was used to represent demographic data from 2005-2009 and the 2010 Census was used to represent demographic data for 2010-2015. We used the official census datasets because Congress officially recognizes these datasets and they have previously been used in other WNV studies (Andreadis et al. 2004, Ruiz et al. 2004, Allan et al. 2009). In addition, we calculated population density by dividing the population size by the area (m^2) of the census tract. The census tract level was used because it was the highest resolution that included all of the variables of interest.

We used the percentage of impervious surface coverage to represent the amount of manmade structures and development. Impervious surface data were obtained from the Multi-

resolution Land Characteristics Consortium (Multi-Resolution Land Characteristics Consortium 2018) of the National Land Cover Database. Finally, elevation data were extracted from the USGS National Elevation Dataset (United States Geological Survey 2018a). Elevation data were then converted to slope and aspect data in ArcGIS.

To extract demographic, impervious surface, elevation, slope, and aspect data, we placed a 750-m buffer around each trap and used ArcGIS to calculate the average or percentage estimate of each variable. This buffer represents the estimated dispersal distance of *Cx. quinquefasciatus* (Medeiros et al. 2017). For alignment purposes, the map layers in ArcGIS used the GCS North American 1983 coordinate system and the Lambert Conformal Conic projection.

3.3.3 Statistical Model

For modeling purposes, we used data collected as mosquito pools of ≤ 50 mosquitoes per pool; such that for observations at operational area i during visit j , if a mosquito pool is WNV positive, it receives a value of 1; and when a mosquito pool does not test positive, it receives a value of 0. The infection rate (IR) was calculated using the Biggerstaff Pooled Infection Rate Excel Add-in, which was developed by the Center for Disease Control and Prevention (Biggerstaff 2009). Mosquito population density was calculated as a relative abundance of mosquito species detected in terms of trapping effort by operational area and by year (Bolling et al. 2009a). We combined the abundance of the mosquito population with the IR to create the vector index (VI) to adequately express arbovirus transmission risk posed by a vector population (Gujral et al. 2007). The VI incorporates the vector species abundance and the vector species infection rate to quantify the risk into a single value to estimate the number of infected

mosquitoes collected per trap night for a given species (Gujral et al. 2007). The VI is expressed by:

$$VI = \sum_i N_i P_i \quad (3.1)$$

where N is the average abundance of mosquito species i (number of female mosquitoes per trap night) and P represents the estimated IR for mosquito species i . The resulting VI was then used as our final response variable in our models. To capture the peak season of WNV in Harris County, we focused primarily on weeks 20-40 for our calculations and analyses.

We selected 12 predictors that were expected to affect aspects of the WNV cycle and therefore, the WNV VI in mosquitoes in Harris County. Our covariates for the model include the proximity to water, elevation, slope, aspect, percentage of impervious surfaces, median age, percentage of males, median income, and percentage of white, black, and Hispanic populations as covariates in our model because these variables have been previously associated with WNV spatial patterns (Deichmeister and Telang 2010, DeGroot and Sugumaran 2012, Degroot et al. 2014). We selected 2005-2015 as our period of interest because the program did not consistently start trapping in all operational areas of the county until 2005.

All covariates were normalized to improve predictive accuracy and to prevent covariates with large numeric value ranges from impacting the prediction process. To evaluate the predictors on the WNV vector index, we used a linear mixed effect model, with a restricted maximum likelihood (REML) method, a variant of the maximum likelihood method (Bates et al. 2015). We used the R statistical software and the *lme4* package to fit the model with the function *lmer* (Bates et al. 2015). The model was fit in the following parameterization:

$$VI_{ij} = \alpha_0 + \alpha_1 x_j + \beta_{0i} + \beta_{1i} x_j + \dots + \epsilon_{ij} \quad (3.2)$$

where VI_{ij} is the WNV vector index in i th operational areas at x_j year, the α_0 and α_1 are the fixed intercept and slope respectively, β_{0i} and β_{1i} are the random intercept and slope, respectively, and ϵ_{ij} is the residual. The assumptions for the random effects β_{0i} , β_{1i} and residuals are:

$$(\beta_{0i}, \beta_{1i})^T \sim N(0,0)^T, G) \text{ and } \epsilon_{ij} \sim \text{Normal}(0, \sigma^2) \quad (3.3)$$

We assessed convergence of the model parameters using the REML criterion. We used a backward elimination process and Akaike's Information Criterion (AIC) to choose the best model, with the smallest AIC values representing the model of best-fit. The AIC measures the relative quality of models for a given number of parameters and models goodness of fit, aiming to minimize the number of parameters (Bozdogan 1987, Hlavac 2015).

To create a predictive map, the best-fit model outputs were mathematically combined using ArcGIS (Rochlin et al., 2011). The variables and their estimates of the best-fit model were used to model the predictive map. To validate our model we calculated the VI for each of the GV and SS traps in each operational area used in 2016-2017 ("observed"), which resulted in 403 and 370 records collected in 2016 and 2017, respectively. These records were imported into ArcGIS, where we extracted the "expected" data from the predictive map. Then, we compared the observed and expected values from 2016-2017 using an R^2 calculation.

Data collection protocols in 2016-2017 were similar to the protocols that were set in 2002, however, collection and testing evolved to include more *Aedes spp.*-specific surveillance

with the possibility of Zika virus arriving in Texas. This included more BG traps, testing more *Aedes spp.* for various arboviruses, and testing fewer *Culex spp.* pools for WNV and SLE.

3.4 Results

We analyzed 11 years of mosquito surveillance data in Harris County (2005-2015). Of the 489 mosquito traps, 268 were GV and 221 were SS mosquito traps (Figure 6). Each operational area has between 1 and 4 trap locations; the average number of traps per operational area is 1.6 (± 0.79 S.D.) GV traps and 1.1 (± 0.37 S.D.) SS traps. During this period, 4,236,487 female *Cx. quinquefasciatus* mosquitoes were identified, pooled, and tested for WNV. A total of 135,692 mosquito pools containing ≤ 50 individual mosquitoes were tested during the 11 years. Of the total number of mosquito pools, 4,717 pools tested positive for WNV, which represent 3.5% of the mosquito pools tested.

The highest WNV VI in Harris County (average between GV and SS traps) occurred in 2014 (554.87 ± 78.27), 2011 (232.49 ± 29.63), and 2012 (176.63 ± 24.78) (Figure 7). The years with the lowest WNV VI's in mosquitoes were in 2008 (4.96 ± 1.92), 2007 (13.21 ± 3.12), and 2013 (48.35 ± 9.64) (Figure 7). The highest mean VI's calculated from GV traps were found in operational areas in north and central Harris County (Figure 8). The highest mean VI's calculated from SS traps were found in areas in central Harris County with a few areas in northwest Harris County (Figure 9).

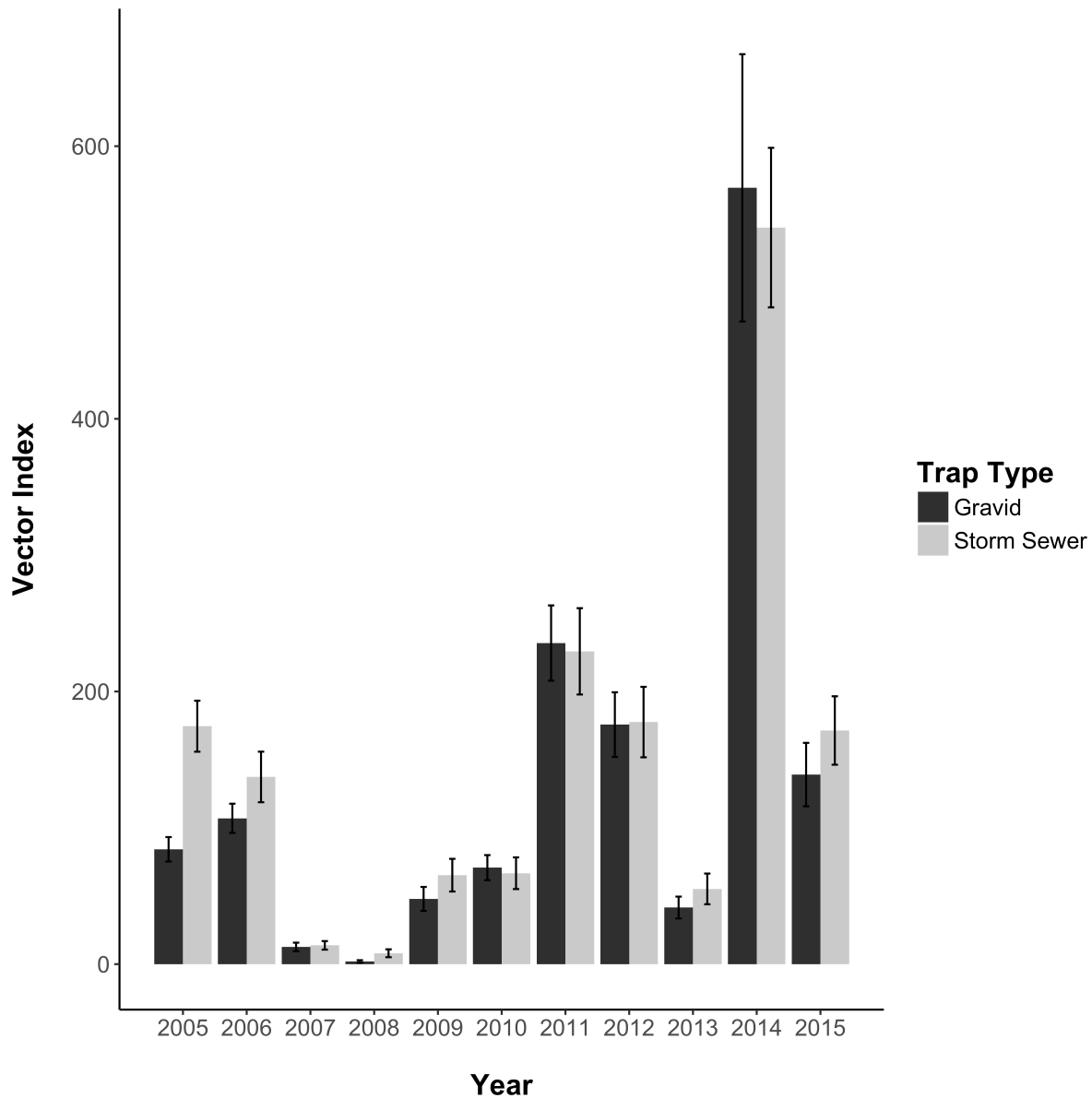


Figure 7 Comparison of WNV vector index (VI) by year and trap type. The vector index is the product of the abundance of female *Cx. quinquefasciatus* mosquitoes and the infection rate. The VI varies per year, with 2014 having the greatest VI and 2008 having the lowest VI. While the VI varies per year, the amount of variability of VI's measured per trap type was minimal.

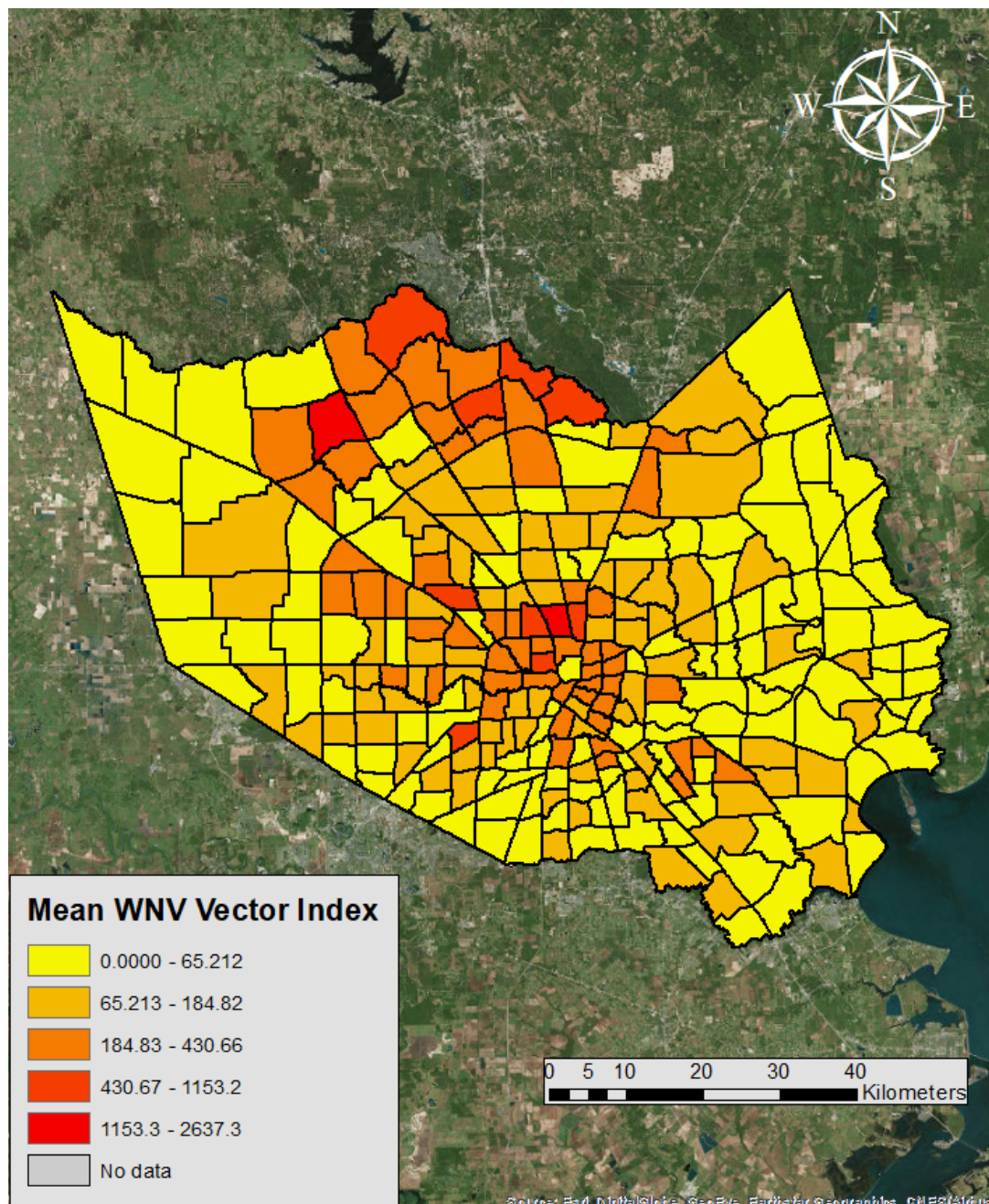


Figure 8 Mean WNV VI for GV traps by operational area in Harris County for years 2005-2015. The VI was calculated based on weeks 20-40, which represents the peak WNV season. The highest VI occurs in central and northern Harris County.

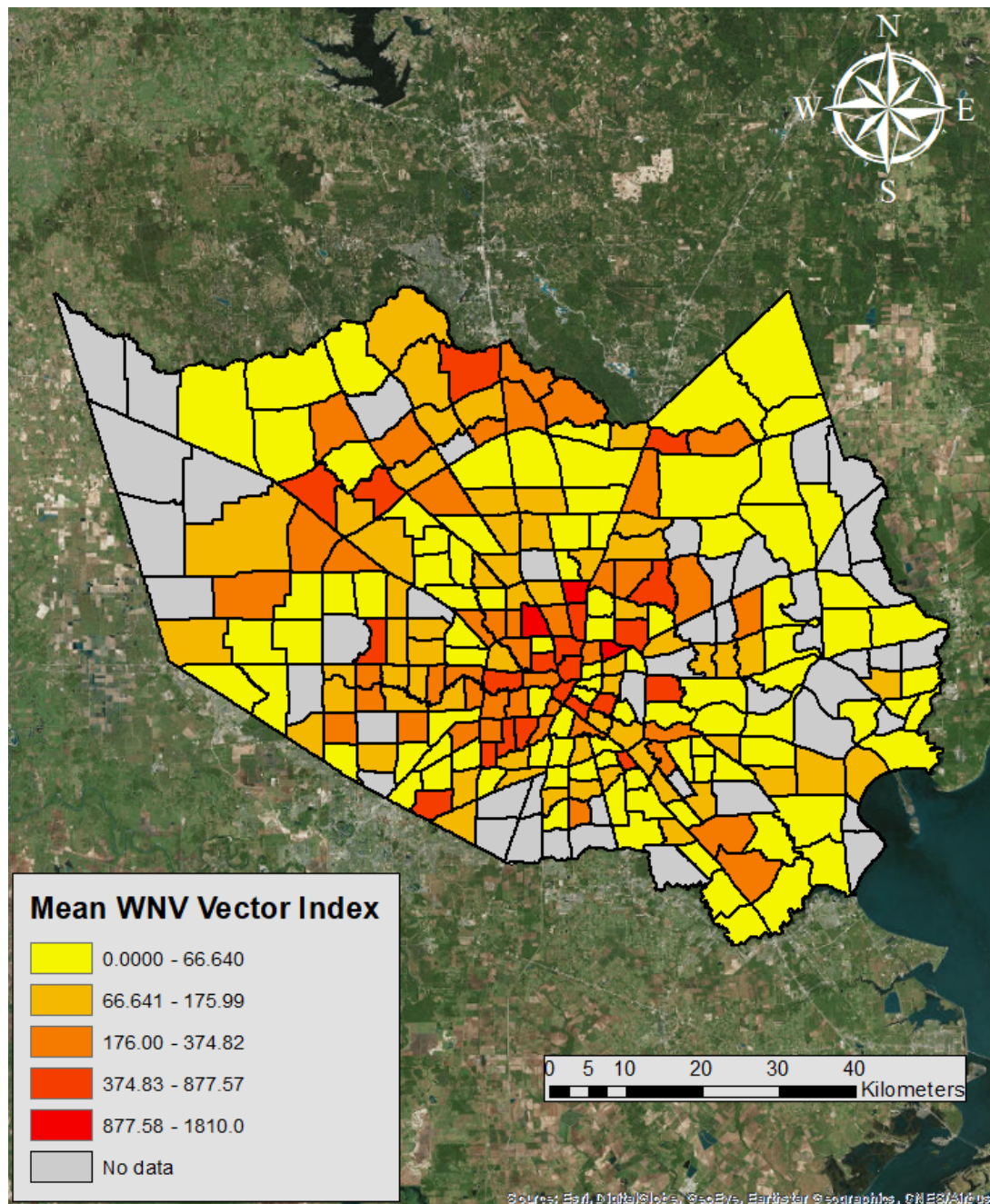


Figure 9 Mean WNV VI for SS traps by operational area in Harris County for years 2005-2015. The VI was calculated based on weeks 20-40, which represents the peak WNV season. Gray represents operational areas that did not have a VI, due to missing calculations for the abundance or infection rate.

The best-fit model (equation 3.4) that explained the WNV VI in Harris County included the following predictors: elevation with an estimate of 44.82 (95% CI: 21.08, 68.56), impervious surfaces with an estimate of 31.37 (95 % CI: 7.76, 54.98), median income with an estimate of 37.83 (95 % CI: 5.75, 69.92), and percent of Hispanic populations with an estimate of 43.38 (95 % CI: 7.31, 79.45) (Table 6). All predictors showed positive associations with the VI. Our best-fit model included operational area, frequency of trap visits/surveys, sampling year, population density, and trap type nested in the operational area as random factors in our model because their variation in the study area was due to stochasticity (Table 6) (Bates et al. 2015).

$$y = \text{meanVI} + (\text{DEM} * 44.82) + (\text{impervious surface} * 31.37) + (\text{median income} * 37.83) + (\text{percent Hispanic} * 43.38) \quad (3.4)$$

When we extrapolated the best-fit model as our predictive map (Figure 10), we found high probabilities of high vector indexes mainly in central and north Harris County with elevated VI's in west Harris County as well. In central Harris County, cities and regions such as Bellaire, Houston, Hunters Creek Village, Jersey Village, and Spring Valley contained operational areas with the highest predicted VI's. In the north, Cypress, Spring, and Tomball also predicted high VI's. Pasadena, which is located in southeast Harris County, was also predicted to have high VI's. The map also highlighted areas with low VI's throughout most of east Harris County.

Our model validation showed that our best-fit model could predict VI's with 38% accuracy and 26% accuracy when using 2016-2017 GV or SS trap data, respectively (Figure 11).

Table 6 Model Comparisons. Comparisons of models using a backwards elimination process. Covariate estimates are given with 95% confidence intervals in (). Random effects and their standard deviations for each model are listed below. The best-fit model is model (6), which is characterized with the lowest AIC and is **bolded**.

Covariates	Models							
	(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)
proximity water	1.23 (-17.24, 19.70)	2.76 (-17.14, 22.66)						
median age	6.00 (-15.67, 27.68)	5.04 (-17.54, 27.62)	5.33 (-17.14, 27.79)					
percentage males	2.49 (-14.17, 19.15)	0.63 (-16.64, 17.89)	0.49 (-16.72, 17.69)	0.73 (-16.44, 17.90)				
population density	7.11 (-9.18, 23.40)	7.11 (-9.54, 23.76)	7.18 (-9.45, 23.82)	6.57 (-9.86, 23.00)	6.55 (-9.87, 22.98)			
slope %	-10.33 (-26.19, 5.54)	-10.62 (-27.88, 6.64)	-10.87 (-28.01, 6.27)	-10.94 (-28.06, 6.19)	-10.97 (-28.08, 6.13)	-13.01 (-32.87, 6.84)		
aspect	4.27 (-10.92, 19.46)	1.80 (-14.73, 18.33)	1.83 (-14.69, 18.34)	1.82 (-14.68, 18.33)	1.76 (-14.67, 18.20)	-0.08 (-18.89, 18.73)	-0.70 (-19.52, 18.12)	
median income	30.32* (4.64, 56.00)	30.86* (4.14, 57.59)	30.89* (4.19, 57.59)	31.89* (5.53, 58.24)	31.84* (5.52, 58.17)	37.83* (5.75, 69.92)	38.45* (6.33, 70.57)	38.45* (6.35, 70.55)
elevation	33.95*** (15.80, 52.11)	36.11*** (16.61, 55.60)	35.91*** (16.49, 55.34)	34.90*** (15.99, 53.81)	34.89*** (16.00, 53.78)	44.82*** (21.08, 68.56)	43.93*** (20.15, 67.71)	43.91*** (20.16, 67.66)
impervious surface	31.97** (12.29, 51.65)	32.10** (10.91, 53.28)	33.22*** (13.64, 52.80)	33.83*** (14.42, 53.24)	33.90*** (14.57, 53.24)	31.37** (7.76, 54.98)	30.78* (7.12, 54.44)	30.77* (7.14, 54.40)
percent white	28.94 (-16.52, 74.39)	29.42 (-17.46, 76.31)	30.24 (-16.29, 76.76)	30.68 (-15.79, 77.15)	30.87 (-15.43, 77.17)	31.09 (-24.33, 86.50)	36.17 (-18.74, 91.09)	36.18 (-18.70, 91.06)
percent black	43.71 (-5.95, 93.37)	43.79 (-7.79, 95.37)	44.95 (-5.96, 95.86)	44.69 (-6.16, 95.54)	44.72 (-6.11, 95.55)	50.28 (-10.51, 111.07)	55.73 (-4.55, 116.02)	55.74 (-4.51, 115.98)
percent Hispanic	41.62** (9.99, 73.25)	41.40* (8.24, 74.57)	41.81* (8.80, 74.82)	38.69* (8.49, 68.90)	38.86* (8.91, 68.81)	43.38* (7.31, 79.45)	46.50* (10.67, 82.32)	46.52* (10.72, 82.32)
Constant	145.77** (53.11, 238.43)	148.19** (55.25, 241.13)	148.31** (55.37, 241.25)	148.42** (55.34, 241.49)	148.37** (55.33, 241.40)	140.79** (49.74, 231.84)	140.60** (49.73, 231.48)	140.61** (49.74, 231.47)

Table 6 Continued

	Models							
	(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)
Observations	3838	3838	3838	3838	3838	3838	3838	3838
Log Likelihood	-28765.48	-28758.19	-28761.46	-28764.92	-28768.02	-28704.84	-28708.89	-28712.08
Akaike Inf. Crit.	57566.97	57552.37	57556.92	57561.85	57566.03	57439.67	57443.79	57450.13
Bayesian Inf. Crit.	57679.52	57664.92	57663.22	57661.89	57659.82	57533.46	57533.31	57531.42
Note:	*p<0.05; **p<0.01; ***p<0.001							

Standard Deviations of Random Effects

	Models							
Random Effects	(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)
area	78.64	54.68	54.69	54.16	53.95	17.15	20.87	20.99
trap visits	27.20	28.91	29.29	29.63	29.53	5.73	9.65	9.87
year	151.43	151.38	151.32	151.51	151.47	149.56	149.32	149.32
trap type	0.00							
area: trap type		97.52	97.07	97.06	96.99	148.99	149.30	148.91
Pop Den						319.97	319.76	319.73
Residual	432.28	425.96	425.96	425.95	425.92	303.23	303.20	303.2

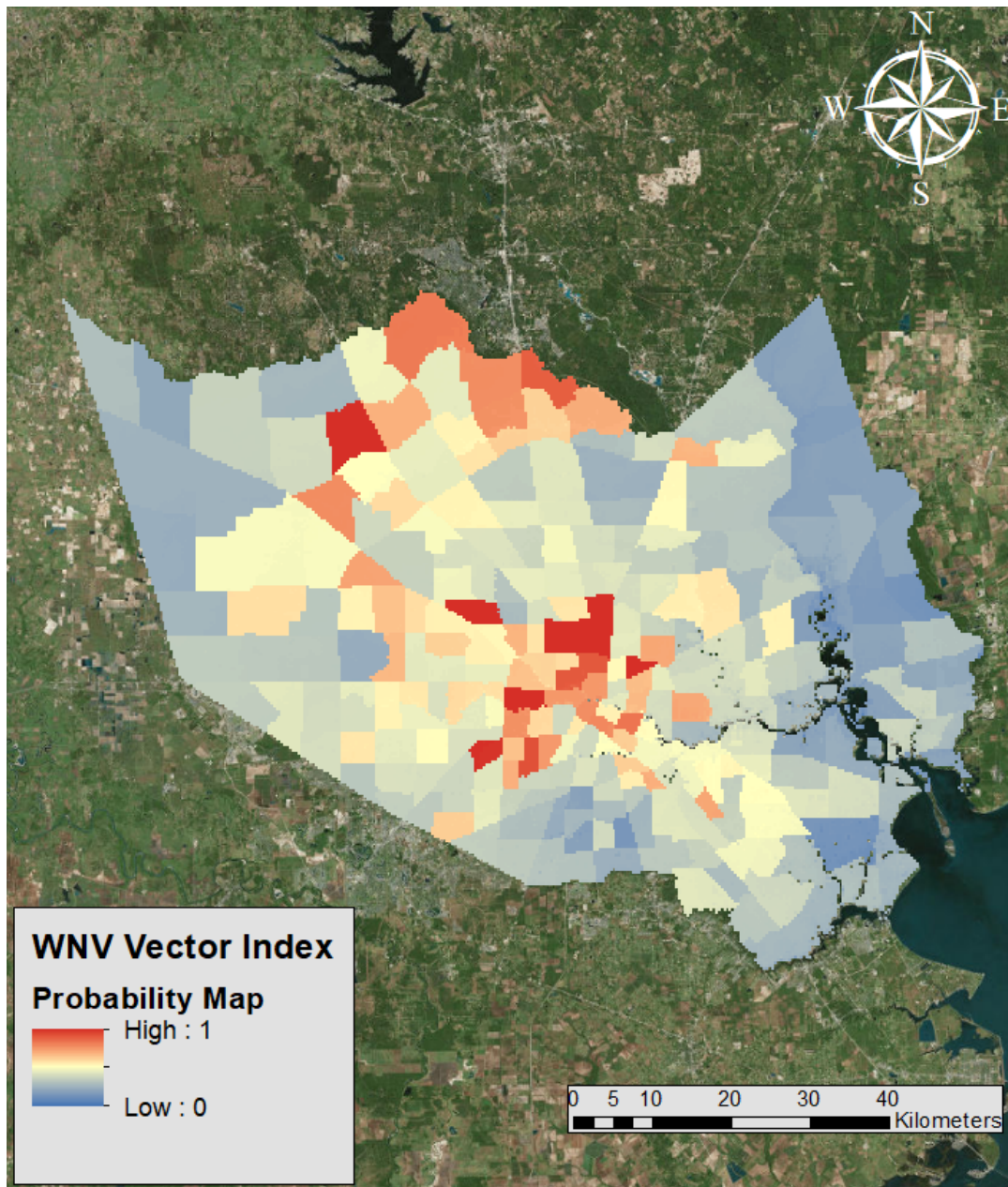


Figure 10 Predictive map of the best-fit model. The predictive map was generated based on equation 3.4 in ArcGIS. The model measured the probability of detecting a high WNV VI throughout Harris County, with darker red regions representing operational areas that have greater risk of high VI's based on higher median income, greater elevation, more impervious surfaces, and a predominantly Hispanic population.

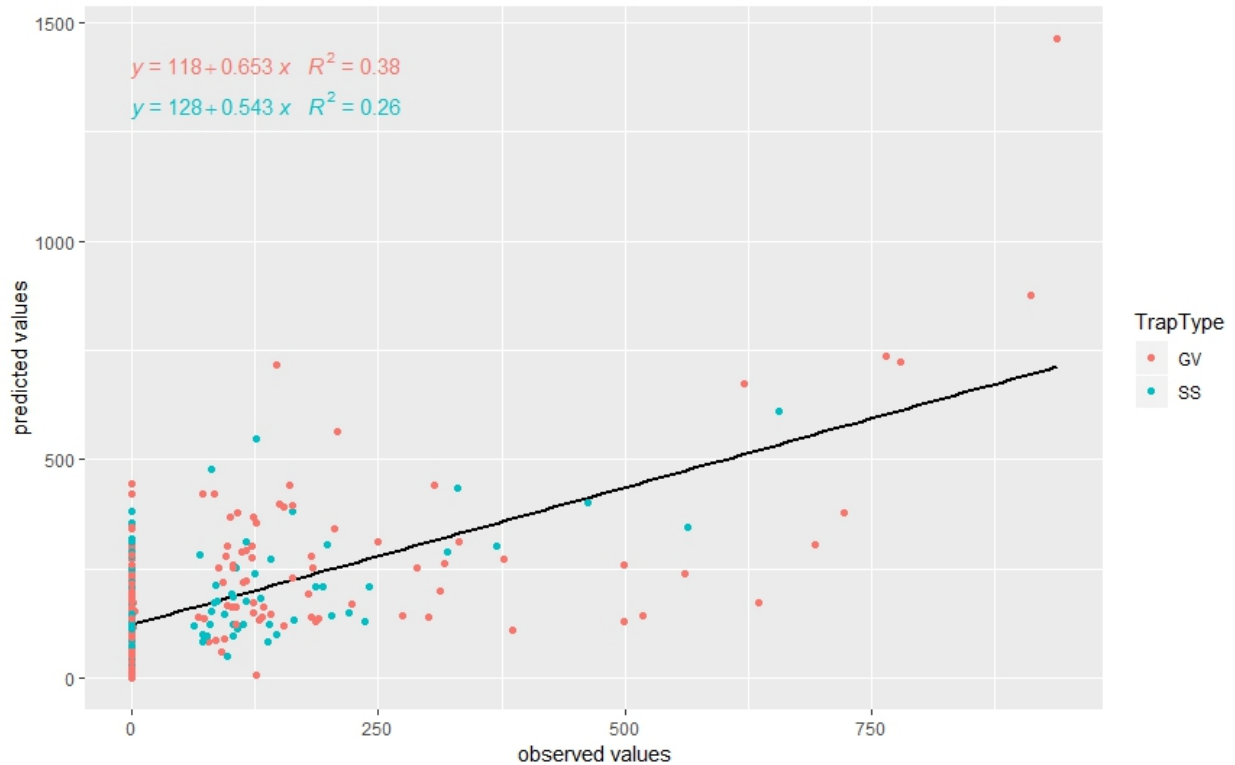


Figure 11 Model Validation Results. The model validation step includes comparing the calculated vector indexes from the 2016-2017 data (“observed values”) and the extracted vector indexes from the map (“predicted values”). The R^2 for each trap type is given. GV = gravid traps (pink), SS = storm-sewer traps (teal).

3.5 Discussion

This study identified significant landscape and socioeconomic factors related to *Cx. quinquefasciatus* abundance and infection with WNV in Harris County, TX, including elevation, impervious surfaces, median income, and percentage of Hispanics in a census tract (Table 6). Landscape determinants were shown to be important factors in the infection of WNV in *Cx. quinquefasciatus* mosquitoes. Elevation had the greatest influence on the model, with an estimate of 44.82 ($p < 0.001$). Harris County’s elevation ranges between 0-310 feet above sea level (Lillibridge et al. 2004). Elevation increases in the northwest direction and decreases when approaching the coastline in the southeast (Figure S3). This gradual increasing elevation in the

northwest direction generally aligns with our model and final risk map (Figure 10). In general, our results for elevation disagree with previous studies from Georgia, Illinois, North Dakota, and South Dakota, where all studies concluded inverse relationships between lower elevation and higher WNV transmission and intensity. In combination with other physiographic elements, higher elevation limits WNV transmission in Georgia where higher elevations have lower temperatures, subsequent smaller mosquito abundances, and greater diversity in avian species composition (Gibbs et al. 2006). Ruiz et al. (2010) found that elevation was a moderate predictor in determining the minimum infection rates of WNV in *Cx. pipiens* in Chicago, IL and suggested that lower elevation could mediate the effect of precipitation by collecting water at lower elevations to allow enrichment of organic materials needed for mosquito population survival. A study in North Dakota found an inverse relationship between the incidence of WNV in horses and elevation (Mongoh et al. 2007). In South Dakota, elevation had a negative relationship with human WNV risk, citing the main vector's (*Cx. tarsalis*) inability to survive at colder temperatures found at higher elevations as the reasoning for the decreased odds of human WNV disease (Chuang et al. 2012).

The differences in our elevation results compared to other studies led us to believe that the extensive flood control systems and storm drains, which follow the elevation gradient may contribute to localized higher vector indexes throughout Harris County. This county is prone to periodic flooding because of its location along the Gulf Coast, abundant rainfall, and relatively low elevation. The county constructed flood control systems comprised of six major bayous and an aging system of storm sewers and underground tunnels to capture floodwaters. The flood control systems may serve as breeding grounds for *Cx. quinquefasciatus*, where stagnant water may collect and become enriched with organic nutrients required for mosquito breeding and

larval development (Lillibridge et al. 2004, Molaei et al. 2007, Curtis et al. 2014). Furthermore, living near bayous was significantly associated with greater odds of human infection with WNV in Harris County (Nolan et al. 2012). This study identified that the highest WNV vector indexes were predicted to occur near White Oak Bayou, Buffalo Bayou and Brays Bayou. The bayous are heavily urbanized and are prone to flooding due to their flat topography. With the amount of heavy urbanization and manmade structures, water cannot be fully absorbed and will pool in the bayous, creating suitable habitats for immature stages of mosquitoes after flooding events. The contribution from flood control systems on WNV transmission intensity warrants further research for Texas.

Impervious surfaces can be used to quantify the degree of urban land use (Arnold Jr and Gibbons 1996). Impervious surfaces represent the amount of human-made materials, which can be used to estimate the extent of artificial structures. Our model found that areas around mosquito traps with greater percentages of impervious surfaces resulted in higher vector indexes. Impervious surfaces prevent rainfall from absorbing into the ground, thereby allowing the accumulation of water and the potential for mosquito habitats (Ruiz et al. 2010). A similar phenomenon was seen in Wisconsin when researchers found that impervious surfaces may contribute to the collection of water in catch basins since these surfaces cannot absorb the water, leading to stagnant water pools and greater abundances of mosquito larvae (Kronenwetter-Koepel et al. 2005). In Chicago, Ruiz et al. (2010) found a relationship between IR and impervious surfaces, where these surfaces can collect stagnant water in low-lying areas and create habitats that are characteristic for *Culex* mosquitoes. Conversely, another study in the same area did not identify impervious surfaces as an important predictor of human WNV illness

(Messina et al. 2011). The differences in their results could be attributed to the uncoupling between mosquito infection and the appearance of human WNV infection.

Notably, socioeconomic factors also emerged as key determinants related to WNV VI's. Median income in Harris County showed a positive association with the WNV VI, with an estimate of 37.83 in our model. According to our study, areas of higher income may have higher vector indexes than areas with lower income possibly due to larger properties that may have more potential mosquito larval habitats, landscapes with abundant container habitats, or yards that may be watered more frequently than lower-income areas, contributing potential larval habitats. In Chicago, people with higher median incomes were more likely to be in clusters of high WNV incidence than those in lower median income areas during the 2002 WNV epidemic (Ruiz et al. 2004). In another study investigating the 2012 WNV outbreak across the country, Degroote et al. (2014) found that higher median income were associated with high WNV ratings in the northcentral and northeast regions. While the authors did not find a significant relationship specifically with median income and WNV incidence in the southeast region, which contains Harris County, another variation of income was significant. Their study found that greater percentages of people living in poverty in the southeast region of the U.S. were associated with more cases of WNV in humans, which contradicts our results (Degroote et al. 2014). It should be noted that socioeconomic status is a single measurement that is conceptualized as a combination of income, education, and occupation to compare the relative economic and social position of one person or family (Green 1970, Mueller and Parcel 1981). Previous studies have used different variations of socioeconomic status, such as the number of foreclosures in an area, abandoned houses, and the type of housing occupation, as predictors for the risk of WNV (Ozdenerol et al. 2008, Reisen et al. 2008a, Chung et al. 2013).

The percentage of Hispanics within a census tract was another significant demographic measurement in our model. Harris County has a high proportion of Hispanics present in the population, with approximately 42.4% of the population in the county identifying themselves with Hispanic origins (United States Census Bureau 2018). Our models identified a significantly positive relationship between higher percentages of the Hispanic population and the WNV VI with an estimate of 43.38. Other studies tasked with identifying demographic variables often utilized the percentage of white and black populations, however, very few studies have specifically included Hispanic populations in their modeling efforts. Therefore, the behaviors of Hispanic populations related to the proliferation and subsequent infection of *Cx. quinquefasciatus* mosquitoes with WNV have yet to be elucidated. Nevertheless, studies investigating associations between the Hispanic population and dengue have contributed to the understanding of the Hispanic population's role in the transmission of mosquito-borne viruses. For example, a study found that Hispanic populations were associated with the higher numbers of containers in North Texas, which can provide habitats for dengue vectors *Ae. aegypti* and *Ae. albopictus* (Lee et al. 2009). *Cx. quinquefasciatus* may also use similar habitats if stagnant water is present in containers.

Harris County used two different trap types consistently during the study period (2005-2015). SS and GV traps did not reveal major differences when calculating the VI (Figure 7). That is, operational areas with high a VI for one trap type often had a high VI in the same area for the other trap type. Both trap types specifically target *Cx. quinquefasciatus* mosquitoes, but they differ in that GV traps are likely to capture gravid female mosquitoes that are ready to lay their eggs and that SS traps are likely to capture female mosquitoes that might not have had a blood meal yet or are in search of a blood meal (Williams and Gingrich 2007). Since female

mosquitoes trapped in the GV trap have already taken a blood meal, they are more likely to be infected with WNV, whereas females from SS traps might not have taken a blood meal and thusly remain uninfected. A comparison of light, gravid, and resting traps were compared for various locations in Delaware and New Jersey, and the authors concluded that the overall infection rate for WNV was 33 times greater for gravid traps compared to the infection rate for light traps (Williams and Gingrich 2007). In addition, the traps differ in their placement. The GV traps are often placed in bushy, shaded areas, while SS traps are placed underground in storm sewers where *Cx. quinquefasciatus* mosquitoes may be found. Despite the differences in how and where the traps are placed, both trap types still produced similar VI's per year (Figure 7).

This 11-year dataset over a large and diverse county yielded several landscape parameters that contributed to the variation in VI, although the fit was low and suggests that our model was not inclusive of additional factors explaining the number of infected *Culex* mosquitoes in the landscape. During our validation step, our best-fit model for the vector index predicted with an accuracy of 20-40%, which could be due to the ubiquitous vector control throughout the county, the ethnic diversity within households and block groups, and the changes in surveillance protocols starting in 2016. In response to WNV-positive mosquito pools, Harris County regularly performs ultra-low volume adulticide spraying by truck in neighborhoods where mosquito pools tested positive for WNV. This mosquito control and occasional aerial applications of adulticides were not considered by the current study. Further research should evaluate the effects of adulticide spraying on the patterns of WNV in Harris County.

In addition, our study did not consider Hispanic populations as a separate race, according to the Census Bureau, but instead the term “Hispanic” is referred to as an ethnicity. That is, individuals can report that they have Hispanic origins in addition to race. The overlap of races

and Hispanic origin in the data resulted in percentages that were sometimes more than 100%. Future work should focus on finding resources that can account for this overlap.

After the threat of Zika found in a traveler from Harris County, TX in 2016, surveillance shifted towards *Aedes spp.* sampling and testing (McCarthy 2016). The majority of the study period biased their sampling methods towards *Culex spp.* collections using GV and SS traps and testing for SLE and WNV. In 2016, however, more BG traps were introduced to sample more *Aedes spp.* mosquitoes and WNV testing was restricted to one pool per trapping event instead of a maximum of three pools, which was the standard prior to 2016.

While not necessarily weaknesses or limitations in our study, the temporal and spatial scales for our covariate layers may have affected the predictive capabilities for 2016-2017. We used data from the 2000 and 2010 Census, which may lose accuracy over time as we move further away from the official census year. While official U.S. Census Bureau data were used, other resources such as the American Community Survey develop yearly estimates for various demographic variables and may serve as a resource for demographic variables for future studies. Our spatial scales for our covariates were also restricted to the census tract level for our demographic variables, which might not necessarily reflect the nuances of local mosquito population dynamics at smaller scales.

Harris County Public Health Mosquito and Vector Control has been an integral program in monitoring and controlling WNV mosquito populations since the arrival of this new arbovirus in 2002 in the third most populous county in the U.S. The extensive surveillance and organizational capacity allowed this long-term spatial analysis of mosquito abundance and infection with WNV. This study highlights features of the landscape and social demographics where WNV is more likely to circulate in vector populations, which will inform future vector

intervention campaigns in this region. Without an available vaccine for humans, mosquito control programs and public awareness campaigns are currently the most effective way to prevent and/or minimize WNV human cases. Understanding the ecology of WNV dynamics with the mosquito, environment, and social demographics plays a key role in effective intervention campaigns. Furthermore, predicting when and where WNV will occur may provide an early warning system, offering an opportunity to control mosquitoes and alert the public with the appropriate messages to reduce the risk of exposure before bridge transmission to humans.

CHAPTER IV

THE ROLE OF WEATHER ON THE TEMPORAL ABUNDANCE AND WEST NILE VIRUS
INFECTION PATTERNS IN *CULEX QUINQUEFASCIATUS* SAY (DIPTERA: CULICIDAE)
IN HARRIS COUNTY, TEXAS

4.1 Synopsis

Early warning systems for vector borne disease (VBD) prediction are an ecological application where data from the interface of several environmental components can be used to predict future VBD transmission. In general, models for early warning systems tend to only consider average environmental conditions ignoring variation in weather variables, despite the prediction from Schmalhausen's law about the importance of environmental variability for biological systems. Here, we present results from a long-term mosquito surveillance program from Harris County, Texas, USA, where we use time series analysis techniques to study the abundance and West Nile virus (WNV) infection patterns in the primary vector, *Culex quinquefasciatus* Say. We found that, as predicted by Schmalhausen's law, mosquito abundance was associated with the standard deviation and kurtosis of environmental variables. By contrast, WNV infection rates were associated with 8-month lagged temperature, suggesting environmental conditions during overwintering might be key for WNV amplification during summer outbreaks. Finally, model validation showed that seasonal autoregressive models successfully predicted mosquito WNV infection rates up to 2 months ahead, but did rather poorly at predicting mosquito abundance, a result that might reflect impacts of vector control for mosquito population reduction, geographic scale, and trap location change artifacts.

4.2 Introduction

Early warning systems of vector-borne diseases (VBDs) are crucial to the effective and efficient control of the disease prior to the appearance of human infections. To develop early warning systems, a complete understanding of the ecology of the disease system and its extrinsic environmental drivers is necessary. Early warning systems have used a variety of methods and data sources such as vegetation and weather in combination with geographic information systems/remote sensing to predict various VBDs including malaria in Africa, American Cutaneous Leishmaniasis in Costa Rica, dengue in Brazil, and West Nile virus (WNV) in the U.S., among many other VBDs (Connor et al. 1999, Craig et al. 1999, Thomson and Connor 2000, Rogers and Randolph 2003, Kuhn et al. 2005, Shaman and Day 2005, Thomson et al. 2006, Chaves and Pascual 2007, Ruiz et al. 2010, Lowe et al. 2013, Manore et al. 2014, Shand et al. 2016).

Most models used in the development of early warning systems for disease prediction have used mean (average) environmental variables as inputs for model development. However, studies suggest that models could be improved by including measurements of environmental variability. For example, it has been observed that higher order statistical moments of environmental variability in weather, such as kurtosis or standard deviation, allow more accurate prediction of abundance in several mosquito species (Hayes and Downs 1980, Shaman and Day 2007, Chaves et al. 2011, Chaves et al. 2012, Chaves 2016, Ng et al. 2018). This prediction follows Schmalhausen's law, the ecological principle stating that organisms are sensitive to not only average patterns, but also to variability patterns (Lewontin and Levins 2000, Chaves and Koenraadt 2010). For instance, organisms are susceptible to variability in their environment when stressed by any single environmental component (Lewontin and Levins 2000, Chaves and

Koenraadt 2010). In principle, environmental variability can be measured by higher order statistical moments, such as the variance, which measures a variable's dispersion around its mean (Figure A1A). Another example is kurtosis, which measures whether a variable is more unpredictable on the extremes of a distribution with respect to the mean, generating a leptokurtic distribution, or if an environmental variable is more unpredictable around the mean, generating a platykurtic distribution (Figure A1B) (Chaves et al. 2011). In general, it will be expected that biological systems are more sensitive to platykurtic environmental components, provided that there is more uncertainty regarding values around a mean, than in a leptokurtic environment, where there is relatively low variability when the environment fluctuates around the mean (Levins 1968). Due to their complex biology, VBDs are excellent model systems to test the hypothesis around Schmalhausen's law, given the confluence of many different organisms that have different degrees of autonomy and interactions with changing environments in both their life cycles and the ecological interactions leading to pathogen transmission (Chaves 2017).

VBDs patterns of interaction with the changing environment might be one of the key components to explain the emergence of new diseases and their successful establishment in new habitats (Levins et al. 1994). Among VBDs, WNV, is a zoonotic disease with an enzootic cycle involving avian amplification hosts and mosquito vectors that recently invaded North America (Weaver and Reisen 2010). Despite the abundance of studies looking at its association with environmental variables (Reisen 1995, Chase and Knight 2003, Reisen et al. 2006b, Reisen et al. 2006a, Brown et al. 2008b, Reisen et al. 2008b, Randolph and Rogers 2010, Reisen et al. 2010, Ruiz et al. 2010, Degroote et al. 2014, Shand et al. 2016), little to no studies inquire about the impacts of environmental variability on its transmission.

WNV is a pathogen that was first introduced to the United States in 1999, and has since spread throughout much of North America. Since its introduction, 46,086 cases of WNV have been reported and 2,017 deaths have been recorded as of 2016 (CDC 2016b). WNV transmission biology consists of avian hosts that amplify virus acquired via infected mosquito bites, and then can infect bloodsucking mosquitoes that can continue transmission among avian hosts or bridge transmission to “dead-end hosts,” such as horses and humans, which are not likely to infect mosquitoes (Weaver and Reisen 2010). *Culex spp.* mosquitoes are the primary enzootic vectors (Turell et al. 2005, Weaver and Reisen 2010), where these mosquitoes serve as a “bridge” between animal and human transmission of WNV (Kilpatrick et al. 2005a, Hamer et al. 2008a, Hamer et al. 2008b).

WNV amplification is highly heterogeneous each season, with periodic outbreak years mixed with low levels of virus transmission, and weather is the suggested key factor driving these patterns (Ruiz et al. 2010, Chung et al. 2013). For example, weather plays a vital role in the abundance of mosquito populations and subsequent pathogen transmission (Chaves 2017). Increasing ambient temperature, up to a point, will increase the rate of development, productivity, and abundance of mosquito populations and decrease the extrinsic incubation period, the time interval between an infectious blood meal until the mosquito is capable of transmitting the virus (Smith 1987, Rueda et al. 1990, Dohm et al. 2002b, Reisen et al. 2006a).

In addition, precipitation is known to have important consequences on mosquito productivity and abundance (Ruiz et al. 2010, Chuang et al. 2011, Degroote et al. 2014), which also influences WNV transmission. However, the influence of prior precipitation on WNV transmission is complex and no clear patterns have emerged from multiple studies (Landesman et al. 2007, Chuang et al. 2012, Chung et al. 2013, Paz and Semenza 2013). Precipitation creates

small pools of water that become enriched, creating suitable oviposition habitats for gravid female mosquitoes (Takeda et al. 2003, Soverow et al. 2009). However, heavy rainfall can flush larval habitats and reduce adult mosquito productivity (Shaman et al. 2002b, Koenraadt and Harrington 2008). Furthermore, drought conditions disrupt the aquatic ecosystem of predators and competitors that serve to limit mosquito larval activity, allowing larvae to fully develop and emerge as adults (Chase and Knight 2003).

Temperature and precipitation can affect the amount of vegetation present. Vegetation can serve as resting habitats for adult mosquitoes, roosting sites for avian hosts that female mosquitoes utilize for a blood meal, and sources of nutrition during the development cycle of the immature stages of mosquitoes (Ward et al. 2005, Brown et al. 2008a, Gardner et al. 2013).

Understanding the ecology of WNV transmission dynamics plays a key role for effective intervention strategies. In central and southeast Texas, the southern house mosquito *Culex quinquefasciatus* Say (Diptera: Culicidae) is the most relevant mosquito species involved in the transmission cycle (Strickman and Lang 1986, Lillibridge et al. 2004, Molaei et al. 2007).

Quantitative predictive models as part of an early warning system for WNV transmission have been developed for certain regions of the U.S., but these types of models have not been parameterized for Texas. Being able to predict when and where WNV may occur in Texas, a hotspot for WNV and other VBDs in the U.S., provides an early warning system and the opportunity to control mosquitoes before bridge transmission to humans and alert the public with the appropriate messages to reduce WNV exposure risk.

Utilizing historical data from Harris County, TX, we examine the influence of weather patterns, including mean conditions and higher order statistical moments like standard deviation (SD) and kurtosis, on the abundance and WNV infection of *Cx. quinquefasciatus* the main WNV

vector in southeast Texas. We hypothesize that annual and seasonal weather patterns affect mosquito biology and WNV transmission dynamics, which contribute to the temporal heterogeneity in the abundance and WNV infection rates of *Cx. quinquefasciatus*. We also expect that previous winter temperatures, which set the conditions for mosquito overwintering (Dohm and Turell 2001, Reisen et al. 2006b, Chung et al. 2013, Chaves et al. 2018), might influence *Cx. quinquefasciatus* WNV infection rates in the subsequent summer, thus creating the expectation of long delays in the association between temperature and WNV infection rates in *Cx. quinquefasciatus*.

4.3 Methods

4.3.1 Study Area

Harris County, TX includes the metropolitan city of Houston and has a population of 4,092,459 according to the 2010 U.S. Census, making it the most populated county in Texas and the third most populated county in the U.S. (United States Census Bureau 2016). The county is located along the Gulf of Mexico, making it an ideal location for international import and export systems for ships, mosquitoes, and vector-borne diseases (Reiter 1998). Its unique location along the Gulf makes the county prone to severe weather such as hurricanes, which result in major flooding events. To counteract flooding events, Harris County has a large flood control system comprised of several different water containment parts, such as bayous, channels, storm drains, and sewers, many of which are aging and rich with organic materials suitable for mosquito breeding (Molaei et al., 2007). Following outbreaks of St. Louis Encephalitis (SLE) virus, a similar arbovirus to WNV, which amplifies in *Culex* mosquitoes and birds with spillover transmission to humans, Harris County first began its mosquito surveillance program in 1965.

Since then, the surveillance program has generated considerable mosquito disease research, expanded their surveillance to include WNV, Dengue virus, and Zika virus, and generated substantial historic records of mosquito trap data (Dennett and Debboun 2017). Other publications have focused on the most populous areas within the I-610 highway loop, which mainly comprises Houston (Rios et al. 2006, Dennett et al. 2007b, Curtis et al. 2014), however, this study will analyze data from the entire county.

4.3.2 Mosquito Data

Harris County Public Health Mosquito and Vector Control (HCPH) conducted mosquito surveillance on a weekly basis between 2002-2016. HCPH consistently used a combination of storm sewer (SS) and gravid (GV) traps for WNV surveillance throughout the county (Figure 12). SS traps are modified CDC Light Traps baited with dry ice and attached to man hole covers underground (Molaei et al. 2007). GV traps are baited with hay infusion water and placed in residential yards, usually under vegetation. The hay infusion is composed of mixing 1.3 kg of Coastal bermudagrass, *Cynodon dactylon* (L.), with 42 gallons of water and then aged for 10-14 days (Dennett et al. 2007a, White et al. 2009). Other trap types were used by HCPH, but only SS and GV traps were analyzed given their ubiquitous usage throughout the study period and the county.

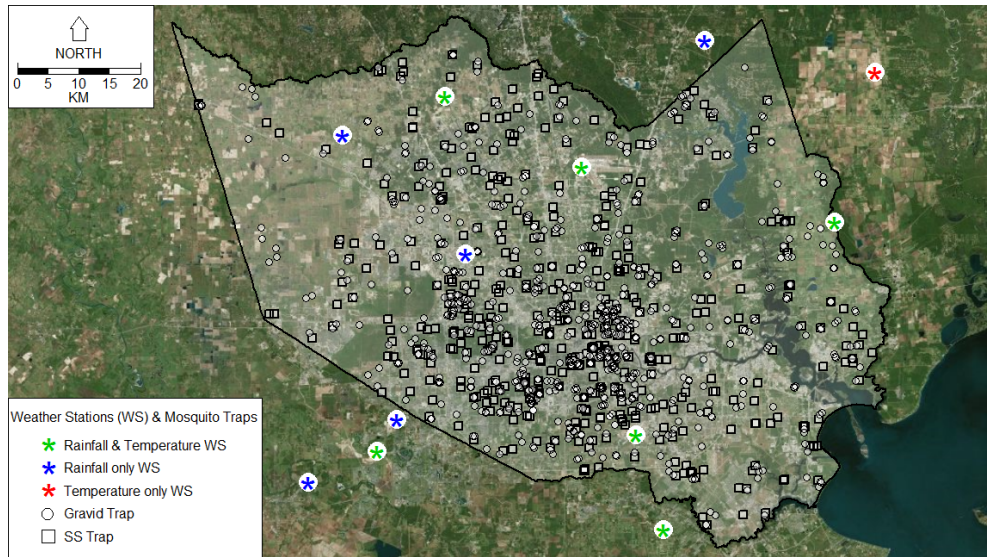


Figure 12 Map of Harris County, weather stations and trap locations. The background map is courtesy of Google Earth - Harris County is highlighted, and the location of mosquito traps, gravid (GV) and storm sewer (SS) traps are indicated with different symbols. Weather stations are color coded according to whether they recorded temperature, rainfall, or both. For details about symbols and color codes, please refer to the inset legend.

The mosquito collection protocol from Harris County has been described in detail elsewhere (Molaei et al. 2007, Curtis et al. 2014). Briefly, traps are placed in the afternoon between 1:30 PM-5:00 PM and then collected the following morning between 7:30 AM-10:30 AM. Traps are placed into “operational areas,” lines that divide the county for surveillance, inspection, surveying, and control purposes (Hunt and Hacker 1984). The 268 operational areas are based on municipal, district, and zip code lines. Live mosquitoes are brought back to the lab and then frozen at -70°C . Mosquitoes are then identified by species and sex on a chill table to preserve the presence of the virus and then sorted into pools of ≤ 50 mosquitoes, with a maximum of three pools per trap. HCPH tested for WNV antigen in mosquito pools using an enzyme-linked immunosorbent assay (ELISA) and positive results were confirmed with a Rapid Analyte Measurement Platform (RAMP) test (Lillibridge et al. 2004, Randle et al. 2016). To be

considered a positive pool, the mosquito pool must test positive on both the ELISA and RAMP test. A positive pool is a mosquito pool that contains at least one *Cx. quinquefasciatus* mosquito positive for WNV. Using data from all the pools tested we estimated monthly infection rates, under the assumption that the diagnostic methods have a sensitivity near 1, using a maximum likelihood estimation method for unequal pool size that is fit with a log link generalized linear model (Farrington 1992) and confidence intervals that are estimated by inverting a likelihood ratio test (Speybroeck et al. 2012).

When generating the time series, we needed to input missing values for December 2003 and January 2004, when no traps were deployed by HCPH, which was done via interpolation using a loess regression as described by Ng et al. (2018).

4.3.3 Weather and Vegetation Data

For this study, we acquired data for global climatic indices and local weather for Harris County, TX. To evaluate the impact of global climatic phenomena on *Cx. quinquefasciatus* abundance and its WNV infection rate, we downloaded monthly data for the El Niño 3.4 index from the U.S. National Oceanic and Atmospheric Administration (NOAA) Climate Prediction Center (NOAA). The El Niño 3.4 index is associated with interannual rainfall dynamics in Texas (Li and Kafatos 2000), is based on the Extended Reconstructed Sea Surface Temperature (Huang et al. 2017), and corresponds to sea surface temperatures measured in the area delimited by 5°N–5°S and 170°W–120°W of the Pacific Ocean.

To evaluate the impact of local climatic indices in our data, we used data from weather stations located inside Harris County or neighboring counties (Figure 12). We used the Climate Data Explorer from the Royal Netherlands Meteorological Institute (KNMI) to download daily

weather data, looking for stations that had at least 10 years of data. We specifically selected the following weather stations (coordinates and Global Historical Climatological Network Code) (Figure 12): Baytown (29.91°N, -94.99°E, USC00410586), Clover Field (29.52°N, -95.24°E, USW00012975), Hobby Airport (29.64°N, -95.28°E, USW00012918), Houston Intercontinental Airport (29.98°N, -95.36°E, USW00012960), Hooks Airport (30.07°N, -95.56°E, USW00053910), and Sugarland (29.62°N, -95.66°E, USW00012977), which had both temperature and rainfall records for our study period. Stations that had data for only rainfall included: Cypress (30.02°N, -95.71°E, USC00412206), New Caney (30.14°N, -95.18°E, USC00416280), North Houston (29.87°N, -95.53°E, USC00414327), Richmond (29.58°N, -95.76°E, USC00417594), and Westbury (29.66°N, -95.63°E, USC00414325). Data for only temperature was available at Dayton (30.10°N, -94.93°E, USR0000TDAY).

We processed the daily data to generate monthly time series for the study period. We specifically computed the monthly mean, SD, and kurtosis for temperature and rainfall in Harris County. For comparison we also downloaded gridded weather data from GHCN/CAMS 2 m (temperature) (NOAA) and GPCP (rainfall) (NOAA), with resolutions of 0.5° and 0.25°, respectively.

We downloaded monthly images for vegetation indices with a 1-km resolution vegetation (M*D13A3) product (Didan 2015). We specifically downloaded images for the normalized difference (NDVI) and enhanced (EVI) vegetation indices, which are proxies for vegetation growth (Pettorelli et al. 2005). The images, which are courtesy of the NASA Land Processes Distributed Active Archive Center (LP DAAC), USGS/Earth Resources Observation and Science (EROS) Center (Sioux Falls, South Dakota), were downloaded from the server (NASALPDAAC) using the package *MODISsp* for the software R (Busetto and Ranghetti

2016). Each image was then clipped to the surface of Harris County, and then stacked into a geotiff using the package *raster* for R (Brunsdon and Comber 2015). Then for each monthly image we estimated the mean, SD and kurtosis for NDVI and EVI during the study period.

4.3.4 Statistical Analysis

4.3.4.1 Seasonality

Seasonal profiles for all the mosquito time series, vegetation, and weather variables were built using monthly boxplots (Venables and Ripley 2002).

4.3.4.2 Non-Stationary Patterns of Association in the Time Series

We studied the association of cycles in the time series using a cross wavelet coherence analysis to identify non-stationary association patterns (i.e., changes through time) and the association between cycles in the time series, or coherence, whose period might be variable and not repetitive or seasonal (Chaves and Pascual 2006, Cazelles et al. 2007). We used this technique to study the association between mosquito abundance and infection with the El Niño 3.4 index, NDVI, EVI, temperature and rainfall.

4.3.4.3 Time Series Modeling

To fit and select variables for monthly time series models of mosquito abundance and WNV infections in pools we used a standard protocol for the time series analysis (Hurtado et al. 2014, Hurtado et al. 2018). The first step consists of assessing the correlation of each time series with itself by inspecting the autocorrelation function (ACF) as well as the correlation of consecutive time lags using a partial autocorrelation function (PACF) (Shumway and Stoffer

2011). Information from the ACF and PACF will identify a null model that considers the autocorrelation structure of the focal time series. This null model was then used to pre-whiten the times series with the Kalman filter. Pre-whitening is a process to remove a common autocorrelative structure that can generate spurious correlations from the climate, weather and vegetation indices (Shumway and Stoffer 2011). Then residuals from the autonomous model and the pre-whitened time series were used to estimate cross correlation functions (CCFs), which show the correlation between two time series as a function of fixed time lags (Hoshi et al. 2014). Once we identified significant lags of the covariates between 1 and 12 months ($P < 0.05$), the lags were then used to fit full models of mosquito abundance and mosquito WNV infection. Models were simplified by model selection through backward elimination (Kuhn and Johnson 2013), following the minimization of the Akaike information criterion (AIC) (Faraway 2016). This process allows model selection among models with similar parameter numbers (Faraway 2016). For the best models we further tested if variables, whose parameters were not significant, could be eliminated using likelihood ratio tests (Faraway 2016), and the resulting models are reported as the best models in the Results section. For the best models, we verified time series model assumptions using standard procedures for the time series analysis (Shumway and Stoffer 2011).

4.3.4.4 Time Series Model Validation

We validated the time series models by leaving one year of observations, 2016, out of model fitting and then forecasting mosquito abundance and WNV infection rates at time steps of 1, 2, 3, 4, 6 and 12 months, and then estimating the predictive R^2 (Chaves and Pascual 2007), which is defined as the variance normalized mean square error of the prediction, in other words:

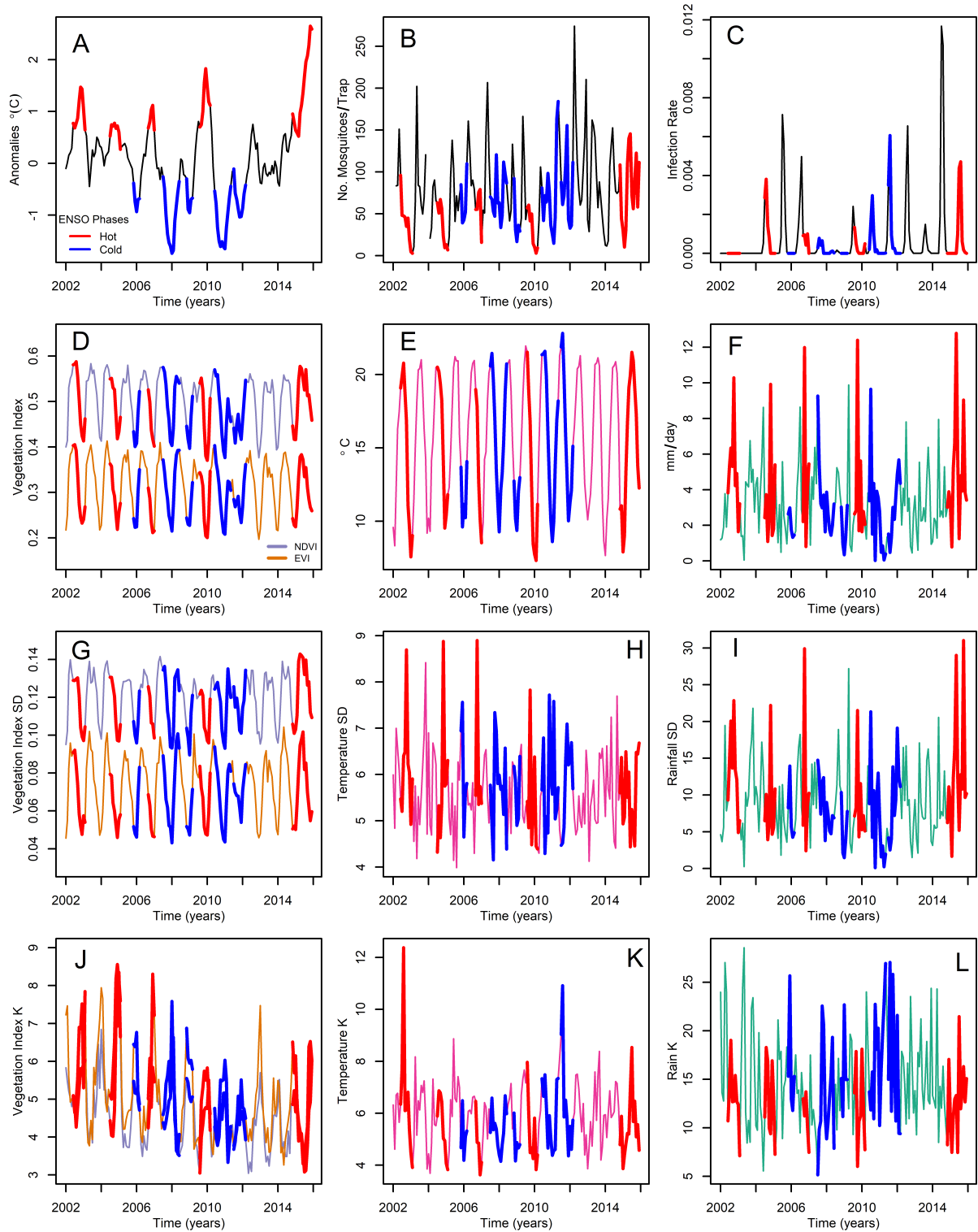
$$_{\text{pred}}R^2=1-(\text{mean square error/variance of the series}). \quad (4.1)$$

The predictive R^2 has a straightforward interpretation, where a $_{\text{pred}}R^2$ of 1 indicates perfect forecasts, but negative values, or near 0, indicate a poor predictive ability (Chaves and Pascual 2007).

4.4 Results

Data time series are presented in Figure 13, where color codes are used to represent the phases of the El Niño Southern Oscillation (ENSO). During the study period, the most extreme ENSO event occurred in 2016, as shown by the El Niño 3.4 index time series (Figure 13A).

Figure 13 Monthly Time Series. (A) Sea Surface Temperature in the El Niño 3.4 region, (B) Average number of mosquito per trap, (C) West Nile virus mosquito infection rate, (D) Vegetation indices, including the Normalized Difference Vegetation Index (NDVI) and the Enhanced Vegetation Index (EVI), (E) Average temperature, (F) Average rainfall, (G) Standard Deviation (SD) of NDVI and EVI, (H) SD of temperature, (I) SD of rainfall, (J) Kurtosis (K) of NDVI and EVI, (K) K of Temperature, (L) K of Rainfall. In all panels ENSO phases are highlighted by colors, for details, see inset legend of panel A. Panels B and C are based on combined data from gravid and storm sewer traps. In panels D, G and J NDVI and EVI are differentiated by color, see inset legend of panel D for details.



A total of 686 and 476 locations for GV and SS traps were used throughout Harris County, respectively. During the study period, 15 GV traps were deployed over 450 times at the same location and 24 SS traps were deployed over 500 times at the same location (Figure 12). A total of 10,533,033 female *Cx. quinquefasciatus* were collected using GV (5,371,840 mosquitoes, 51% of the samples) and SS (5,161,193 mosquitoes, 49%) traps. The total sampling effort was 130,567 trap-nights, with 55% of the sampling effort coming from GV traps (71,849 trap-nights) and the remaining 45% coming from SS traps (58,718 trap nights). Monthly mosquito abundance, based on combined GV and SS trap collections, was highly variable (Figure 13B), having an average (\pm SD) of 74.84 ± 47.89 . Mosquito abundance peaks were observed when ENSO was not going through its extreme hot and cold phases, a pattern also observed for the time series based on GV (Figure A2A) and SS (Figure A2B) traps.

Even though WNV infections in humans were first detected in 2002, WNV infections in mosquito pools (Figure 13C) were first detected in Texas in 2004. The monthly average WNV infection rate (\pm SD), estimated only for months with positive pools, was 1.58 ± 2.35 , per 1,000 mosquitoes. As observed with mosquito abundance, WNV infection peaks were observed at times when ENSO activity did not go through its cold and hot extremes, but overall, tended to follow cold ENSO peaks (Figure 13C). Unlike what was observed for mosquito abundance, temporal patterns of mosquito infection were nearly identical when comparing infection estimates from GV (Figure A2C) and SS (Figure A2D) traps. For this reason, we used the WNV infection time series based on GV and SS traps for subsequent analyses. NDVI and EVI had similar temporal patterns during the study period (Figure 13D), with data suggesting that vegetation growth at Harris County slows down during the cold ENSO phase. Meanwhile, temperature (Figure 13E) is higher during the cold ENSO phase, and the estimate using station

data was very similar to the data from the gridded database (Figure A2E). Rainfall peaked during the hot ENSO phase followed by the cold phase (Figure 13F), but this temporal pattern was not so clear for the gridded rainfall (Figure A2F). Given the more apparent patterns of ENSO impacts on the weather observed from weather stations, we used this data for subsequent analyses.

The SD of the vegetation indices (Figure 13G) were similar to the mean time series of the vegetation indices. By contrast, the SD of temperature (Figure 13H) and rainfall (Figure 13I) reached maximum values during the hot ENSO phase, followed by the cold phase. The vegetation indices (Figure 13J) and temperature (Figure 13K) were more leptokurtic during the ENSO hot and cold phases, meaning that most of the variability occurred around the mean value than in the extremes, and more platykurtic when ENSO was not passing through an extreme phase. In contrast, rainfall (Figure 13L) was most leptokurtic during the ENSO cold phase and most platykurtic during the ENSO hot phase.

Seasonal patterns of mosquito abundance for *Cx. quinquefasciatus*, based on both GV and SS traps, (Figure 14A) were bimodal having a large peak in May and a second smaller peak in November. When separating the abundance by trap type, this bimodal pattern was not observed in GV traps, which had a single peak in May (Figure A3A). However, the bimodal seasonality was observed in SS traps (Figure A3B), which had peaks in May and November. Given these marked differences in abundance between GV and SS traps, we decided to perform time series analyses of the combined abundance time series, but also of mosquito abundance based on GV and SS traps separately.

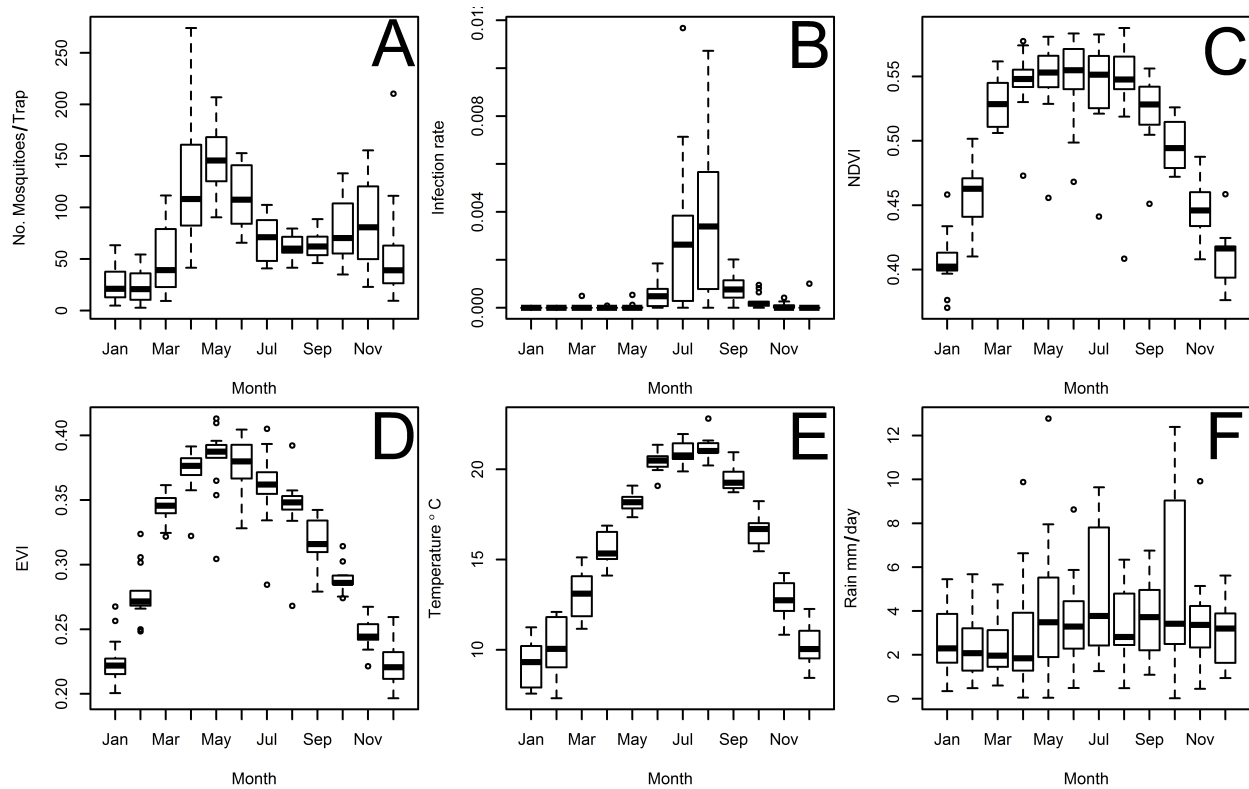


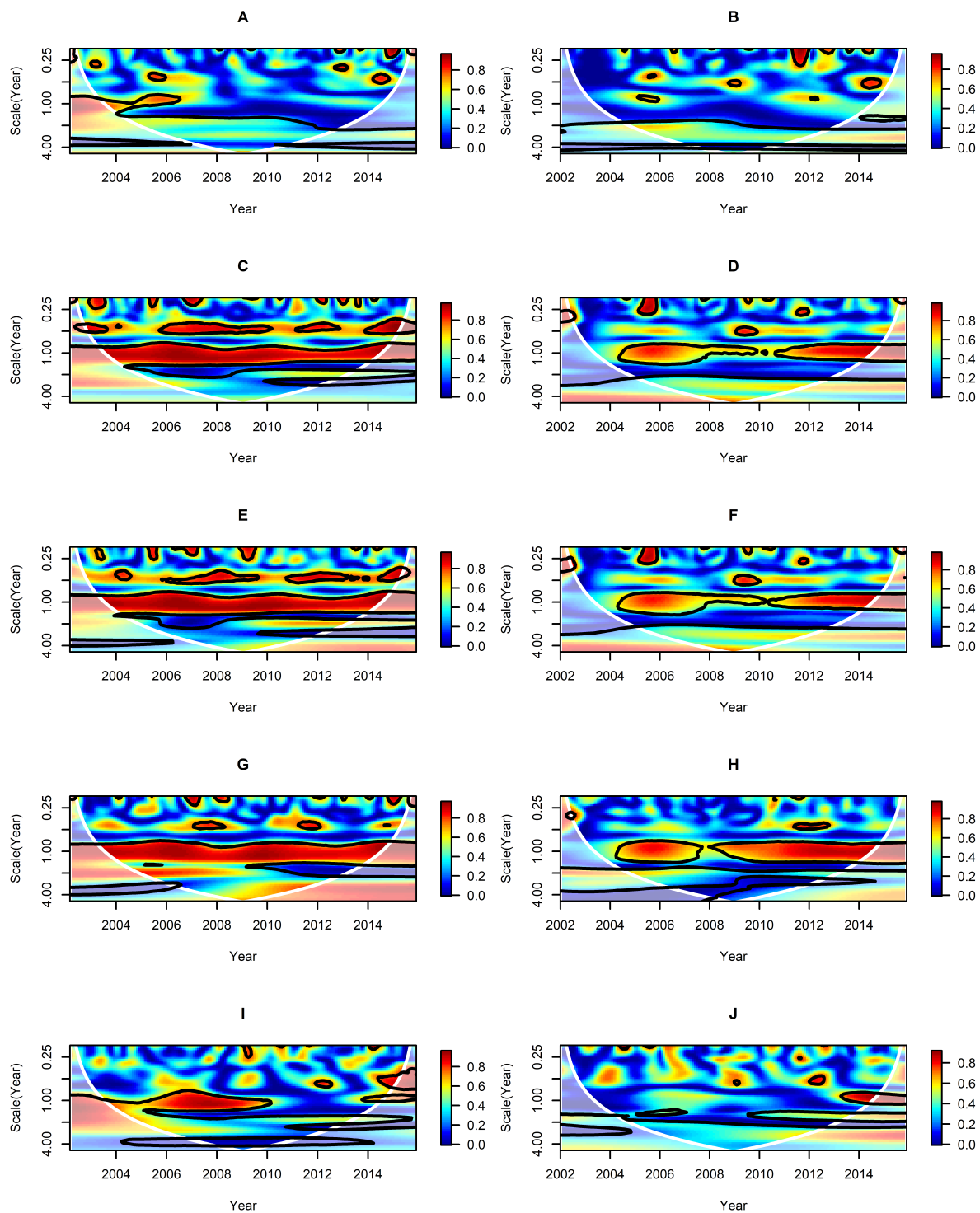
Figure 14 Seasonal Monthly Boxplots. (A) Average number of mosquitoes per trap, (B) West Nile virus mosquito infection rate, (C) Normalized Difference Vegetation Index (NDVI), (D) Enhanced Vegetation Index (EVI), (E) Average temperature (F) Average rainfall. Panels A and B are based on combined data from gravid and storm sewer traps.

Seasonal WNV infection patterns were unimodal with a seasonal peak in August (Figure 14B), a pattern also observed separately for GV (Figure A3C) and SS (Figure A3D) traps. NDVI has a seasonal peak from April to August (Figure 14C), while EVI (Figure 14D) has a unique peak in May. Temperature also had a unimodal pattern (Figure 14E), with a peak in August, which was also observed in the gridded temperature data (Figure A3E). Rainfall had two seasons, one relatively dry from January to April, and a wet season for the rest of the year, with August being consistently the driest month during the wet season (Figure 14F), a similar pattern was also observed in the gridded data (Figure A3F).

The cross wavelet coherence analyses show that interannual cycles, with a period between 3 and 4 years, of mosquito abundance (Figure 15A) and WNV mosquito infection rates

(Figure 15B) were coherent with those observed in ENSO. Meanwhile, NDVI and EVI had seasonal cycles, with periods of 1 year, associated with mosquito abundance (Figure 15C, 15E) and WNV infection rate (Figure 15D, 15F). Temperature cycles were both seasonal and interannual, with cycles of 2 to 4 years, coherent with mosquito abundance (Figure 15G) and WNV infection rates (Figure 15H). Meanwhile rainfall cycles were associated at an interannual scale, with cycles of 3 to 4 years, with cycles of mosquito abundance, which between 2002 and 2010 were also highly coherent at the seasonal scale with rainfall, (Figure 15I) and with WNV infection rates (Figure 15J).

Figure 15 Cross Wavelet Coherence Analysis. Coherence between Sea Surface Temperature 3.4 and (A) Monthly average mosquito abundance per trap (MAMAPT), (B) West Nile virus mosquito infection rate (WNV MIR). Normalized Difference Vegetation Index (NDVI) and (C) MAMAPT, (D) WNV MIR. Enhanced Vegetation Index, EVI and (E) MAMAPT, (F) WNV MIR. Temperature and (G) MAMAPT, (H) WNV MIR. Rainfall and (I) MAMAPT, (J) WNV MIR. In all plots, the y-axis represents the scale, or period measured in years, at which two time series are coherent, while the x-axis represents time. A guide for coherence values is presented to the right of each panel. Coherence goes from zero (blue) to one (red). Red regions in the plots indicate frequencies and times for which the two series share power (i.e., variability). The cone of influence (where results are not influenced by the edges of the data) and significantly coherent ($p < 0.05$) scales through time are indicated by solid lines. MAMAPT and WNV MIR are based on combined data from gravid and storm sewer traps.



The autocorrelation functions of mosquito abundance (Figure 16A) and WNV infection rates (Figure 16B) suggested that both time series were at most second order, i.e., autocorrelated up to the second lag, and seasonal, i.e., being significantly correlated at lag 12 months, meaning time series were autocorrelated at any time lag with itself 12 months before. That autocorrelation structure was observed using the partial autocorrelation function of both mosquito abundance (Figure 16C) and WNV infection rates (Figure 16D).

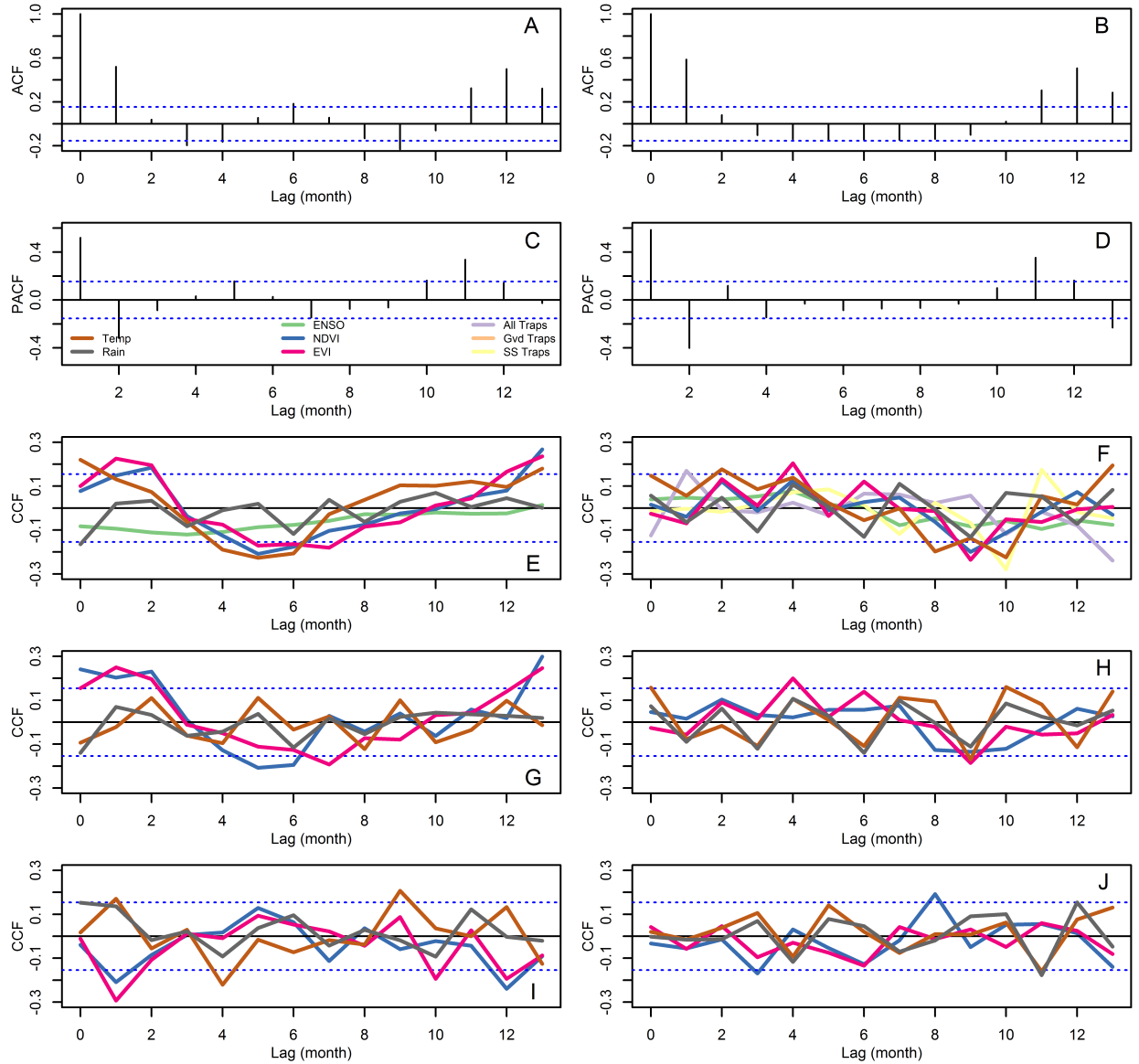


Figure 16 Auto-Correlation Functions (ACF). (A) Monthly average mosquito abundance per trap, MAMAPT and (B) West Nile virus mosquito infection rate, WNV MIR. Partial Auto-Correlation Functions (PACF) of (C) MAMAPT and (D) WNV MIR. Cross-Correlation Functions (CCF) of the average value of environmental variables with (E) MAMAPT and (F) WNV MIR. CCF of the Standard Deviation (SD) of environmental variables with (G) MAMAPT and (H) WNV MIR. CCF of the Kurtosis (K) of environmental variables with (I) MAMAPT and (J) WNV MIR. In panels E to J, environmental variables are color coded, for details, please refer to the inset legend of panel D. MAMAPT and WNV MIR are based on combined data from gravid and storm sewer traps.

With this information, a seasonal autoregressive model was fitted as the null model with the following form:

$$x_t = \mu + \varphi_1 x_{t-1} + \varphi_2 x_{t-2} + \varphi_{12} x_{t-12} - \varphi_1 \varphi_{12} x_{t-13} - \varphi_2 \varphi_{12} x_{t-14} + \varepsilon_t \quad (4.2)$$

where μ is the mean of the time series $x_t = y_t - \mu$, where y_t is either monthly mosquito abundance or WNV infection rates, t indicates time, and ε_t is a normally and identically distributed error. Model selection for mosquito abundance, the model presented in (4.2), suggested that the following model:

$$x_t = \mu + \varphi_1 x_{t-1} + \varphi_{12} x_{t-12} - \varphi_1 \varphi_{12} x_{t-13} + \varepsilon_t \quad (4.3)$$

was the best null model for abundance estimates based on the combined SS and GV data. This null model was used to pre-whiten the time series of the weather and vegetation covariates, which were then used to estimate cross-correlation functions between the average values of the covariates with mosquito abundance (Figure 16E) and WNV infection rate (Figure 16F), the SD of the covariates with mosquito abundance (Figure 16G) and WNV infection rate (Figure 16H), and the kurtosis of the covariates with mosquito abundance (Figure 16I) and WNV infection rate (Figure 16J). We also estimated the ACF and PACF of mosquito abundance with GV (Figure A4A, A4C) and SS traps (Figure A4B, A4D), and the cross-correlation function of mean, SD, and kurtosis of the covariates with GV (Figure A4E, A4G, A4I) and SS (Figure A4F, A4H, A4J) traps.

The variables that were significantly associated with mosquito abundance were then considered in a full model:

$$x_t = \mu + \varphi_1 x_{t-1} + \varphi_{12} x_{t-12} + \varphi_{13} x_{t-13} + \sum cov_{t-j} + \varepsilon_t \quad (4.4)$$

that included covariates (*cov*) with time lags $j \geq 0$. The process of model selection for the mosquito abundance model is presented in Table S1, and the process for model selection of mosquito abundance model based only on GV traps in Table S2, and for the model based only on SS traps in Table S3.

Parameter estimates for the best mosquito abundance model are presented in Table 7. Parameters included a positive association with the standard deviation of NDVI (2-month lag) and temperature kurtosis (9-month lag). Meanwhile abundance was negatively associated with rainfall (no time lag), NDVI kurtosis (12-month lag) and EVI kurtosis (1-month lag). Significant parameters in the best models for mosquito abundance based on GV and SS traps separately (Table S4) had similarities with the model for data from both traps (Table 7). Both of those models did not have a significant seasonal autoregressive parameter, i.e., both time series were not significantly autocorrelated with themselves with a 12-month lag. Interestingly, both of these models (Table S4) were associated with EVI kurtosis with 1 month of lag, the association being negative like in the model of Table 7. Other parameters shared with the model presented in Table 7 also had the same sign such as the kurtosis of NDVI with a 12-month lag and a 9-month lag temperature kurtosis for the model based on SS traps (Table S4). Other parameters included variables that were not included in the best model presented in Table 7, and included both mean, SD, and kurtosis parameters (Table S4).

Table 7 Parameter estimates for the best time series model explaining changes in *Culex quinquefasciatus* abundance sampled with gravid and storm-sewer traps in Harris County, TX. For parameters, Intercept represents the mean value of the time series, AR = autoregressive term, SAR = seasonal autoregressive term, Rainfall = average rainfall, NDVI = Normalized Difference Vegetation Index, EVI = Enhanced Vegetation Index, SD = standard deviation, Temperature = average temperature, K = kurtosis. Lags are in months. σ^2 is the variance of the time series.

Parameters (lag)	Estimates	Standard Error
Intercept	74.1795	6.3822
AR (1)	0.3873	0.0790
SAR (12)	0.3414	0.0822
Rainfall (0)	-2.6398	1.0384
NDVI SD (2)	792.7775	379.7516
NDVI K (12)	-14.7881	4.8280
EVI K (1)	-8.8210	3.7416
Temperature K (9)	7.78873	2.4330

$\sigma^2 = 1045$; Log likelihood = -705.7; AIC = 1429.4

The best model for mosquito WNV infection rates (Table 8) was a second order seasonal autoregressive model, i.e., with an autoregressive component similar to the one described in equation 4.2, with mean temperature at an 8-month lag as a significant covariate. The process of model selection is presented in Table S5.

Table 8 Parameter estimates for the best time series model explaining changes in West Nile virus infection rate of *Culex quinquefasciatus* in Harris County, TX. For parameters, Intercept represents the mean value of the time series, AR = autoregressive term, SAR = seasonal autoregressive term, Temperature = average temperature. Lags are in months. σ^2 is the variance of the time series.

Parameters (lag)	Estimates	Standard Error
Intercept	0.0008	0.0001
AR (1)	0.7620	0.0787
AR (2)	-0.3199	0.0797
SAR (12)	0.4757	0.0946
Temperature (8)	0.0003	0.0001

$\sigma^2 = 1.37e^{-6}$; Log likelihood = 765.73; AIC = -1519.47

Finally, the process of model validation suggested the predictive ability of the mosquito abundance model was overall low (Figure 17A), a pattern shared with the models based on GV (Figure A5A) and SS (Figure A5B) traps separately, which nevertheless outperformed the model combining the data from both types of traps. By contrast, the predictive accuracy of the WNV infection rate (Figure 17B) model was high for 1 (80%) and 2 (60%) months, negative at 3 months, and overall decreased as the prediction step increased the number of months predicted at once.

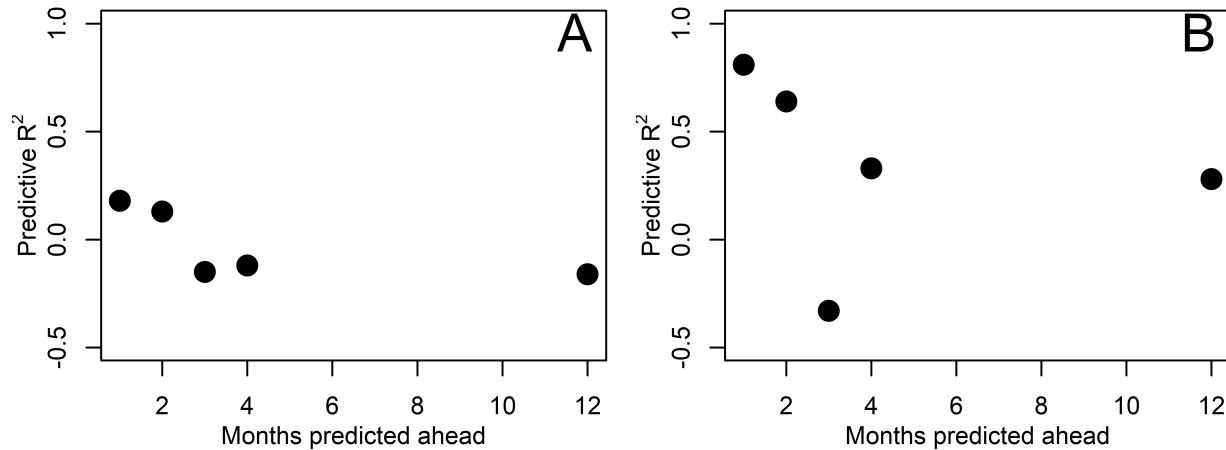


Figure 17 Predictive R^2 . Predictive capability of models selected as best to explain (A) Monthly average mosquito abundance per trap (B) West Nile Virus mosquito infection rate.

4.5 Discussion

Our study found significant weather factors and measurements of their variability were significantly associated with *Cx. quinquefasciatus* abundance and WNV infection rates during the study period (2002-2016) in Harris County, TX. Mosquito abundance generally peaked following the ENSO cold phases when ENSO activity did not go through extreme hot or cold phases (Figure 13B). During the hot ENSO phase, we generally saw peaks in rainfall and greater variation in temperature and rainfall (Figure 13). On the other hand, the cold ENSO phases were characterized by hotter temperature peaks and less rainfall, which resulted in less vegetation growth in Harris County (Figure 13). During these extreme hot and cold ENSO phases, we found lower *Cx. quinquefasciatus* abundance, which could be due to the excess rainfall and higher temperatures/low vegetation in the hot and cold phases, respectively. The increased amount of precipitation during the hot ENSO phase might wash out larval habitats for *Cx. quinquefasciatus* above- and belowground (Shaman et al. 2002b, Koenraadt and Harrington 2008). This phenomenon was true for *Cx. quinquefasciatus* in the U.S. as observed in California (Heft and

Walton 2008) and Georgia (Chaves and Kitron 2011, Nguyen et al. 2012) and *Cx. pipiens* in Illinois (Hamer et al. 2011). Extremely high temperatures are known to decrease the life span of the mosquito and prematurely kill mosquitoes before they are able to transmit the virus to a new host (Reisen 1995, Reisen et al. 2006a, Brault 2009). Vegetation is required for larval development of *Cx. quinquefasciatus* as it provides a source of organic matter and nutrients. The importance of vegetation has been investigated in other areas of the U.S. such as the cities of New York, Chicago and Houston, where the presence of vegetation was positively associated with human risk for WNV (Brownstein et al. 2002, Ruiz et al. 2004, Nolan et al. 2012).

Our results demonstrate that increased variability in both temperature and rainfall result in higher abundances of mosquitoes. Measurements of variability were significant covariates in the abundance models (Table 7). The significant covariates in the abundance model further highlight the importance in including measurements of environmental variability to investigate association patterns between mosquito abundance dynamics and the weather. The covariates for the best mosquito abundance model that combined mosquito counts from both GV and SS traps included positive associations with the standard deviation of NDVI with a 2-month lag and the kurtosis of temperature with a 9-month lag. Rainfall with no lag, NDVI kurtosis with a 12-month lag, and EVI kurtosis with a 1-month lag had a negative association with mosquito abundance. The phenomenon emphasizing the importance of significant variation in weather and vegetation on mosquito abundance follows Schmalhausen's Law, the biological principle stating that organisms are sensitive to both average environmental conditions and environmental variability, which has been previously reported for *Cx. quinquefasciatus* and other vectors of disease (Hayes 1975, Hayes and Hsi 1975, Hayes and Downs 1980, Chaves and Koenraadt 2010, Ng et al.

2018). Therefore, the more neutral conditions seen when ENSO is not going through extreme hot and cold phases may allow for greater abundances of *Cx. quinquefasciatus*.

Interestingly, our study did not include temperature within the same summer season as a significant variable, which other studies have found among other mosquito species (Degaetano 2005, Chuang et al. 2011). Rather, temperature with an 8-month lag was a significantly positive covariate in our WNV infection rate model (Table 8). Given that infection rates generally peak around August in Harris County (Figure 14B), warmer temperatures during the winter are expected to increase the infection rates the following summer. In general, warmer winter seasons preceding a WNV season has been a significant factor of interest in other studies using various measurements of WNV, including mosquito abundance of different *Culex* species, infection rates/vector indexes, and human cases (Reisen et al. 2010, Chung et al. 2013, Degroote et al. 2014, Wimberly et al. 2014).

One mechanism for increased infection rates in the summer following a mild winter is that warmer temperatures in the winter allow *Cx. quinquefasciatus* to remain active gonotrophically and maintain their populations. Alternatively, *Cx. quinquefasciatus* can survive through the winter by entering quiescence when temperatures drop, but can become active once temperatures increase again (Eldridge 1968, Reisen et al. 1986, Diniz et al. 2017). Quiescence is a period of non-seasonal dormancy characterized by slowed metabolism in response to environmental stimuli (Clements 1992). Since *Cx. quinquefasciatus* do not enter diapause and are not hormonally-controlled to enter a state of dormancy, physiological activity can be restored once the stimulus that induces quiescence ceases (Vinogradova 2007, Denlinger and Armbruster 2014, Lacour et al. 2015, Diniz et al. 2017). The sustained activity in mosquito populations through warmer winter temperatures also permit the dissemination of WNV throughout the

vector and consequently vertically transmit WNV to their progeny, thereby maintaining the virus in the vector population (Dohm and Turell 2001, Nasci et al. 2001a, Goddard et al. 2003, Nelms et al. 2013). For example, when *Cx. pipiens* is inoculated with WNV and held at reduced temperatures (10°C) for 21-42 days, the virus is not fully disseminated. Once the mosquito is transferred to an incubation temperature of 26°C, the dissemination rates increased (Dohm and Turell 2001). Studies on the effects of overwintering in *Cx. quinquefasciatus* and WNV infection in Texas are worth investigating.

Another mechanism for increased WNV infection rates during a warm winter relates to the opportunistic feeding patterns of *Cx. quinquefasciatus*, which more frequently feed on avian hosts (Molaei et al. 2007). Warmer winter temperatures can signal the arrival of an early spring, allowing birds to initiate recruitment of young earlier (Forchhammer et al. 1998, Walther et al. 2002). Consequently, increased populations of susceptible juvenile birds are known to fuel the amplification of WNV (Hamer et al. 2008b). Mosquito feeding may coincide with warmer temperatures in the winter, allowing mosquitoes to become infected even during periods of expected low activity since birds may still be viremic or become infected from exposure to feces containing WNV (Eldridge 1968, Dawson et al. 2007, Hinton et al. 2015).

An interesting observation was the difference in mosquito abundance between GV and SS traps. GV traps exhibited a unimodal abundance distribution, however, SS traps showed a bimodal distribution (Figure A2). The second peak in abundance of *Cx. quinquefasciatus* trapped in SS traps during November could be attributed to the life history of this mosquito species. SS traps are placed underground in storm sewers and baited to capture host-seeking mosquitoes. However, *Cx. quinquefasciatus* will also use storm sewers as hibernacula or shelter during cooler months to overwinter into the next spring season (Strickman and Lang 1986). With this in mind,

the second peak in the abundance in November could be attributed to the mosquito's retreat into underground storm sewers to avoid harsh winter conditions since this species does not enter diapause, but instead undergoes quiescence when retreating to storm drains (Reisen et al. 1986, Strickman and Lang 1986, Strickman 1988, Tesh et al. 2004b, Reisen et al. 2010, Nguyen et al. 2012, Reisen 2012, Nelms et al. 2013). These dissection studies investigating overwintering techniques in California have demonstrated that *Cx. quinquefasciatus* mosquitoes undergo quiescence rather than diapause, which is seen in *Cx. pipiens* and *Cx. tarsalis*. However, this type of study, to the best of our knowledge, has not been performed in Texas and warrants further consideration to elucidate overwintering patterns for mosquitoes found in storm drains.

The abundance models for this study performed poorly (Figure 17A), but the infection rate model performed well when predicting between 1-2 months ahead (Figure 17B). The low predictive ability of the abundance model can be attributed to unavoidable logistical constraints that emerge in large-scale vector surveillance systems such as in Harris County, TX. For example, our model did not take into account the mosquito control efforts by HCPH that may have affected local mosquito populations. Mosquito control in Harris County consists of aboveground-based ultra-low volume (ULV) adulticiding in response to a positive pool. Within the same week of detecting a positive pool, ULV adulticiding will occur in the operational area (Fredregill et al. 2011). During peak seasons, spraying may occur more than once a week, which may affect the abundance of mosquitoes trapped by SS and GV traps that target adult mosquitoes.

Another challenge is related to the temporal and spatial scales of our study. Given the long temporal range of our data, we used a monthly scale for the time series analysis. Having a temporal scale of weekly data would better reflect the finer nuances in mosquito abundance and

improve model predictive ability (Chaves et al. 2013, Chuang et al. 2017). Spatial scale is also an important factor when considering infection data since results and conclusions may differ depending on the scale chosen for the study (Winters et al. 2010). We summarized data over a large spatial scale, with Harris County covering 4,600 km². At smaller spatial scales, we might have been able to better capture local population dynamics, as observed in more finely grained studies on mosquito population dynamics (Chaves et al. 2013, Ng et al. 2018).

Another factor to consider when explaining the low predictive ability of our mosquito abundance models is the movement of trap locations throughout the county during the study period. A total of 686 and 476 trap locations were used for GV and SS traps, respectively. Throughout the study period, only 15 GV traps and 24 SS traps remained in the same location. In contrast, 392 GV traps and 324 SS traps were deployed less than 50 times at the same location, which demonstrates the lack of consistency in trap locations throughout the study period (Figure A6). Inconsistencies from trapping may lead to artifacts and biases that do not necessarily reflect local population dynamics of the previous trap locations. The location of the trap influences the mosquito abundance estimates, which may explain the low predictive capability of the abundance models given that many of these traps moved throughout the study period (Brown et al. 2008c).

Finally, our study demonstrates the importance of long-term systematic sampling of mosquitoes to build a predictive model as part of an early warning system. This is the first study in Texas, and overall the southwestern U.S., to use a long-term dataset to examine weather factors and variability to explain WNV vector abundance and WNV infection rates. We developed and validated models that can accurately predict WNV infection rates in response to weather phenomena. After one of the largest epidemics of WNV in 2012, which was centered in

Dallas County, TX, Harris County can integrate these models into a proactive system to initiate interventions and allocate resources for vector control and disease prevention before the appearance of human cases in order to prevent another devastating epidemic.

CHAPTER V

SUMMARY AND FUTURE RESEARCH

West Nile virus (WNV) is characterized by an enzootic cycle, involving avian hosts and mosquito vectors, as well as limited spillover into humans and horses. The temporal and spatial heterogeneity of WNV is complicated given the specific ecologies of the host and vector and generalizations from one region that may not necessarily be true for other regions. This dissertation provides information on how the ecologies related to the landscape, sociodemographics, and weather play a role in WNV transmission dynamics in Texas. Specifically, I used two datasets from Dallas and Harris Counties to identify significant factors that are related to the abundance, infection rate, or vector index of WNV.

We identified increased urbanization (based on an index composed of greater population density, lower normalized difference vegetation index, higher coverage of urban land types, and more impervious surfaces), lower elevation, and older populations as significant predictors of the presence of WNV in *Cx. quinquefasciatus* during the 2012 WNV epidemic in Dallas County. The relationships identified between the landscape, social demographics, and WNV infection help to identify regions of Dallas County with the highest risk for positive mosquito pools. While other studies have identified factors related to human WNV incidence, this is the only project that identified landscape and social demographic variables related to the vector of WNV during this epidemic.

We conducted a similar study for Harris County, TX, which includes the metropolitan city of Houston. We used Harris County Public Health Mosquito and Vector Control's surveillance dataset, which first started its surveillance for WNV in 2002. The best-fit model found that higher vector indexes are related to greater elevation, more impervious surfaces,

higher median income, and a predominantly Hispanic population. This model was then used to create a predictive map, which highlighted north and central Harris County as areas predicted to have high vector indexes. When we validated the predictive map with 2016-2017 trap data, we found that the map predicted with 38% accuracy for gravid traps and 26% accuracy for storm-sewer traps. The low validation measurements could be due to continuous vector control efforts throughout the county that were not taken into account for this study, the unclear nature of Hispanics as an ethnic group rather than race, changes in surveillance practice starting in 2016, and the spatial and temporal scale of the covariates.

Finally, this dissertation uncovered correlations between weather events and WNV vector abundance and infection rates in Harris County, TX. Using a time series analysis, our abundance model included measurements of variability for temperature, rainfall, and vegetation (normalized difference vegetation index and enhanced vegetation index). The abundance model follows Schmalhausen's law, which states that organisms are not only affected by the mean (average) of environmental variables, but also the variability and extremes of their environment. The WNV infection rates were associated with an 8-month lag of mean temperature, suggesting that ambient temperature during the winter might be key for WNV amplification during summer outbreaks. Model validation showed that the WNV infection rate model successfully predicted infection rates up to two months ahead (80% accuracy for 1 month ahead and 60% accuracy for 2 months ahead), but the abundance model performed poorly when predicting mosquito abundance. The low predictive accuracy of the abundance model reflects the effects of vector control, spatial and temporal scale of our time series, and trap location artifacts.

The landscape analyses for Dallas and Harris Counties resulted in a similar finding regarding urbanization and the built environment. The models for Dallas and Harris Counties

emphasized a positive association with WNV infection in *Cx. quinquefasciatus*, with an index of urbanization and impervious surfaces, respectively. However, even within the same state, these two major metropolitan counties in Texas emphasized different variables responsible for WNV infection in *Cx. quinquefasciatus*. For example, the models for both counties highlighted elevation as a significant variable, but they had opposite effects, with a negative association found in Dallas County and a positive association found in Harris County. In addition, there were differences in the demographic variables that were important in their respective counties such as median age in Dallas County and median income and percentage of Hispanics in Harris County.

While there were differences in the final models for each county, it should be noted that the availability of data drove our choices in the landscape/demographic covariates and the outcome variables entering the models. When developing the models for Dallas County, we found that race and ethnicity did not play a significant role in the models (i.e., did not explain a large proportion of variance), however, these factors were more apparent in the study for Harris County. Furthermore, the abundance data from Dallas County during the epidemic were considered unreliable given the lack of systematic sampling in 2012, therefore, we relied on the binary results for WNV testing as the outcome in our model. Since Harris County had an extensive database, relying on the binary result for each WNV test was not practical. Therefore, we summarized the data and calculated the vector index as a measurement of WNV-infected mosquitoes in Harris County. On the other hand, we were also able to analyze long-term patterns in the weather and WNV because of the extensive database from Harris County, but this was not possible with Dallas County since we only had data for a single year (2012). Regardless, the models for both counties revealed important relationships between WNV-infected mosquitoes and the environment that will be useful in constructing an early-warning system.

While this dissertation established the foundations for WNV patterns based on landscape, sociodemographic, and weather parameters for Texas, future work to build an early warning system is still warranted. For Dallas County, an analysis comparing the entomological risk map developed here with an epidemiological risk map utilizing human case data would inform us of human WNV risk and whether a high-risk area for WNV-positive mosquitoes equates to a high-risk area for WNV human cases. In addition, long-term surveillance data for Dallas County is needed to identify long-term patterns of WNV. Future directions for the dataset from Harris County, TX include investigations of population composition and phenology of various mosquito species known to vector diseases, effects of adulticide spraying on WNV vector abundance and infection, and outcomes based on weekly weather conditions compared to monthly conditions. Data gathered from these studies will add to the current knowledge of WNV and vectors of disease in Texas, which has largely remained underinvestigated compared to other states such as California and Illinois. Overall, we hope to bring these important variables to the forefront against WNV by creating a tool that will use weather, landscape, and sociodemographic parameters to predict WNV and track the intensity of the oncoming WNV season or give real-time forecasts of the risk of WNV infection in vectors. This kind of tool is currently unavailable for Texas, but these models can be used as parameters for the first early warning system in the state.

While vector data are important in understanding the biology and ecology of WNV transmission, we are also interested in data involving the hosts for WNV, including avian and human data, to fully understand the drivers of the disease cycle. For example, Harris County has an extensive avian database, which includes active mist-netting activities, passive surveillance of dead birds that citizens can report, and results of WNV testing in the bird samples. This avian

database could be used to further inform us of the enzootic cycle that precedes human infection with WNV. Finally, human case data will indicate if infected vectors and avian reservoirs align with WNV-positive human cases spatially and temporally, which have important implications for creating a proactive mosquito abatement program such as when and where to apply control methods using quantitative thresholds for vector infection with WNV. However, this process brings into question the ethics of initiating control measures based on quantitative thresholds for vector control. That is, a decision needs to be made regarding vector control when a situation arises where human cases exist, but the overall abundance/infection rate/vector index is low. The decision will change depending on the disease, but will ultimately rely on things such as the transmission cycle of the disease (humans as a reservoir vs. accidental host) and if the geographic location is in a high-risk area for the disease.

Early warning systems for WNV have been developed and parameterized for different regions of the U.S., but no such system exists for Texas even though the state is considered a hot spot for vector-borne diseases. The research presented here adds to the accumulating knowledge of the determinants of spatial and temporal differences of WNV transmission dynamics, with a focus on WNV in Texas and the major vector of WNV in the region *Cx. quinquefasciatus*. While we expect to see differences between our results and other studies, we find that even between two major cities in Texas, there are still large differences in the findings, exemplifying the complexities in the local interactions between the landscape, sociodemographics, weather, and the ecology of the vector. Furthermore, this dissertation emphasizes the importance of systematic sampling for WNV vectors in that we can draw conclusions about the spatial and temporal patterns and how they relate to various measures of WNV. Finally, we hope to use the significant factors identified in these studies to build early-warning systems specifically for Texas to prevent

another devastating event such as the major national WNV outbreak in 2012, which was centered in Dallas County, TX. With these results, we can predict when and where WNV outbreaks may happen within a county based on environmental parameters, which will ultimately save time, money, resources, and human lives.

REFERENCES

- Allan, B. F., R. B. Langerhans, W. A. Ryberg, W. J. Landesman, N. W. Griffin, R. S. Katz, B. J. Oberle, M. R. Schutzenhofer, K. N. Smyth, A. de St Maurice, L. Clark, K. R. Crooks, D. E. Hernandez, R. G. McLean, R. S. Ostfeld, and J. M. Chase. 2009.** Ecological correlates of risk and incidence of West Nile virus in the United States. *Oecologia* 158: 699-708.
- Anderson, J. F., T. G. Andreadis, A. J. Main, F. J. Ferrandino, and C. R. Vossbrinck. 2006.** West Nile Virus from Female and Male Mosquitoes (Diptera: Culicidae) in Subterranean, Ground, and Canopy Habitats in Connecticut. *J. Med. Entomol.* 43: 1010-1019.
- Andreadis, T. G. 2012.** The contribution of *Culex pipiens* complex mosquitoes to transmission and persistence of West Nile virus in North America. *J. Am. Mosq. Control Assoc.* 28: 137-151.
- Andreadis, T. G., J. F. Anderson, C. R. Vossbrinck, and A. J. Main. 2004.** Epidemiology of West Nile Virus in Connecticut: A Five-Year Analysis of Mosquito Data 1999–2003. *Vector Borne Zoonotic Dis.* 4: 360-378.
- Apperson, C. S., B. A. Harrison, T. R. Unnasch, H. K. Hassan, W. S. Irby, H. M. Savage, S. E. Aspen, D. W. Watson, L. M. Rueda, B. R. Engber, and R. S. Nasci. 2002.** Host-feeding habits of *Culex* and other mosquitoes (Diptera: Culicidae) in the Borough of Queens in New York City, with characters and techniques for identification of *Culex* mosquitoes. *J. Med. Entomol.* 39: 777-785.
- Arnold Jr, C. L., and C. J. Gibbons. 1996.** Impervious surface coverage: the emergence of a key environmental indicator. *J. Am. Plann. Assoc.* 62: 243-258.

- Artsob, H., D. J. Gubler, D. A. Enria, M. A. Morales, M. Pupo, M. L. Bunning, and J. P. Dudley. 2009.** West Nile Virus in the New World: trends in the spread and proliferation of West Nile Virus in the Western Hemisphere. *Zoonoses Public Health* 56: 357-369.
- Austgen, L. E., R. A. Bowen, M. L. Bunning, B. S. Davis, C. J. Mitchell, and G.-J. J. Chang. 2004.** Experimental infection of cats and dogs with West Nile virus. *Emerg. Infect. Dis.* 10: 82.
- Bakonyi, T., Z. Hubálek, I. Rudolf, and N. Nowotny. 2005.** Novel flavivirus or new lineage of West Nile virus, central Europe. *Emerg. Infect. Dis.* 11: 225.
- Baqar, S., C. G. Hayes, J. R. Murphy, and D. M. Watts. 1993.** Vertical transmission of West Nile virus by *Culex* and *Aedes* species mosquitoes. *Am. J. Trop. Med. Hyg.* 48: 757-762.
- Bates, D., M. Mächler, B. Bolker, and S. Walker. 2015.** Fitting Linear Mixed-Effects Models Using lme4. 67: 48.
- Beehler, J., J. Webb, and M. Mulla. 1993.** Spatial and circadian oviposition patterns in an urban population of *Culex quinquefasciatus*. *J. Am. Mosq. Control Assoc.* 9: 385-388.
- Bell, J. A., N. J. Mickelson, and J. A. Vaughan. 2005.** West Nile virus in host-seeking mosquitoes within a residential neighborhood in Grand Forks, North Dakota. *Vector Borne Zoonotic Dis.* 5: 373-382.
- Bentler, K. T., J. S. Hall, J. J. Root, K. Klenk, B. Schmit, B. F. Blackwell, P. C. Ramey, and L. Clark. 2007.** Serologic evidence of West Nile virus exposure in North American mesopredators. *Am. J. Trop. Med. Hyg.* 76: 173-179.
- Bernard, K. A., J. G. Maffei, S. A. Jones, E. B. Kauffman, G. Ebel, A. Dupuis 2nd, K. A. Ngo, D. C. Nicholas, D. M. Young, and P.-Y. Shi. 2001.** West Nile virus infection in birds and mosquitoes, New York State, 2000. *Emerg. Infect. Dis.* 7: 679.

- Bernkopf, H., S. Levine, and R. Nerson. 1953.** Isolation of West Nile Virus in Israel. *J. Infect. Dis.* 93: 207-218.
- Berthet, F., H. Zeller, M. Drouet, J. Rauzier, J. Digoutte, and V. Deubel. 1997.** Extensive nucleotide changes and deletions within the envelope glycoprotein gene of Euro-African West Nile viruses. *J. Gen. Virol.* 78: 2293-2297.
- Bian, L., L. Li, and G. Yan. 2006.** Combining Global and Local Estimates for Spatial Distribution of Mosquito Larval Habitats. *GISci. Remote Sens.* 43: 128-141.
- Biggerstaff, B. 2009.** PooledInfRate, Version 4.0: a Microsoft Office Excel Add-Into compute prevalence estimates from pooled samples. Centers for Disease Control and Prevention, Fort Collins.
- Bisanzio, D., M. Giacobini, L. Bertolotti, A. Mosca, L. Balbo, U. Kitron, and G. M. Vazquez-Prokopec. 2011.** Spatio-temporal patterns of distribution of West Nile virus vectors in eastern Piedmont Region, Italy. *Parasit. Vectors* 4: 230.
- Blaine, T. W., P. S. Grewal, A. Dawes, and D. Snider. 2010.** Profiling community gardeners. *J. Extension* 48: 1-12.
- Blitvich, B. J., L. I. Juarez, B. J. Tucker, W. A. Rowley, and K. B. Platt. 2009.** Antibodies to West Nile virus in raccoons and other wild peridomestic mammals in Iowa. *J. Wildl. Dis.* 45: 1163-1168.
- Bolling, B. G., J. H. Kennedy, and E. G. Zimmerman. 2005.** Seasonal dynamics of four potential West Nile vector species in north-central Texas. *J. Vector Ecol.* 30: 186-194.
- Bolling, B. G., C. M. Barker, C. G. Moore, W. J. Pape, and L. Eisen. 2009a.** Seasonal Patterns for Entomological Measures of Risk for Exposure to *Culex* Vectors and West

- Nile Virus in Relation to Human Disease Cases in Northeastern Colorado. J. Med. Entomol. 46: 1519-1531.
- Bolling, B. G., C. M. Barker, C. G. Moore, W. J. Pape, and L. Eisen. 2009b.** Modeling/GIS, risk assessment, economic impact: Seasonal patterns for entomological measures of risk for exposure to *Culex* vectors and West Nile virus in relation to human disease cases in Northeastern Colorado. J. Med. Entomol. 46: 1519.
- Bondre, V. P., R. S. Jadi, A. C. Mishra, P. N. Yergolkar, and V. A. Arankalle. 2007.** West Nile virus isolates from India: evidence for a distinct genetic lineage. J. Gen. Virol. 88: 875-884.
- Bowden, S. E., K. Magori, and J. M. Drake. 2011.** Regional differences in the association between land cover and West Nile virus disease incidence in humans in the United States. Am. J. Trop. Med. Hyg. 84: 234-238.
- Bozdogan, H. 1987.** Model selection and Akaike's information criterion (AIC): The general theory and its analytical extensions. Psychometrika 52: 345-370.
- Brault, A. C. 2009.** Changing patterns of West Nile virus transmission: altered vector competence and host susceptibility. Vet. Res. 40: 43.
- Brown, E. 2012.** California mosquito-borne virus surveillance & response plan. California: California Department of Public Health Mosquito & Vector Control Association of California University of California.
- Brown, H., M. Duik-Wasser, T. Andreadis, and D. Fish. 2008a.** Remotely-sensed vegetation indices identify mosquito clusters of West Nile virus vectors in an urban landscape in the northeastern United States. Vector Borne Zoonotic Dis. 8: 197-206.

- Brown, H. E., J. E. Childs, M. A. Diuk-Wasser, and D. Fish. 2008b.** Ecological factors associated with West Nile virus transmission, northeastern United States. *Emerg. Infect. Dis.* 14: 1539-1545.
- Brown, H. E., M. Paladini, R. A. Cook, D. Kline, D. Barnard, and D. Fish. 2008c.** Effectiveness of mosquito traps in measuring species abundance and composition. *J. Med. Entomol.* 45: 517-521.
- Brownstein, J. S., H. Rosen, D. Purdy, J. R. Miller, M. Merlino, F. Mostashari, and D. Fish. 2002.** Spatial Assessment of West Nile Virus: Rapid Risk Assessment of an Introduced Vector-Borne Zoonosis. *Vector Borne Zoonotic Dis.* 2: 157-164.
- Brunsdon, C., and L. Comber. 2015.** An introduction to R for spatial analysis and mapping, Sage Publications LTD., London.
- Burakoff, A., J. Lehman, M. Fischer, J. E. Staples, and N. P. Lindsey. 2018.** West Nile Virus and Other Nationally Notifiable Arboviral Diseases—United States, 2016. *MMWR Morb. Mortal. Wkly. Rep.* 67: 13.
- Busetto, L., and L. Ranghetti. 2016.** MODISstp: An R package for automatic preprocessing of MODIS Land Products time series. *Computers & Geosciences* 97: 40-48.
- Bustamante, D. M., and C. C. Lord. 2010.** Sources of error in the estimation of mosquito infection rates used to assess risk of arbovirus transmission. *Am. J. Trop. Med. Hyg.* 82: 1172-1184.
- Calhoun, L. M., M. Avery, L. Jones, K. Gunarto, R. King, J. Roberts, and T. R. Burkot. 2007.** Combined sewage overflows (CSO) are major urban breeding sites for *Culex quinquefasciatus* in Atlanta, Georgia. *Am. J. Trop. Med. Hyg.* 77: 478-484.

- Cazelles, B., M. Chavez, G. C. De Magny, J.-F. Guégan, and S. Hales. 2007.** Time-dependent spectral analysis of epidemiological time-series with wavelets. *J. R. Soc. Interface* 4: 625-636.
- CDC. 2002a.** Intrauterine West Nile virus infection--New York, 2002. *MMWR Morb. Mortal. Wkly. Rep.* 51: 1135.
- CDC. 2002b.** Possible West Nile virus transmission to an infant through breast-feeding--Michigan, 2002. *MMWR Morb. Mortal. Wkly. Rep.* 51: 877.
- CDC. 2002c.** Laboratory-acquired West Nile virus infections--United States, 2002. *MMWR Morb. Mortal. Wkly. Rep.* 51: 1133.
- CDC. 2003.** West Nile virus infection among turkey breeder farm workers--Wisconsin, 2002. *MMWR Morb. Mortal. Wkly. Rep.* 52: 1017.
- CDC. 2013.** West Nile Virus in the United States: Guidelines for Surveillance.
- CDC. 2014.** West Nile Virus: Final Annual Maps and Data for 1999-2014.
- CDC. 2016a.** Final Cumulative Maps & Data for 1999-2016.
- CDC. 2016b.** Final Annual Maps & Data for 1999-2016.
- CDC. 2017.** West Nile Virus & Dead Birds.
- Chancey, C., A. Grinev, E. Volkova, and M. Rios. 2015.** The global ecology and epidemiology of West Nile Virus. *Biomed. Res. Int.* 2015: 376230.
- Chase, J. M., and T. M. Knight. 2003.** Drought-induced mosquito outbreaks in wetlands. *Ecol. Lett.* 6: 1017-1024.
- Chaves, L., and U. Kitron. 2011.** Weather variability impacts on oviposition dynamics of the southern house mosquito at intermediate time scales. *Bull. Entomol. Res.* 101: 633-641.

- Chaves, L. F. 2016.** Globally invasive, withdrawing at home: *Aedes albopictus* and *Aedes japonicus* facing the rise of *Aedes flavopictus*. Int. J. Biometeorol. 60: 1727-1738.
- Chaves, L. F. 2017.** Climate change and the biology of insect vectors of human pathogens, John Wiley & Sons, Ltd.
- Chaves, L. F., and M. Pascual. 2006.** Climate cycles and forecasts of cutaneous leishmaniasis, a nonstationary vector-borne disease. PLoS Med. 3: e295.
- Chaves, L. F., and M. Pascual. 2007.** Comparing models for early warning systems of neglected tropical diseases. PLoS Negl. Trop. Dis. 1: e33.
- Chaves, L. F., and C. J. Koenraadt. 2010.** Climate change and highland malaria: fresh air for a hot debate. Q. Rev. Biol. 85: 27-55.
- Chaves, L. F., J.-Y. Jian, and K. Moji. 2018.** Overwintering in the Bamboo Mosquito *Tripteroides bambusa* (Diptera: Culicidae) During a Warm, But Unpredictably Changing, Winter. Environ. Entomol. 47: 148-158.
- Chaves, L. F., C. L. Keogh, G. M. Vazquez-Prokopec, and U. D. Kitron. 2009.** Combined Sewage Overflow Enhances Oviposition of *Culex quinquefasciatus* (Diptera: Culicidae) in Urban Areas. J. Med. Entomol. 46: 220-226.
- Chaves, L. F., A. C. Morrison, U. D. Kitron, and T. W. Scott. 2012.** Nonlinear impacts of climatic variability on the density-dependent regulation of an insect vector of disease. Global Change Biol. 18: 457-468.
- Chaves, L. F., G. L. Hamer, E. D. Walker, W. M. Brown, M. O. Ruiz, and U. D. Kitron. 2011.** Climatic variability and landscape heterogeneity impact urban mosquito diversity and vector abundance and infection. Ecosphere 2: 1-21.

- Chaves, L. F., Y. Higa, S. H. Lee, J. Y. Jeong, S. T. Heo, M. Kim, N. Minakawa, and K. H. Lee. 2013.** Environmental forcing shapes regional house mosquito synchrony in a warming temperate island. *Environ. Entomol.* 42: 605-613.
- Chuang, T.-W., M. B. Hildreth, D. L. Vanroekel, and M. C. Wimberly. 2011.** Weather and Land Cover Influences on Mosquito Populations in Sioux Falls, South Dakota. *J. Med. Entomol.* 48: 669-679.
- Chuang, T. W., and M. C. Wimberly. 2012.** Remote sensing of climatic anomalies and West Nile virus incidence in the northern Great Plains of the United States. *PloS one* 7: e46882.
- Chuang, T. W., L. F. Chaves, and P. J. Chen. 2017.** Effects of local and regional climatic fluctuations on dengue outbreaks in southern Taiwan. *PloS one* 12: e0178698.
- Chuang, T. W., C. W. Hockett, L. Kightlinger, and M. C. Wimberly. 2012.** Landscape-level spatial patterns of West Nile virus risk in the northern Great Plains. *Am. J. Trop. Med. Hyg.* 86: 724-731.
- Chung, W. M., C. M. Buseman, S. N. Joyner, S. M. Hughes, T. B. Fomby, J. P. Luby, and R. W. Haley. 2013.** The 2012 West Nile encephalitis epidemic in Dallas, Texas. *J. A. Med. Assoc.* 310: 297-307.
- Clements, A. 1992.** *The Biology of Mosquitoes, Vol I: Development, Nutrition and Reproduction*, Chapman Hall, Florida.
- Colborn, J. M., K. A. Smith, J. Townsend, D. Damian, R. S. Nasci, and J. P. Mutebi. 2013.** West Nile virus outbreak in Phoenix, Arizona--2010: entomological observations and epidemiological correlations. *J. Am. Mosq. Control Assoc.* 29: 123-132.
- Colorado Department of Public Health and Environment. 2018.** West Nile virus.

- Colton, L., and R. S. Nasci. 2006.** Quantification of West Nile virus in the saliva of *Culex* species collected from the southern United States. *J. Am. Mosq. Control Assoc.* 22: 57-63.
- Condotta, S. A., F. F. Hunter, and M. J. Bidochka. 2004.** West Nile virus infection rates in pooled and individual mosquito samples. *Vector Borne Zoonotic Dis.* 4: 198-203.
- Connor, S., M. Thomson, and D. Molyneux. 1999.** Forecasting and prevention of epidemic malaria: new perspectives on an old problem. *Parassitologia* 41: 439-448.
- Constantine, D. G. 1970.** Bats in relation to the health, welfare, and economy of man, Academic Press Incorporated.
- Cooke, W. H., 3rd, K. Grala, and R. C. Wallis. 2006.** Avian GIS models signal human risk for West Nile virus in Mississippi. *Int. J. Health Geogr.* 5: 36.
- Coppedge, B. R., D. M. Engle, R. E. Masters, and M. S. Gregory. 2001.** Avian response to landscape change in fragmented southern Great Plains grasslands. *Ecol. Appl.* 11: 47-59.
- Craig, M. H., R. Snow, and D. le Sueur. 1999.** A climate-based distribution model of malaria transmission in sub-Saharan Africa. *Parasitol. Today* 15: 105-111.
- Curtis, A., X. Ye, E. Heob, J. Targhetta, V. Salvato, M. Reyna, R. Bueno, and L. Holmes. 2014.** A comparison of three approaches to identify West Nile Virus mosquito space-time hotspots in the Houston Vicinity for the period 2002–2011. *Appl. Geogr.* 51: 58-64.
- Davis, A., M. Bunning, P. Gordy, N. Panella, B. Blitvich, and R. Bowen. 2005.** Experimental and natural infection of North American bats with West Nile virus. *Am. J. Trop. Med. Hyg.* 73: 467-469.

- Dawson, J. R., W. B. Stone, G. D. Ebel, D. S. Young, D. S. Galinski, J. P. Pensabene, M. A. Franke, M. Eidson, and L. D. Kramer. 2007.** Crow deaths caused by West Nile virus during winter. *Emerg. Infect. Dis.* 13: 1912.
- Degaetano, A. T. 2005.** Meteorological effects on adult mosquito (*Culex*) populations in metropolitan New Jersey. *Int. J. Biometeorol.* 49: 345-353.
- DeGroote, J. P., and R. Sugumaran. 2012.** National and regional associations between human West Nile virus incidence and demographic, landscape, and land use conditions in the coterminous United States. *Vector Borne Zoonotic Dis.* 12: 657-665.
- Degroote, J. P., R. Sugumaran, and M. Ecker. 2014.** Landscape, demographic and climatic associations with human West Nile virus occurrence regionally in 2012 in the United States of America. *Geospat. Health* 9: 153-168.
- DeGroote, J. P., R. Sugumaran, S. M. Brend, B. J. Tucker, and L. C. Bartholomay. 2008.** Landscape, demographic, entomological, and climatic associations with human disease incidence of West Nile virus in the state of Iowa, USA. *Int. J. Health Geogr.* 7: 19.
- Deichmeister, J. M., and A. Telang. 2010.** Abundance of West Nile virus mosquito vectors in relation to climate and landscape variables. *J. Vector Ecol.* 36: 75-85.
- Denlinger, D. L., and P. A. Armbruster. 2014.** Mosquito diapause. *Annu. Rev. Entomol.* 59: 73-93.
- Dennett, J., and M. Debboun. 2017.** Case Study: Surveillance and Control Operations in Harris County, Texas. *Pest Control Technology*.
- Dennett, J. A., T. Wuithiranyagool, M. Reyna-Nava, A. Bala, R. B. Tesh, R. E. Parsons, and R. Bueno. 2007a.** Description And Use Of The Harris County Gravid trap For West Nile Virus Surveillance 2003–061. *J. Am. Mosq. Control Assoc.* 23: 359-362.

- Dennett, J. A., A. Bala, T. Wuithiranyagool, Y. Randle, C. B. Sargent, H. Guzman, M. Siirin, H. K. Hassan, M. Reyna-Nava, T. R. Unnasch, R. B. Tesh, R. E. Parsons, and R. Bueno. 2007b.** Associations between Two Mosquito Populations and West Nile Virus in Harris County, Texas, 2003–061. *J. Am. Mosq. Control Assoc.* 23: 264-275.
- Desalvo, D., P. Roy-Chaudhury, R. Peddi, T. Merchen, K. Konijetti, M. Gupta, R. Boardman, C. Rogers, J. Buell, and M. Hanaway. 2004.** West Nile virus encephalitis in organ transplant recipients: another high-risk group for meningoencephalitis and death. *Transplantation* 77: 466-469.
- Didan, K. 2015.** MYD13A3 MODIS/Aqua Vegetation Indices Monthly L3 Global 1km SIN Grid V006 *In* N. E. L. DAAC. [ed.].
- DiMenna, M. A., R. Bueno, R. R. Parmenter, D. E. Norris, J. M. Sheyka, J. L. Molina, E. M. LaBeau, E. S. Hatton, and G. E. Glass. 2006.** Emergence of West Nile Virus in Mosquito (Diptera: Culicidae) Communities of the New Mexico Rio Grande Valley. *J. Med. Entomol.* 43: 594-599.
- Diniz, D. F. A., C. M. R. de Albuquerque, L. O. Oliva, M. A. V. de Melo-Santos, and C. F. J. Ayres. 2017.** Diapause and quiescence: dormancy mechanisms that contribute to the geographical expansion of mosquitoes and their evolutionary success. *Parasit. Vectors* 10: 310.
- Diuk-Wasser, M. A., H. E. Brown, T. G. Andreadis, and D. Fish. 2006.** Modeling the Spatial Distribution of Mosquito Vectors for West Nile Virus in Connecticut, USA. *Vector Borne Zoonotic Dis.* 6: 283-295.

- Dohm, D. J., and M. J. Turell. 2001.** Effect of incubation at overwintering temperatures on the replication of West Nile virus in New York *Culex pipiens* (Diptera: Culicidae). J. Med. Entomol. 38: 462-464.
- Dohm, D. J., M. R. Sardelis, and M. J. Turell. 2002a.** Experimental Vertical Transmission of West Nile Virus by *Culex pipiens* (Diptera: Culicidae). J. Med. Entomol. 39: 640-644.
- Dohm, D. J., M. L. O'Guinn, and M. J. Turell. 2002b.** Effect of Environmental Temperature on the Ability of *Culex pipiens* (Diptera: Culicidae) to Transmit West Nile Virus. J. Med. Entomol. 39: 221-225.
- Dowling, Z., S. L. Ladeau, P. Armbruster, D. Biehler, and P. T. Leisnham. 2013.** Socioeconomic Status Affects Mosquito (Diptera: Culicidae) Larval Habitat Type Availability and Infestation Level. J. Med. Entomol. 50: 764-772.
- Du, Y., and J. G. Millar. 1999.** Oviposition responses of gravid *Culex quinquefasciatus* and *Culex tarsalis* to bulrush (*Schoenoplectus acutus*) infusions. J. Am. Mosq. Control Assoc. 15: 500-509.
- Ebel, G. D., I. Rochlin, J. Longacker, and L. D. Kramer. 2005.** *Culex restuans* (Diptera: Culicidae) Relative Abundance and Vector Competence for West Nile Virus. J. Med. Entomol. 42: 838-843.
- Edgell, M. C. 1984.** Trans-hemispheric movements of Holarctic Anatidae: the Eurasian wigeon (*Anas penelope* L.) in North America. J. Biogeogr.: 27-39.
- Eisen, L., B. G. Bolling, C. D. Blair, B. J. Beaty, and C. G. Moore. 2008.** Mosquito Species Richness, Composition, and Abundance along Habitat-Climate-Elevation Gradients in the Northern Colorado Front Range. J. Med. Entomol. 45: 800-811.

Eisen, L., C. M. Barker, C. G. Moore, W. J. Pape, A. M. Winters, and N. Cheronis. 2010.

Irrigated agriculture is an important risk factor for West Nile virus disease in the hyperendemic Larimer-Boulder-Weld area of north central Colorado. *J. Med. Entomol.* 47: 939-951.

Eisen, R. J., and L. Eisen. 2008. Spatial Modeling of Human Risk of Exposure to Vector-Borne

Pathogens Based on Epidemiological Versus Arthropod Vector Data. *J. Med. Entomol.* 45: 181-192.

Eldridge, B. F. 1968. The effect of temperature and photoperiod on blood-feeding and ovarian

development in mosquitoes of the *Culex pipiens* complex. *Am. J. Trop. Med. Hyg.* 17: 133-140.

Epstein, P. R. 2001. West Nile virus and the climate. *J. Urban Health* 78: 367-371.

Epstein, P. R., and C. Defilippo. 2001. West Nile virus and drought. *Glob. Chang. Hum. Health*

2: 105-107.

Farajollahi, A., D. M. Fonseca, L. D. Kramer, and A. Marm Kilpatrick. 2011. "Bird biting"

mosquitoes and human disease: a review of the role of *Culex pipiens* complex mosquitoes in epidemiology. *Infect. Genet. Evol.* 11: 1577-1585.

Faraway, J. J. 2016. Linear models with R, Chapman and Hall/CRC.

Farrington, C. 1992. Estimating prevalence by group testing using generalized linear models.

Stat. Med. 11: 1591-1597.

Forchhammer, M. C., E. Post, and N. C. Stenseth. 1998. Breeding phenology and climate....

Nature 391: 29.

- Fredregill, C. L., G. C. Motl, J. A. Dennett, K. L. Flatt, and R. Bueno, Jr. 2011.** Thermal fog efficacy tests against mosquitoes in storm drains in Harris County, Texas 2008-09. J. Am. Mosq. Control Assoc. 27: 61-68.
- Gardner, A. M., T. K. Anderson, G. L. Hamer, D. E. Johnson, K. E. Varela, E. D. Walker, and M. O. Ruiz. 2013.** Terrestrial vegetation and aquatic chemistry influence larval mosquito abundance in catch basins, Chicago, USA. Parasit. and Vectors 6: 9.
- Geographic Information Science and Spatial Epidemiology. 2015.** MIR Model.
- Gibbs, S. E. J., M. C. Wimberly, M. Madden, J. Masour, M. J. Yabsley, and D. E. Stallknecht. 2006.** Factors Affecting the Geographic Distribution of West Nile Virus in Georgia, USA: 2002–2004. Vector Borne Zoonotic Dis. 6: 73-82.
- Gleiser, R. M., and L. P. Zalazar. 2010.** Distribution of mosquitoes in relation to urban landscape characteristics. Bull. Entomol. Res. 100: 153-158.
- Goddard, L. B., A. E. Roth, W. K. Reisen, and T. W. Scott. 2002.** Vector competence of California mosquitoes for West Nile virus. Emerging infectious diseases 8: 1385.
- Goddard, L. B., A. E. Roth, W. K. Reisen, and T. W. Scott. 2003.** Vertical Transmission of West Nile Virus by Three California *Culex* (Diptera: Culicidae) Species. J. Med. Entomol. 40: 743-746.
- Gould, L. H., and E. Fikrig. 2004.** West Nile virus: a growing concern? J. Clin. Invest. 113: 1102-1107.
- Green, L. W. 1970.** Manual for scoring socioeconomic status for research on health behavior. Public Health Rep. 85: 815.

- Gu, W., T. R. Unnasch, C. R. Katholi, R. Lampman, and R. J. Novak. 2008.** Fundamental issues in mosquito surveillance for arboviral transmission. *T. Roy. Soc. Trop. Med. H.* 102: 817-822.
- Gujral, I. B., E. C. Zielinski-Gutierrez, A. LeBailly, and R. Nasci. 2007.** Behavioral risks for West Nile virus disease, northern Colorado, 2003. *Emerg. Infect. Dis.* 13: 419.
- Hahn, M. B., A. J. Monaghan, M. H. Hayden, R. J. Eisen, M. J. Delorey, N. P. Lindsey, R. S. Nasci, and M. Fischer. 2015.** Meteorological conditions associated with increased incidence of West Nile virus disease in the United States, 2004-2012. *Am. J. Trop. Med. Hyg.* 92: 1013-1022.
- Hamer, G. L., P. H. Kelly, D. A. Focks, T. L. Goldberg, and E. D. Walker. 2011.** Evaluation of a novel emergence trap to study *Culex* mosquitoes in urban catch basins. *J. Am. Mosq. Control Assoc.* 27: 142-147.
- Hamer, G. L., U. D. Kitron, J. D. Brawn, S. R. Loss, M. O. Ruiz, T. L. Goldberg, and E. D. Walker. 2008a.** *Culex pipiens* (Diptera: Culicidae): a bridge vector of West Nile virus to humans. *J. Med. Entomol.* 45: 125-128.
- Hamer, G. L., U. D. Kitron, T. L. Goldberg, J. D. Brawn, S. R. Loss, M. O. Ruiz, D. B. Hayes, and E. D. Walker. 2009.** Host selection by *Culex pipiens* mosquitoes and West Nile virus amplification. *Am. J. Trop. Med. Hyg.* 80: 268-278.
- Hamer, G. L., E. D. Walker, J. D. Brawn, S. R. Loss, M. O. Ruiz, T. L. Goldberg, A. M. Schotthoefer, W. M. Brown, E. Wheeler, and U. D. Kitron. 2008b.** Rapid amplification of West Nile virus: the role of hatch-year birds. *Vector Borne Zoonotic Dis.* 8: 57-67.

- Harrigan, R. J., H. A. Thomassen, W. Buermann, R. F. Cummings, M. E. Kahn, and T. B. Smith. 2010.** Economic conditions predict prevalence of West Nile virus. *PloS one* 5: e15437.
- Harrington, L. C., and R. L. Poulson. 2008.** Considerations for accurate identification of adult *Culex restuans* (Diptera: Culicidae) in field studies. *J. Med. Entomol.* 45: 1-8.
- Hayes, C., and T. Monath. 1989.** West Nile fever, pp. 59–88. CRC Boca Raton, FL.
- Hayes, J. 1975.** Seasonal Changes in Population Structure of *Culex pipiens quinquefasciatus* Say (Diptera: Culicidae): Study of an Isolated Population. *J. Med. Entomol.* 12: 167-178.
- Hayes, J., and B. P. Hsi. 1975.** Interrelationships between selected meteorologic phenomena and immature stages of *Culex pipiens quinquefasciatus* Say: study of an isolated population. *J. Med. Entomol.* 12: 299-308.
- Hayes, J., and T. D. Downs. 1980.** Seasonal changes in an isolated population of *Culex pipiens quinquefasciatus* (Diptera: Culicidae): A time series analysis. *J. Med. Entomol.* 17: 63-69.
- Heft, D. E., and W. E. Walton. 2008.** Effects of the El Niño-Southern Oscillation (ENSO) cycle on mosquito populations in southern California. *J. Vector Ecol.* 33: 17-29.
- Hinton, M. G., W. K. Reisen, S. S. Wheeler, and A. K. Townsend. 2015.** West Nile Virus Activity in a Winter Roost of American Crows (*Corvus brachyrhynchos*): Is Bird-To-Bird Transmission Important in Persistence and Amplification? *J. Med. Entomol.* 52: 683-692.
- Hlavac, M. 2015.** Stargazer: Well-formatted regression and summary statistics tables (R package version 5.2).

Hongoh, V., L. Berrang-Ford, N. H. Ogden, R. Lindsay, M. E. Scott, and H. Artsob. 2009.

A review of environmental determinants and risk factors for avian-associated mosquito arboviruses in Canada. *Biodiversity* 10: 83-91.

Hoshi, T., Y. Higa, and L. F. Chaves. 2014. *Uranotaenia novobscura ryukyuana* (Diptera:

Culicidae) population dynamics are denso-dependent and autonomous from weather fluctuations. *Ann. Entomol. Soc. Am.* 107: 136-142.

Hribar, L. J., J. M. Smith, J. J. Vlach, and T. N. Verna. 2001. Survey of container-breeding

mosquitoes from the Florida Keys, Monroe County, Florida. *J. Am. Mosq. Control Assoc.* 17: 245-248.

Huang, B., P. W. Thorne, V. F. Banzon, T. Boyer, G. Chepurin, J. H. Lawrimore, M. J.

Menne, T. M. Smith, R. S. Vose, and H.-M. Zhang. 2017. Extended reconstructed sea surface temperature, version 5 (ERSSTv5): upgrades, validations, and intercomparisons. *J. Clim.* 30: 8179-8205.

Hunt, G. J., and C. S. Hacker. 1984. Computer-generated maps as an aid to mosquito control

(Diptera: Culicidae). *J. Med. Entomol.* 21: 489-500.

Hurtado, L. A., L. Cáceres, L. F. Chaves, and J. E. Calzada. 2014. When climate change

couples social neglect: malaria dynamics in Panamá. *Emerg. Microbes Infect.* 3: e28.

Hurtado, L. A., J. E. Calzada, C. A. Rigg, M. Castillo, and L. F. Chaves. 2018. Climatic

fluctuations and malaria transmission dynamics, prior to elimination, in Guna Yala, República de Panamá. *Malar. J.* 17: 85.

Hwang, J., H.-S. Ryu, H. Kim, and S.-A. Lee. 2015. The first reported case of West Nile

encephalitis in Korea. *J. Korean Med. Sci.* 30: 343-345.

- Iwamoto, M., D. B. Jernigan, A. Guasch, M. J. Trepka, C. G. Blackmore, W. C. Hellinger, S. M. Pham, S. Zaki, R. S. Lanciotti, and S. E. Lance-Parker. 2003.** Transmission of West Nile virus from an organ donor to four transplant recipients. *New Engl. J. Med.* 348: 2196-2203.
- Jones, R. C., K. N. Weaver, S. Smith, C. Blanco, C. Flores, K. Gibbs, D. Markowski, and J. P. Mutebi. 2011.** Use of the vector index and geographic information system to prospectively inform West Nile virus interventions. *J. Am. Mosq. Control Assoc.* 27: 315-319.
- Karabatsos, N. 1985.** International catalogue of arboviruses, including certain other viruses of vertebrates.
- Karki, S., G. L. Hamer, T. K. Anderson, T. L. Goldberg, U. D. Kitron, B. L. Krebs, E. D. Walker, and M. O. Ruiz. 2016.** Effect of Trapping Methods, Weather, and Landscape on Estimates of the *Culex* Vector Mosquito Abundance. *Environ. Health Insights* 10: 93-103.
- Kilpatrick, A. M., M. A. Meola, R. M. Moudy, and L. D. Kramer. 2008.** Temperature, viral genetics, and the transmission of West Nile virus by *Culex pipiens* mosquitoes. *PLoS Pathog.* 4: e1000092.
- Kilpatrick, A. M., L. D. Kramer, M. J. Jones, P. P. Marra, and P. Daszak. 2006.** West Nile virus epidemics in North America are driven by shifts in mosquito feeding behavior. *PLoS Biol.* 4: e82.
- Kilpatrick, A. M., L. D. Kramer, S. R. Campbell, E. O. Alleyne, A. P. Dobson, and P. Daszak. 2005a.** West Nile virus risk assessment and the bridge vector paradigm. *Emerg. Infect. Dis.* 11: 425-429.

- Kilpatrick, A. M., L. D. Kramer, S. R. Campbell, E. O. Alleyne, A. P. Dobson, and P. Daszak. 2005b.** West Nile virus risk assessment and the bridge vector paradigm. *Emerging infectious diseases* 11: 425-429.
- Klenk, K., and N. Komar. 2003.** Poor replication of West Nile virus (New York 1999 strain) in three reptilian and one amphibian species. *Am. J. Trop. Med. Hyg.* 69: 260-262.
- KNMI.** KNMI Climate Explorer.
- Koenraadt, C. J. M., and L. Harrington. 2008.** Flushing effect of rain on container-inhabiting mosquitoes *Aedes aegypti* and *Culex pipiens* (Diptera: Culicidae). *J. Med. Entomol.* 45: 28-35.
- Komar, N. 2000.** West Nile viral encephalitis. *Rev. Sci. Tech. OIE* 19: 166-171.
- Komar, N., N. A. Panella, A. J. Golnar, and G. L. Hamer. 2018.** Forage Ratio Analysis of the Southern House Mosquito in College Station, Texas. *Vector Borne Zoonotic Dis.*
- Komar, N., N. A. Panella, J. E. Burns, S. W. Dusza, T. M. Mascarenhas, and T. O. Talbot. 2001.** Serologic evidence for West Nile virus infection in birds in the New York City vicinity during an outbreak in 1999. *Emerg. Infect. Dis.* 7: 621.
- Komar, N., S. Langevin, S. Hinten, N. M. Nemeth, E. Edwards, D. Hettler, B. Davis, R. Bowen, and M. Bunning. 2003.** Experimental Infection of North American Birds with the New York 1999 Strain of West Nile Virus. *Emerging infectious diseases* 9: 311-322.
- Kostyukov, M., A. Alekseev, V. Bul'chev, and Z. Gordeeva. 1986.** Experimentally proven infection of *Culex pipiens* L. mosquitoes with West Nile fever virus via the Lake Pallas (*Rana ridibunda*) frog and its transmission via bites. *Med. Parazitol. (Mosk)* 6: 76-78.

- Kostyukov, M., Z. Gordeeva, V. Bulychev, N. Nemova, and O. Daniyarov. 1985.** The lake frog (*Rana ridibunda*)--one of the food hosts of blood-sucking mosquitoes in Tadzhikistan--a reservoir of the West Nile fever virus. *Med. Parazitol. (Mosk)*: 49-50.
- Kramer, L. D., L. M. Styer, and G. D. Ebel. 2008.** A global perspective on the epidemiology of West Nile virus. *Annu. Rev. Entomol.* 53: 61-81.
- Kronenwetter-Koepel, T. A., J. K. Meece, C. A. Miller, and K. D. Reed. 2005.** Surveillance of above-and below-ground mosquito breeding habitats in a rural midwestern community: baseline data for larvicidal control measures against West Nile Virus vectors. *Clinical medicine & research* 3: 3-12.
- Kuhn, K., D. Campbell-Lendrum, A. Haines, J. Cox, C. Corvalán, and M. Anker. 2005.** Using climate to predict infectious disease epidemics. Geneva: WHO.
- Kuhn, M., and K. Johnson. 2013.** Applied predictive modeling, vol. 26, Springer.
- Kumar, D., G. R. Prasad, J. Zaltzman, G. A. Levy, and A. Humar. 2004.** Community-acquired West Nile virus infection in solid-organ transplant recipients. *Transplantation* 77: 399-402.
- Kwan, J. L., B. K. Park, T. E. Carpenter, V. Ngo, R. Civen, and W. K. Reisen. 2012.** Comparison of enzootic risk measures for predicting West Nile disease, Los Angeles, California, USA, 2004-2010. *Emerg. Infect. Dis.* 18: 1298-1306.
- LaBeaud, A. D., A.-M. Gorman, J. Koonce, C. Kippes, J. McLeod, J. Lynch, T. Gallagher, C. H. King, and A. M. Mandalakas. 2008.** Rapid GIS-based profiling of West Nile virus transmission: defining environmental factors associated with an urban-suburban outbreak in Northeast Ohio, USA. *Geospat. Health* 2: 215.

- Lacour, G., L. Chanaud, G. L'Ambert, and T. Hance. 2015.** Seasonal synchronization of diapause phases in *Aedes albopictus* (Diptera: Culicidae). PloS one 10: e0145311.
- Lanciotti, R., J. Roehrig, V. Deubel, J. Smith, M. Parker, K. Steele, B. Crise, K. Volpe, M. Crabtree, and J. Scherret. 1999.** Origin of the West Nile virus responsible for an outbreak of encephalitis in the northeastern United States. Science 286: 2333-2337.
- Lanciotti, R. S., G. D. Ebel, V. Deubel, A. J. Kerst, S. Murri, R. Meyer, M. Bowen, N. McKinney, W. E. Morrill, M. B. Crabtree, L. D. Kramer, and J. T. Roehrig. 2002.** Complete Genome Sequences and Phylogenetic Analysis of West Nile Virus Strains Isolated from the United States, Europe, and the Middle East. Virology 298: 96-105.
- Landesman, W. J., B. F. Allan, R. B. Langerhans, T. M. Knight, and J. M. Chase. 2007.** Inter-annual associations between precipitation and human incidence of West Nile virus in the United States. Vector Borne Zoonotic Dis. 7: 337-343.
- Lee, J.-H., M. Stahl, S. Sawlis, and S. Suzuki. 2009.** A Potential Risk Assessment of a Dengue Outbreak in North Central Texas, USA (Part 2 of 2). J. Environ. Health 71: 36-39.
- Levins, R. 1968.** Evolution in changing environments: some theoretical explorations, Princeton University Press.
- Levins, R., T. Awerbuch, U. Brinkmann, I. Eckardt, P. Epstein, N. Makhoul, C. A. de Possas, C. Puccia, A. Spielman, and M. E. Wilson. 1994.** The emergence of new diseases. Am. Sci. 82: 52-60.
- Lewontin, R., and R. Levins. 2000.** Schmalhausen's law. Capitalism Nat. Social. 11: 103-108.
- Li, X.-L., S.-H. Fu, W.-B. Liu, H.-Y. Wang, Z. Lu, S.-X. Tong, Z.-X. Li, R. S. Nasci, O. Kosoy, and Y. Cui. 2013.** West Nile Virus infection in Xinjiang, China. Vector Borne Zoonotic Dis. 13: 131-133.

- Li, Z., and M. Kafatos. 2000.** Interannual variability of vegetation in the United States and its relation to El Nino/Southern Oscillation. *Remote Sens. Environ.* 71: 239-247.
- Lillesand, T., R. W. Kiefer, and J. Chipman. 2014.** Remote sensing and image interpretation, John Wiley & Sons.
- Lillibridge, K. M., R. Parsons, Y. Randle, A. P. T. DA ROSA, H. Guzman, M. Siirin, T. Wuithiranyagool, C. Hailey, S. Higgs, and A. A. Bala. 2004.** The 2002 introduction of West Nile virus into Harris County, Texas, an area historically endemic for St. Louis encephalitis. *Am. J. Trop. Med. Hyg.* 70: 676-681.
- Lowe, R., T. C. Bailey, D. B. Stephenson, T. E. Jupp, R. J. Graham, C. Barcellos, and M. S. Carvalho. 2013.** The development of an early warning system for climate-sensitive disease risk with a focus on dengue epidemics in Southeast Brazil. *Stat. Med.* 32: 864-883.
- Machain-Williams, C., S. E. Padilla-Paz, M. Weber, R. Cetina-Trejo, J. A. Juarez-Ordaz, M. A. Loroño-Pino, A. Ulloa, C. Wang, J. Garcia-Rejon, Blitvich, and B. J. 2013.** Antibodies to West Nile virus in wild and farmed crocodiles in southeastern Mexico. *J. Wildl. Dis.* 49: 690-693.
- Mackenzie, J., A. Barrett, and V. Deubel. 2002.** The Japanese encephalitis serological group of flaviviruses: a brief introduction to the group, pp. 1-10, Japanese encephalitis and West Nile viruses. Springer.
- Manore, C. A., J. Davis, R. C. Christofferson, D. Wesson, J. M. Hyman, and C. N. Mores. 2014.** Towards an early warning system for forecasting human West Nile Virus incidence. *PLoS Curr.* 6.

- Marfin, A. A., L. R. Petersen, M. Eidson, J. Miller, J. Hadler, C. Farello, B. Werner, G. L. Campbell, M. Layton, and P. Smith. 2001.** Widespread West Nile virus activity, eastern United States, 2000. *Emerg. Infect. Dis.* 7: 730.
- Marra, P. P., S. Griffing, C. Caffrey, A. M. Kilpatrick, R. McLean, C. Brand, E. M. I. Saito, A. P. Dupuis, L. Kramer, and R. Novak. 2004a.** West Nile Virus and Wildlife. *BioScience* 54: 393.
- Marra, P. P., S. Griffing, C. Caffrey, M. A. Kilpatrick, R. McLean, C. Brand, E. Saito, A. P. Dupuis, L. Kramer, and R. Novak. 2004b.** West Nile virus and wildlife. *BioScience* 54: 393-402.
- Marzluff, J. M. 2001.** Worldwide urbanization and its effects on birds, pp. 19-47, *Avian ecology and conservation in an urbanizing world*. Springer.
- McCarthy, M. 2016.** First US case of Zika virus infection is identified in Texas. *Bmj* 352: i212.
- Medeiros, M. C., E. C. Boothe, E. B. Roark, and G. L. Hamer. 2017.** Dispersal of male and female *Culex quinquefasciatus* and *Aedes albopictus* mosquitoes using stable isotope enrichment. *PLoS Negl. Trop. Dis.* 11: e0005347.
- Messina, J. P., W. Brown, G. Amore, U. D. Kitron, and M. O. Ruiz. 2011.** West Nile virus in the Greater Chicago area: A geographic examination of human illness and risk from 2002 to 2006. *URISA J.* 23: 5-22.
- Meyer, R., J. Hardy, and W. Reisen. 1990.** Diel changes in adult mosquito microhabitat temperatures and their relationship to the extrinsic incubation of arboviruses in mosquitoes in Kern County, California. *J. Med. Entomol.* 27: 607-614.
- Miller, D. L., M. J. Mauel, C. Baldwin, G. Burtle, D. Ingram, and M. E. Hines. 2003.** West Nile virus in farmed alligators. *Emerg. Infect. Dis.* 9: 794.

- Milligan, K., E. Moretti, and P. Oreopoulos. 2004.** Does education improve citizenship? Evidence from the United States and the United Kingdom. *J. Public Econ.* 88: 1667-1695.
- Mirski, T., A. Bielawska-Drózd, and M. Bartoszcze. 2012.** Impact of Climate Change on Infectious Diseases. *Pol. J. Environ. Stud.* 21.
- Molaei, G., T. G. Andreadis, P. M. Armstrong, R. Bueno, Jr., J. A. Dennett, S. V. Real, C. Sargent, A. Bala, Y. Randle, H. Guzman, A. Travassos da Rosa, T. Wuithiranyagool, and R. B. Tesh. 2007.** Host Feeding Pattern of *Culex quinquefasciatus* (Diptera: Culicidae) and Its Role in Transmission of West Nile Virus in Harris County, Texas. *Am. J. Trop. Med. Hyg.* 77: 73-81.
- Mongoh, M. N., M. L. Khaita, and N. W. Dyer. 2007.** Environmental and ecological determinants of West Nile virus occurrence in horses in North Dakota, 2002. *Epidemiol. Infect.* 135: 57-66.
- Mouchet, J., J. Rageau, C. Laumond, C. Hannoun, D. Beytout, J. Oudar, B. Corniou, and A. Chippaux. 1970.** Epidemiologie du virus West Nile: etude d'un foyer en Camargue. *Inst. Pasteur Ann.*
- Mueller, C. W., and T. L. Parcel. 1981.** Measures of socioeconomic status: Alternatives and recommendations. *Child Dev.*: 13-30.
- Multi-Resolution Land Characteristics Consortium. 2018.** National Land Cover Database.
- Murray, K. O., E. Mertens, and P. Desprès. 2010.** West Nile virus and its emergence in the United States of America. *Vet. Res.* 41: 67.
- Murray, K. O., D. Ruktanonchai, D. Hesalroad, E. Fonken, and M. S. Nolan. 2013.** West Nile virus, Texas, USA, 2012. *Emerg. Infect. Dis.* 19: 1836-1838.

NASALPDAAC. NASA Land Processes Distributed Active Archive Center.

Nasci, R., M. Doyle, B. Biggerstaff, and A. LeBailly. Year. Published. Calculation and application of a vector index (VI) reflecting the number of WN virus infected mosquitoes in a population, pp. 3-7. *In*, 71st annual meeting of the American Mosquito Control Association, 2005.

Nasci, R. S., H. M. Savage, D. J. White, J. R. Miller, B. C. Cropp, M. S. Godsey, A. J. Kerst, P. Bennett, K. Gottfried, and R. S. Lanciotti. 2001a. West Nile virus in overwintering *Culex* mosquitoes, New York City, 2000. *Emerg. Infect. Dis.* 7: 742.

Nasci, R. S., D. J. White, H. Stirling, J. Oliver, T. J. Daniels, R. C. Falco, S. Campbell, W. J. Crans, H. M. Savage, R. S. Lanciotti, C. G. Moore, M. S. Godsey, K. L. Gottfried, and C. J. Mitchell. 2001b. West Nile Virus Isolates from Mosquitoes in New York and New Jersey, 1999. *Emerg. Infect. Diseases.* 7: 626-630.

Nava, M. R., and M. Debboun. 2016. A taxonomic checklist of the mosquitoes of Harris County, Texas. *J. Vector Ecol.* 41: 190-194.

Nelms, B. M., P. A. Macedo, L. Kothera, H. M. Savage, and W. K. Reisen. 2013. Overwintering Biology of *Culex* (Diptera: Culicidae) Mosquitoes in the Sacramento Valley of California. *J. Med. Entomol.* 50: 773-790.

Ng, K. C., L. F. Chaves, K. H. Tsai, and T. W. Chuang. 2018. Increased Adult *Aedes aegypti* and *Culex quinquefasciatus* (Diptera: Culicidae) Abundance in a Dengue Transmission Hotspot, Compared to a Coldspot, within Kaohsiung City, Taiwan. *Insects* 9.

Nguyen, A. T., A. J. Williams-Newkirk, U. D. Kitron, and L. F. Chaves. 2012. Seasonal Weather, Nutrients, and Conspecific Presence Impacts on the Southern House Mosquito

- Oviposition Dynamics in Combined Sewage Overflows. *J. Med. Entomol.* 49: 1328-1338.
- Nielsen, C. F., M. V. Armijos, S. Wheeler, T. E. Carpenter, W. M. Boyce, K. Kelley, D. Brown, T. W. Scott, and W. K. Reisen. 2008.** Risk factors associated with human infection during the 2006 West Nile virus outbreak in Davis, a residential community in northern California. *Am. J. Trop. Med. Hyg.* 78: 53-62.
- NOAA.** GPCP Version 2.3 Combined Precipitation Data Set.
- NOAA.** GHCN_CAMS Gridded 2m Temperature (Land).
- NOAA.** NOAA Climate Prediction Center.
- Nolan, M. S., J. Schuermann, and K. O. Murray. 2013.** West Nile virus infection among humans, Texas, USA, 2002-2011. *Emerg. Infect. Dis.* 19: 137-139.
- Nolan, M. S., A. Zangeneh, S. A. Khuwaja, D. Martinez, S. N. Rossmann, V. Cardenas, and K. O. Murray. 2012.** Proximity of residence to bodies of water and risk for west nile virus infection: a case-control study in Houston, Texas. *J. Biomed. Biotechnol.* 2012: 159578.
- Odelola, H., and O. Oduye. 1977.** West Nile virus infection of adult mice by oral route. *Arch. Virol.* 54: 251-253.
- Ozdenerol, E., E. Bialkowska-Jelinska, and G. N. Taff. 2008.** Locating suitable habitats for West Nile Virus-infected mosquitoes through association of environmental characteristics with infected mosquito locations: a case study in Shelby County, Tennessee. *Int. J. Health Geogr.* 7: 12.

- Paul, S., P. Rajagopalan, and M. Sreenivasan. 1970.** Isolation of the West Nile virus from the frugivorous bat, *Rousettus leschenaulti*. Indian Journal of Medical Research 58: 1169-1171.
- Paz, S., and I. Albersheim. 2008.** Influence of warming tendency on *Culex pipiens* population abundance and on the probability of West Nile fever outbreaks (Israeli Case Study: 2001-2005). EcoHealth 5: 40-48.
- Paz, S., and J. C. Semenza. 2013.** Environmental drivers of West Nile fever epidemiology in Europe and Western Asia--a review. Int. J. Environ. Res. Public Health 10: 3543-3562.
- Paz, S., D. Malkinson, M. S. Green, G. Tsioni, A. Papa, K. Danis, A. Sirbu, C. Ceianu, K. Katalin, E. Ferenczi, H. Zeller, and J. C. Semenza. 2013.** Permissive summer temperatures of the 2010 European West Nile fever upsurge. PloS one 8: e56398.
- Pealer, L. N., A. A. Marfin, L. R. Petersen, R. S. Lanciotti, P. L. Page, S. L. Stramer, M. G. Stobierski, K. Signs, B. Newman, and H. Kapoor. 2003.** Transmission of West Nile virus through blood transfusion in the United States in 2002. New Engl. J. Med. 349: 1236-1245.
- Peper, S. T., D. E. Dawson, N. Dacko, K. Athanasiou, J. Hunter, F. Loko, S. Almas, G. E. Sorensen, K. N. Urban, and A. N. Wilson-Fallon. 2018.** Predictive Modeling for West Nile Virus and Mosquito Surveillance in Lubbock, Texas. J. Am. Mosq. Control Assoc. 34: 18-24.
- Petersen, L. R., J. T. Roehrig, and J. J. Sejvar. 2007.** West Nile virus in the Americas, pp. 3-56, New and Evolving Infections of the 21st Century. Springer.
- Petersen, L. R., A. C. Brault, and R. S. Nasci. 2013a.** West Nile virus: review of the literature. J. A. Med. Assoc. 310: 308-315.

- Petersen, L. R., P. J. Carson, B. J. Biggerstaff, B. Custer, S. M. Borchardt, and M. P. Busch. 2013b.** Estimated cumulative incidence of West Nile virus infection in US adults, 1999-2010. *Epidemiol. Infect.* 141: 591-595.
- Peterson, A. T., D. A. Vieglaiss, and J. K. Andreasen. 2003.** Migratory birds modeled as critical transport agents for West Nile virus in North America. *Vector Borne Zoonotic Dis.* 3: 27-37.
- Peterson, A. T., N. Komar, O. Komar, A. Navarro-Sigüenza, M. B. Robbins, and E. Martínez-Meyer. 2004.** Priority contribution West Nile Virus in the New World: potential impacts on bird species. *Bird Conserv. Int.* 14: 215-232.
- Pettorelli, N., J. O. Vik, A. Mysterud, J.-M. Gaillard, C. J. Tucker, and N. C. Stenseth. 2005.** Using the satellite-derived NDVI to assess ecological responses to environmental change. *Trends Ecol. Evol.* 20: 503-510.
- Platt, K. B., B. J. Tucker, P. G. Halbur, S. Tiawsirisup, B. J. Blitvich, F. G. Fabiosa, L. C. Bartholomay, and W. A. Rowley. 2007.** West Nile virus viremia in eastern chipmunks (*Tamias striatus*) sufficient for infecting different mosquitoes. *Emerg. Infect. Dis.* 13: 831.
- Pollock, C. G. 2008.** West Nile virus in the Americas. *J. Avian Med. Surg.* 22: 151-157.
- Prilipov, A., R. Kinney, E. Samokhvalov, H. Savage, S. Al'khovskii, K. Tsuchiya, V. Gromashevskii, G. Sadykova, A. Shatalov, and O. Vyshemirskii. 2002.** Analysis of new variants of West Nile fever virus. *Vop. Virusol.* 47: 36-41.
- Randle, Y. H., C. B. Freeman, M. Jackson, M. Reyna, and M. Debboun. 2016.** 2014: A Record-Breaking Year for West Nile Virus Positive Mosquito Pools in Harris County and the City of Houston, Texas. *US Army Med. Dep. J.*

- Randolph, S. E., and D. J. Rogers. 2010.** The arrival, establishment and spread of exotic diseases: patterns and predictions. *Nat. Rev. Microbiol.* 8: 361-371.
- Rappole, J. H., S. R. Derrickson, and Z. Hubálek. 2000.** Migratory birds and spread of West Nile virus in the Western Hemisphere. *Emerg. Infect. Dis.* 6: 319.
- Reed, K. D., J. K. Meece, J. S. Henkel, and S. K. Shukla. 2003.** Birds, migration and emerging zoonoses: West Nile virus, Lyme disease, influenza A and enteropathogens. *Clin. Med. Res.* 1: 5-12.
- Reisen, W., and W. Reeves. 1990.** Bionomics and ecology of *Culex tarsalis* and other potential mosquito vector species.
- Reisen, W., and A. C. Brault. 2007.** West Nile virus in North America: perspectives on epidemiology and intervention. *Pest Manag. Sci.* 63: 641-646.
- Reisen, W. K. 1995.** Effect of temperature on *Culex tarsalis* (Diptera: Culicidae) from the Coachella and San Joaquin valleys of California. *J. Med. Entomol.* 32: 636-645.
- Reisen, W. K. 2010.** Landscape epidemiology of vector-borne diseases. *Annu. Rev. Entomol.* 55: 461-483.
- Reisen, W. K. 2012.** The contrasting bionomics of *Culex* mosquitoes in western North America. *J. Am. Mosq. Control Assoc.* 28: 82-91.
- Reisen, W. K., R. P. Meyer, and M. M. Milby. 1986.** Overwintering studies on *Culex tarsalis* (Diptera: Culicidae) in Kern County, California: temporal changes in abundance and reproductive status with comparative observations on *C. quinquefasciatus* (Diptera: Culicidae). *Ann. Entomol. Soc. Am.* 79: 677-685.

- Reisen, W. K., M. M. Milby, and R. P. Meyer. 1992.** Population dynamics of adult *Culex* mosquitoes (Diptera: Culicidae) along the Kern River, Kern County, California, in 1990. J. Med. Entomol. 29: 531-543.
- Reisen, W. K., Y. Fang, and V. M. Martinez. 2006a.** Effects of Temperature on the Transmission of West Nile Virus *Culex tarsalis* (Diptera: Culicidae). J. Med. Entomol. 43: 309-317.
- Reisen, W. K., R. M. Takahashi, B. D. Carroll, and R. Quiring. 2008a.** Delinquent mortgages, neglected swimming pools, and West Nile virus, California. Emerg. Infect. Dis. 14: 1747-1749.
- Reisen, W. K., D. Cayan, M. Tyree, C. M. Barker, B. Eldridge, and M. Dettinger. 2008b.** Impact of climate variation on mosquito abundance in California. J. Vector Ecol. 33: 89-98.
- Reisen, W. K., B. D. Carroll, R. Takahashi, Y. Fang, S. Garcia, V. M. Martinez, and R. Quiring. 2009.** Repeated west Nile virus epidemic transmission in Kern County, California, 2004–2007. J. Med. Entomol. 46: 139-157.
- Reisen, W. K., T. Thiemann, C. M. Barker, H. Lu, B. Carroll, Y. Fang, and H. D. Lothrop. 2010.** Effects of Warm Winter Temperature on the Abundance and Gonotrophic Activity of *Culex* (Diptera: Culicidae) in California. J. Med. Entomol. 47: 230-237.
- Reisen, W. K., H. D. Lothrop, S. S. Wheeler, M. Kennsington, A. Gutierrez, Y. Fang, S. Garcia, and B. Lothrop. 2008c.** Persistent West Nile virus transmission and the apparent displacement St. Louis encephalitis virus in southeastern California, 2003-2006. J. Med. Entomol. 45: 494-508.

- Reisen, W. K., Y. Fang, H. D. Lothrop, V. M. Martinez, J. Wilson, P. O'Connor, R. Carney, B. Cahoon-Young, M. Shafii, and A. C. Brault. 2006b.** Overwintering of West Nile Virus in Southern California. *J. Med. Entomol.* 43: 344-355.
- Reiter, M. E., and D. A. LaPointe. 2007.** Landscape factors influencing the spatial distribution and abundance of mosquito vector *Culex quinquefasciatus* (Diptera: Culicidae) in a mixed residential-agricultural community in Hawai'i. *J. Med. Entomol.* 44: 861-868.
- Reiter, P. 1998.** *Aedes albopictus* and the world trade in used tires, 1988-1995: the shape of things to come? *J. Am. Mosq. Control Assoc.* 14: 83-94.
- Richards, S. L., C. N. Mores, C. C. Lord, and W. J. Tabachnick. 2007.** Impact of extrinsic incubation temperature and virus exposure on vector competence of *Culex pipiens quinquefasciatus* Say (Diptera: Culicidae) for West Nile virus. *Vector Borne Zoonotic Dis.* 7: 629-636.
- Rios, J., C. S. Hacker, C. A. Hailey, and R. E. Parsons. 2006.** Demographic and Spatial Analysis of West Nile Virus and St. Louis Encephalitis in Houston, Texas. *J. Am. Mosq. Control Assoc.* 22: 254-263.
- Rochlin, I., D. Turbow, F. Gomez, D. V. Ninivaggi, and S. R. Campbell. 2011.** Predictive mapping of human risk for West Nile virus (WNV) based on environmental and socioeconomic factors. *PloS one* 6: e23280.
- Roehr, B. 2012.** US hit by massive West Nile virus outbreak centred around Texas. *Bmj* 345: e5633.
- Roehrig, J., M. Layton, P. Smith, G. Campbell, R. Nasci, and R. Lanciotti. 2002.** The emergence of West Nile virus in North America: ecology, epidemiology, and surveillance, pp. 223-240, Japanese encephalitis and West Nile viruses. Springer.

- Rogers, D. J., and S. E. Randolph. 2003.** Studying the global distribution of infectious diseases using GIS and RS. *Nat. Rev. Microbiol.* 1: 231-237.
- Rueda, L., K. Patel, R. Axtell, and R. Stinner. 1990.** Temperature-dependent development and survival rates of *Culex quinquefasciatus* and *Aedes aegypti* (Diptera: Culicidae). *J. Med. Entomol.* 27: 892-898.
- Ruiz, M. O., C. Tedesco, T. J. McTighe, C. Austin, and U. Kitron. 2004.** Environmental and social determinants of human risk during a West Nile virus outbreak in the greater Chicago area, 2002. *Int. J. Health Geogr.* 3: 1-11.
- Ruiz, M. O., E. D. Walker, E. S. Foster, L. D. Haramis, and U. D. Kitron. 2007.** Association of West Nile virus illness and urban landscapes in Chicago and Detroit. *Int. J. Health Geogr.* 6: 10.
- Ruiz, M. O., L. F. Chaves, G. L. Hamer, T. Sun, W. M. Brown, E. D. Walker, L. Haramis, T. L. Goldberg, and U. D. Kitron. 2010.** Local impact of temperature and precipitation on West Nile virus infection in *Culex* species mosquitoes in northeast Illinois, USA. *Parasit. Vectors* 3: 19.
- Ruktanonchai, D. J., S. Stonecipher, N. Lindsey, J. McAllister, S. K. Pillai, K. Horiuchi, M. Delorey, B. J. Biggerstaff, T. Sidwa, J. Zoretic, R. Nasci, M. Fischer, and S. L. Hills. 2014.** Effect of aerial insecticide spraying on West Nile virus disease--north-central Texas, 2012. *Am. J. Trop. Med. Hyg.* 91: 240-245.
- Savage, H., and B. Miller. 1995.** House mosquitoes of the USA, *Culex pipiens* complex. *Wing Beats* 6: 8-9.
- Schweitzer, B. K., N. M. Chapman, and P. C. Iwen. 2009.** Overview of the Flaviviridae With an Emphasis on the Japanese Encephalitis Group Viruses. *Lab. Med.* 40: 493-499.

- Shaman, J., and J. F. Day. 2005.** Achieving operational hydrologic monitoring of
mosquitoborne disease. *Emerg. Infect. Dis.* 11: 1343.
- Shaman, J., and J. F. Day. 2007.** Reproductive Phase Locking of Mosquito Populations in
Response to Rainfall Frequency. *PLoS One* 2: e331.
- Shaman, J., J. F. Day, and M. Stieglitz. 2002a.** Drought-induced amplification of Saint Louis
encephalitis virus, Florida. *Emerg. Infect. Dis.* 8: 575.
- Shaman, J., J. F. Day, and M. Stieglitz. 2005.** Drought-Induced Amplification and Epidemic
Transmission of West Nile Virus in Southern Florida. *J. Med. Entomol.* 42: 134-141.
- Shaman, J., M. Stieglitz, C. Stark, S. Le Blancq, and M. Cane. 2002b.** Using a Dynamic
Hydrology Model To Predict Mosquito Abundances in Flood and Swamp Water. *Emerg.
Infect. Dis.* 8: 6-13.
- Shand, L., W. M. Brown, L. F. Chaves, T. L. Goldberg, G. L. Hamer, L. Haramis, U.
Kitron, E. D. Walker, and M. O. Ruiz. 2016.** Predicting West Nile Virus Infection Risk
From the Synergistic Effects of Rainfall and Temperature. *J. Med. Entomol.* 53: 935-944.
- Shumway, R. H., and D. S. Stoffer. 2011.** Time series regression and exploratory data analysis,
pp. 47-82, *Time series analysis and its applications.* Springer.
- Simmonds, P., P. Becher, J. Bukh, E. A. Gould, G. Meyers, T. Monath, S. Muerhoff, A.
Pletnev, R. Rico-Hesse, and D. B. Smith. 2017.** ICTV virus taxonomy profile:
Flaviviridae. *J. Gen. Virol.* 98: 2-3.
- Smith, C. G. 1987.** Factors influencing the transmission of western equine encephalomyelitis
virus between its vertebrate maintenance hosts and from them to humans. *Am. J. Trop.
Med. Hyg.* 37: 33S-39S.

- Soverow, J. E., G. A. Wellenius, D. N. Fisman, and M. A. Mittleman. 2009.** Infectious disease in a warming world: how weather influenced West Nile virus in the United States (2001-2005). *Environ. Health Perspect.* 117: 1049-1052.
- Speybroeck, N., C. J. Williams, K. B. Lafia, B. Devleesschauwer, and D. Berkvens. 2012.** Estimating the prevalence of infections in vector populations using pools of samples. *Med. Vet. Entomol.* 26: 361-371.
- Stanley, E., R. Ratard, J. Staples, R. Royce, W. Bower, K. Ellingson, and M. Kuehnert. 2009.** West Nile virus transmission via organ transplantation and blood transfusion-Louisiana, 2008. *MMWR Morb. Mortal. Wkly. Rep.* 58: 1263-1267.
- Steinman, A., C. Banet-Noach, S. Tal, O. Levi, L. Simanov, S. Perk, M. Malkinson, and N. Shpigel. 2003.** West Nile virus infection in crocodiles. *Emerg. Infect. Dis.* 9: 887.
- Strickman, D. 1988.** Rate of oviposition by *Culex quinquefasciatus* in San Antonio, Texas, during three years. Walter Reed Army Institution of Research Washington D.C. Department of Entomology.
- Strickman, D., and J. T. Lang. 1986.** Activity of *Culex quinquefasciatus* in an underground storm drain in San Antonio, Texas. Walter Reed Biosystematics Unit Washington D.C.
- Su, T., J. Webb, R. Meyer, and M. Mulla. 2003.** Spatial and temporal distribution of mosquitoes in underground storm drain systems in Orange County, California. *J. Vector Ecol.* 28: 79-89.
- Takasaki, T. 2007.** West Nile fever/encephalitis. *Uirusu* 57: 199-205.
- Takeda, T., C. A. Whitehouse, M. Brewer, A. D. Gettman, and T. N. Mather. 2003.** Arbovirus surveillance in Rhode Island: assessing potential ecologic and climatic correlates. *J. Am. Mosq. Control Assoc.* 19: 179-189.

- Tempelis, C., W. Reeves, R. Bellamy, and M. Lofy. 1965.** A three-year study of the feeding habits of *Culex tarsalis* in Kern County, California. *Am. J. Trop. Med. Hyg.* 14: 170-177.
- Tesh, R. B., R. Parsons, M. Siirin, Y. Randle, C. Sargent, H. Guzman, T. Wuithiranyagool, S. Higgs, D. L. Vanlandingham, and A. A. Bala. 2004a.** Year-round West Nile virus activity, Gulf coast region, Texas and Louisiana. *Emerging infectious diseases* 10: 1649.
- Tesh, R. B., R. Parsons, M. Siirin, Y. Randle, C. Sargent, H. Guzman, T. Wuithiranyagool, S. Higgs, D. L. Vanlandingham, A. A. Bala, K. Haas, and B. Zerinque. 2004b.** Year-round West Nile Virus Activity, Gulf Coast Region, Texas and Louisiana. *Emerg. Infect. Dis.* 10: 1649-1652.
- Thomson, M., and S. Connor. 2000.** Environmental information systems for the control of arthropod vectors of disease. *Med. Vet. Entomol.* 14: 227-244.
- Thomson, M. C., F. J. Doblas-Reyes, S. J. Mason, R. Hagedorn, S. J. Connor, T. Phindela, A. P. Morse, and T. N. Palmer. 2006.** Malaria early warnings based on seasonal climate forecasts from multi-model ensembles. *Nature* 439: 576-579.
- Tiawsirisup, S., K. B. Platt, B. J. Tucker, and W. A. Rowley. 2005.** Eastern cottontail rabbits (*Sylvilagus floridanus*) develop West Nile virus viremias sufficient for infecting select mosquito species. *Vector Borne Zoonotic Dis.* 5: 342-350.
- Trawinski, P. R., and D. S. Mackay. 2010.** Identification of environmental covariates of West Nile virus vector mosquito population abundance. *Vector Borne Zoonotic Dis.* 10: 515-526.
- Turell, M., M. Sardelis, M. O'guinn, and D. Dohm. 2002.** Potential vectors of West Nile virus in North America, pp. 241-252, Japanese encephalitis and West Nile viruses. Springer.

- Turell, M. J., M. O'Guinn, and J. Oliver. 2000.** Potential for New York mosquitoes to transmit West Nile virus. *Am. J. Trop. Med. Hyg.* 62: 413-414.
- Turell, M. J., M. L. O'Guinn, D. J. Dohm, and J. W. Jones. 2001.** Vector competence of North American mosquitoes (Diptera: Culicidae) for West Nile virus. *J. Med. Entomol.* 38: 130-134.
- Turell, M. J., D. J. Dohm, M. R. Sardelis, M. L. O'guinn, T. G. Andreadis, and J. A. Blow. 2005.** An Update on the Potential of North American Mosquitoes (Diptera: Culicidae) to Transmit West Nile Virus. *J. Med. Entomol.* 42: 57-62.
- Uejio, C. K., A. Kemp, and A. C. Comrie. 2012.** Climatic controls on West Nile virus and Sindbis virus transmission and outbreaks in South Africa. *Vector Borne Zoonotic Dis.* 12: 117-125.
- United States Census Bureau. 2016.** QuickFacts.
- United States Census Bureau. 2018.** American FactFinder.
- United States Department of Agriculture Animal and Plant Health Inspection Service. 2018.** Equine West Nile Virus Case Reporting and Surveillance Information.
- United States Geological Survey. 2018a.** The National Map - Elevation.
- United States Geological Survey. 2018b.** Hydrography: Links to Data Products and Map Services.
- Unlu, I., A. J. Mackay, A. Roy, M. M. Yates, and L. D. Foil. 2010.** Evidence of vertical transmission of West Nile virus in field-collected mosquitoes. *J. Vector Ecol.* 35: 95-99.
- Venables, W. N., and B. D. Ripley. 2002.** Modern applied statistics with S, Springer, New York.

- Vinogradova, E. B. 2007.** Diapause in aquatic insects, with emphasis on mosquitoes, pp. 83-113, Diapause in Aquatic Invertebrates Theory and Human Use. Springer.
- Walsh, A. S., G. E. Glass, C. R. Lesser, and F. C. Curriero. 2007.** Predicting seasonal abundance of mosquitoes based on off-season meteorological conditions. Environ. Ecol. Stat. 15: 279-291.
- Walther, G.-R., E. Post, P. Convey, A. Menzel, C. Parmesan, T. J. Beebee, J.-M. Fromentin, O. Hoegh-Guldberg, and F. Bairlein. 2002.** Ecological responses to recent climate change. Nature 416: 389.
- Walton, W. E., A. R. Van Dam, and D. A. Popko. 2009.** Ovipositional Responses of Two *Culex* (Diptera: Culicidae) Species to Larvivorous Fish. J. Med. Entomol. 46: 1338-1343.
- Ward, M. P., B. H. Ramsay, and K. Gallo. 2005.** Rural cases of equine West Nile virus encephalomyelitis and the normalized difference vegetation index. Vector Borne Zoonotic Dis. 5: 181-188.
- Weaver, S. C., and W. K. Reisen. 2010.** Present and future arboviral threats. Antivir. Res. 85: 328-345.
- White, S. L., M. P. Ward, C. M. Budke, T. Cyr, and R. Bueno Jr. 2009.** A comparison of gravid and under-house CO₂-baited CDC light traps for mosquito species of public health importance in Houston, Texas. J. Med. Entomol. 46: 1494-1497.
- Wiens, J. A. 1995.** Habitat fragmentation: island v landscape perspectives on bird conservation. Ibis 137: S97-S104.
- Williams, G. M., and J. B. Gingrich. 2007.** Comparison of light traps, gravid traps, and resting boxes for West Nile virus surveillance. J. Vector Ecol. 32: 285.

- Wimberly, M. C., A. Lamsal, P. Giacomo, and T. W. Chuang. 2014.** Regional variation of climatic influences on West Nile virus outbreaks in the United States. *Am. J. Trop. Med. Hyg.* 91: 677-684.
- Winters, A. M., B. G. Bolling, B. J. Beaty, C. D. Blair, R. J. Eisen, A. M. Meyer, W. J. Pape, C. G. Moore, and L. Eisen. 2008.** Combining mosquito vector and human disease data for improved assessment of spatial West Nile virus disease risk. *Am. J. Trop. Med. Hyg.* 78: 654-665.
- Winters, A. M., R. J. Eisen, M. J. Delorey, M. Fischer, R. S. Nasci, E. Zielinski-Gutierrez, C. G. Moore, W. J. Pape, and L. Eisen. 2010.** Spatial risk assessments based on vector-borne disease epidemiologic data: importance of scale for West Nile virus disease in Colorado. *Am. J. Trop. Med. Hyg.* 82: 945-953.
- Work, T. H., H. S. Hurlbut, and R. Taylor. 1953.** Isolation of West Nile virus from hooded crow and rock pigeon in the Nile delta. *P. Soc. Exp. Biol. Med.* 84: 719-722.

APPENDIX

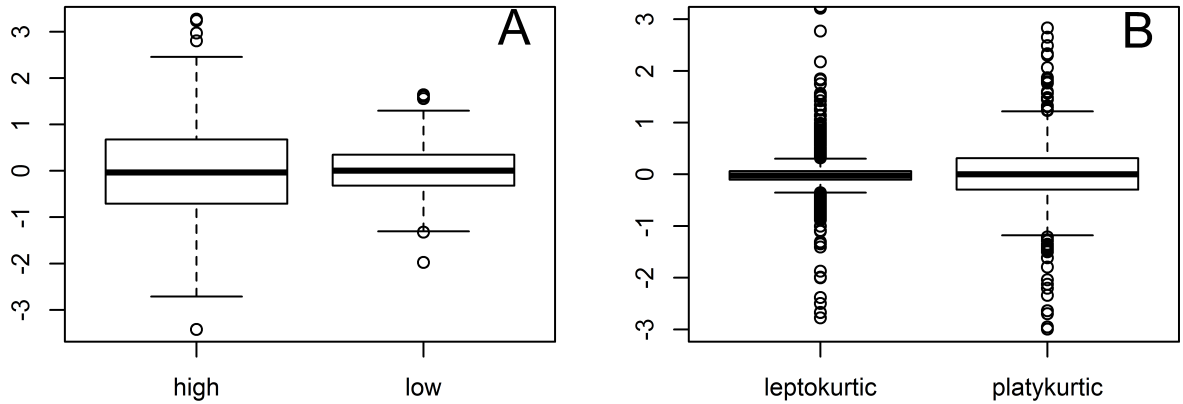


Figure A1 Variance and kurtosis distributions (A) Distributions with different variance: high=1, low=0.25. These distributions were obtained simulating 1000 replicates from a normal distribution with mean 0 using R. (B) Distributions with different kurtosis: leptokurtic=178.8; platykurtic=115.2. Here is worth highlighting how the leptokurtic variable has low variability within the 2nd to 3rd quartile and how such variability is wider in the platykurtic distribution. These distributions were obtained simulating 1000 replicates from T-student distributions using R, with 3 and 1 degrees of freedom, respectively, for the leptokurtic and platykurtic distributions. Both distributions have variance of 1, and mean 0.

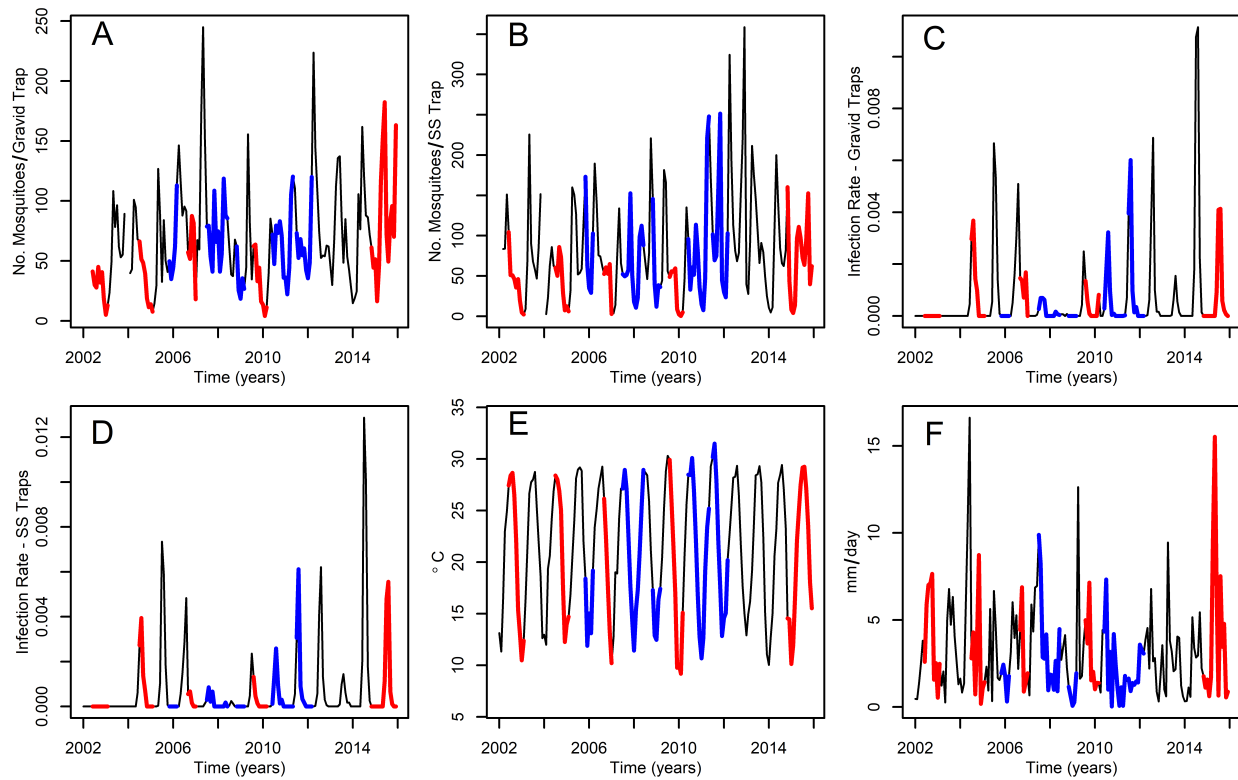


Figure A2 Additional Monthly Time Series. (A) Monthly average mosquito abundance per gravid trap and (B) Monthly average mosquito abundance per storm sewer trap. (C) West Nile Virus mosquito infection rate based on gravid traps. (D) West Nile Virus mosquito infection rate based on storm sewer traps (E) Gridded temperature. (F) Gridded rainfall. In all panels ENSO phases are highlighted by colors, for details, see inset legend of panel A.

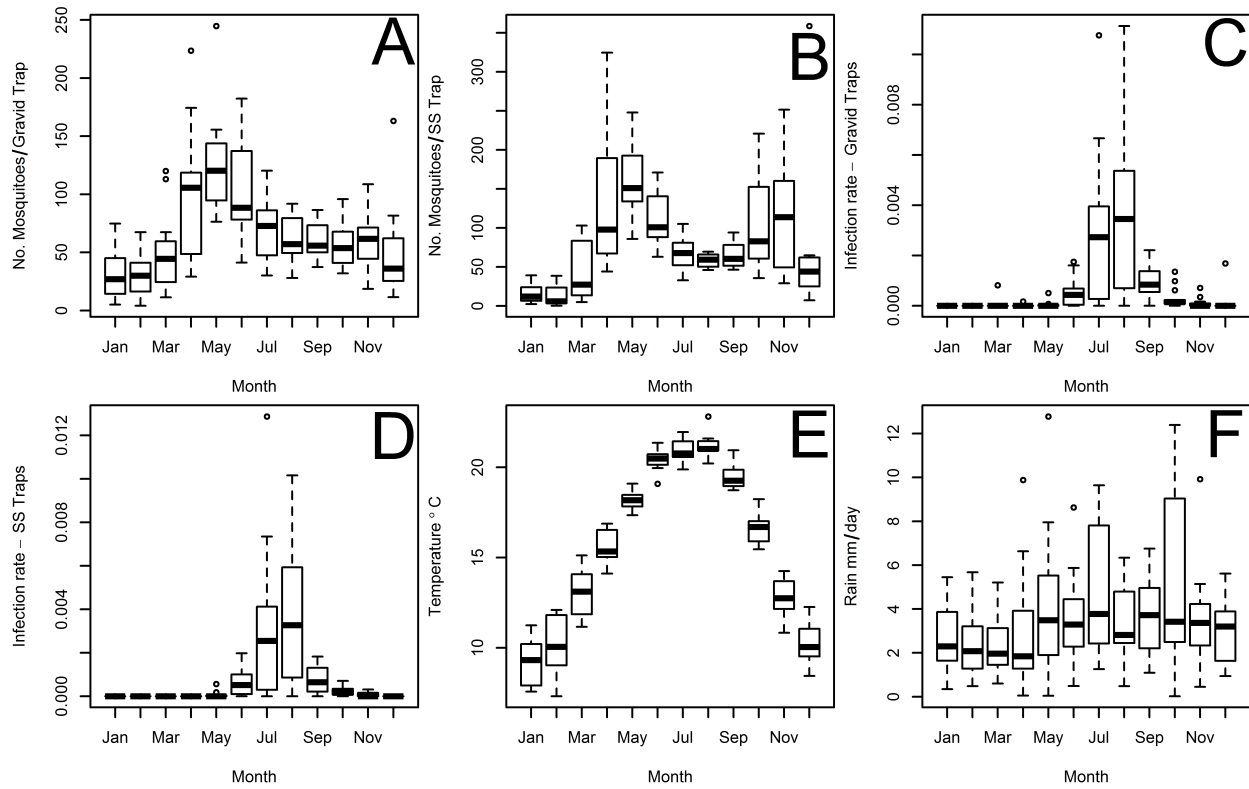


Figure A3 Additional Seasonal Monthly Boxplots. (A) Monthly average mosquito abundance per gravid trap and (B) Monthly average mosquito abundance per storm sewer trap. (C) West Nile Virus mosquito infection rate based on gravid traps. (D) West Nile Virus mosquito infection rate based on storm sewer traps. (E) Gridded temperature. (F) Gridded rainfall.

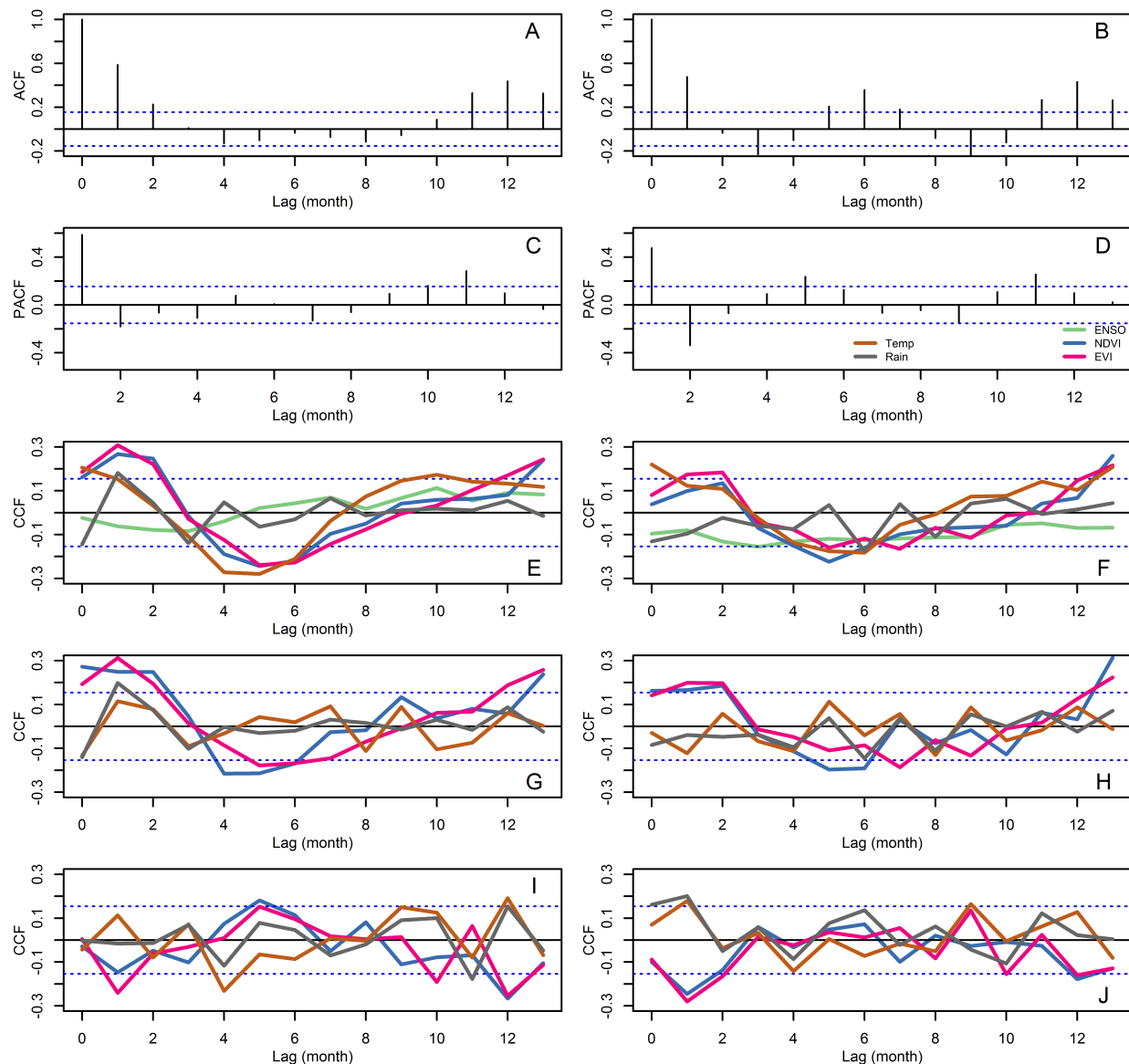


Figure A4 Correlation Functions. Auto-Correlation function (ACF) of (A) Monthly average mosquito abundance per gravid trap (MAMAPGT) and (B) Monthly average mosquito abundance per storm sewer trap (MAMAPSST). Partial Auto-Correlation Function (PACF) of (C) MAMAPGT and (D) MAMAPSST. Cross-Correlation Functions (CCF) of the average value of environmental variables with (E) MAMAPGT and (F) MAMAPSST. CCF of the Standard Deviation (SD) of environmental variables with (G) MAMAPGT and (H) MAMAPSST. CCF of the Kurtosis (K) of environmental variables with (I) MAMAPGT and (J) MAMAPSST. In panels E to J environmental variables are color coded, for details, please refer to the inset legend of panel D.

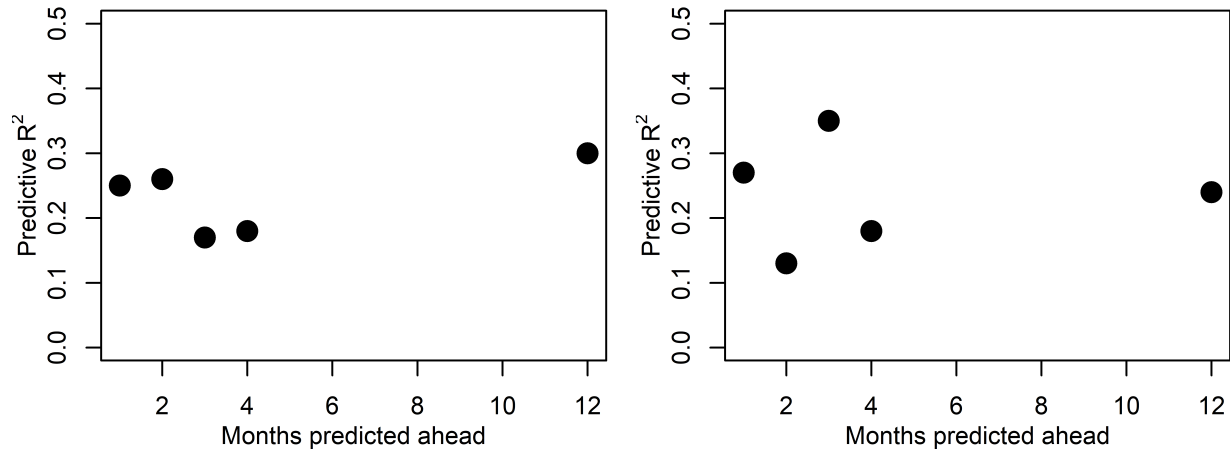


Figure A5 Predictive R². Predictive capability for models selected as best to explain (left) Monthly average mosquito abundance per gravid trap (right) Monthly average mosquito abundance per storm sewer trap.

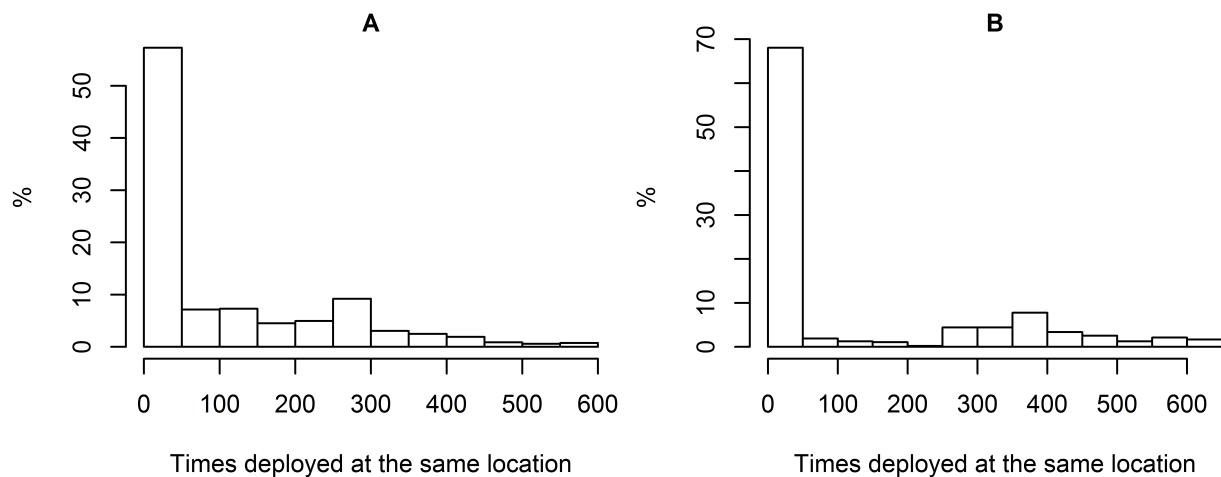


Figure A6 Trapping Consistency. Percentage of the number of times a trap was visited at the same location. The x-axis represents the number of times a trap was deployed at the same location and the y-axis represents the percentage of times the trap was deployed at the same location.

Additional supplementary maps (Figures S1-S6) for Chapter III are included as separate files.

Supplementary tables (Tables S1-S5, Chapter IV) for Chapter IV are included as separate files.