Urbanization, Land Cover, Weather, and Incidence Rates of Neuroinvasive West Nile Virus Infection

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THESIS

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LIST OF ABBREVIATIONS

BRACE – Building Resilience Against Climate Effects

CBSA - Core Based Statistical Areas

CDC - United States Centers for Disease Control and Prevention

EIP - Extrinsic Incubation Period

IDPH – Illinois Department of Public Health

IgG – Immunoglobulin G

IgM – Immunoglobulin M

MRCC - Midwest Regional Climate Center

NOAA - National Oceanic and Atmospheric Administration

NWNV - Neuroinvasive West Nile Virus

RUCC - Rural to Urban Continuum Code

UK – United Kingdom

US - United States

USDA – United States Department of Agriculture

USGS – United States Geological Survey

WND - West Nile Disease

WNF - West Nile Fever

WNV - West Nile Virus

ZINB – Zero-inflated Negative Binomial

CHAPTER I. INTRODUCTION

West Nile Virus (WNV) is a widespread zoonotic disease that infects birds, humans, and horses. WNV is part of the Flavivirus family which includes the yellow fever virus, dengue virus, St. Louis encephalitis virus, and Zika virus (Hayes et al. 2005, CDC 2016). Flaviviruses are generally transmitted by bites from an infected arthropod, usually ticks or mosquitoes; some flaviviral infections result in severe neurological consequences (Fernandez-Garcia et al. 2009). WNV is a global infectious disease with laboratory confirmed cases reported in every continent except Antarctica (Hayes et al. 2005a). The virus was first discovered when it was isolated from a blood sample of an infected Ugandan woman in 1937. It appears that WNV originated in Africa and then spread to the Middle East eventually making its way to Asia, Europe, Australia, and North and South America (Komar et al. 2003). WNV is relatively new to the Western Hemisphere; the first reported human case of WNV was in New York City in 1999. Since then, WNV quickly spread throughout the Western Hemisphere and has become endemic in North America, with some regions consistently having high annual incidence rates compared to the rest of the continent. From 1999-2014 in the US a total of 41,762 cases of WNV were reported to the US Centers for Disease Control and Prevention (CDC) (CDC 2015b), 18,810 of these cases were neuroinvasive, and 1,765 illnesses were fatal (CDC 2015b). Most WNV cases in the US occur in late summer from July to September. WNV is highly underreported; a seroprevalence study conducted by Peterson et al. estimated that 3 million people in the United States (US) were infected from 1999 to 2010 (Peterson et al. 2013). Based on Peterson's study, 6.3 per 1000 WNV cases results were neuroinvasive, with a case fatality rate of 0.6 deaths per 1000 WNV cases. In some published literature, WNV infections are sometimes referred to as WNV, West Nile Fever (WNF), neuroinvasive WNV (NWNV), or West Nile Disease (WND) where WND refers to neuroinvasive disease. For the purposes of this paper neuroinvasive infections are referred to as NWNV.

A. Transmission

The WNV transmission cycle is maintained between female mosquitoes and birds. Male mosquitoes feed exclusively on plant sugars, but female mosquitoes need to feed on blood to for proper egg development (Capinera et al. 2008). After feeding on an infected bird, and after the extrinsic incubation period (EIP) the female mosquito can transmit WNV in a subsequent blood meal. The EIP describes the time it takes from the acquisition of the by a mosquito to the time it is able to transmit WNV. The EIP is at least 12 days but is dependent on viral dose, temperature, and type of transmission (Anderson et al. 2008, Reisen et al. 2006). Humans and horses are considered “dead end hosts” as WNV concentrations in the blood of infected humans and horses do not reach levels that would make further transmission (from human to uninfected mosquitos) likely (Petersen et al. 2013, Zou et al. 2010, Pealer et al. 2003). Certain species of mosquitos have been shown to be more competent vectors than others, and certain types of birds have been shown to be more efficient reservoirs. Generally, the primary vectors for WNV are mosquitoes from the genus Culex and the primary reservoirs are birds from the order Passeriformes. The specific species of primary WNV vectors and reservoirs varies by geographic region.

Vectorial capacity refers to all processes that affect the ability of a vector to function as a vector for a pathogen; this includes biological and environmental processes. Virus, mosquito, and host genetics, feeding and migration patterns of both the vector and host, and longevity of vector and host. In the case of WNV factors that affect vectorial capacity includes: viral lineage, sex of mosquito, mosquito and bird species, type of land cover, temperature, precipitation, mosquito and bird population dynamics, mosquito and bird ecology, and proximity of mosquitoes to birds and humans. Each of these factors will be discussed in further detail.

Laboratory studies have shown that it is possible for some species of ticks to become infected with WNV and transmit the infection to chickens and birds (Hutcheson et al. 2005), but ticks are unlikely to be a major vector for WNV transmission (Hayes et. al 2005a). Cases of WNV have also been reported through less common transmission pathways including: blood transfusion, organ donation, intrauterine, breast milk, and aerosol exposure in an occupational setting (Lindsey et al. 2010).

1. Transmission Vectors

WNV has been detected in 65 different species of mosquitoes, but only a few species are considered a primary vector for the WNV transmission cycle. Laboratory studies have shown mosquitoes in the genus Culex appear to be the most competent vectors of WNV (Hayes et. al, 2005a). The predominant Culex species varies geographically and some Culex can inter-breed and hybridize in areas where there is overlap of two Culex species (CDC 2013a). *Culex pipiens* accounts for over half of WNV positive pools of mosquitoes in the US, but *Culex quinquefasciatus* is the predominant WNV vector in the south and *Culex tarsalis* is the predominant WNV vector along the Mississippi River (Hayes et. al 2005a). *Culex restuans* and *Culex pipiens* are often found in similar regions and habitats, but mosquito population density for each species peaks at different times of the year. The two species have similar morphology and differentiating these species on morphological characteristics has high error rates (Harrington et al. 2008).

a. Culex Mosquito Life Cycle

The life cycle of Culex mosquitoes is sensitive to environmental factors including temperature and precipitation. The mosquito habitat varies throughout the life cycle that begins as an egg then goes through a complete metamorphosis to larvae, pupae, and finally an adult (Capinera et al. 2008). Female mosquitoes generally lay between 50-200 eggs on the surface of water or in an area that will flood;

water is necessary for mosquito eggs to hatch. Mosquitoes in larval and pupal stages live in the aquatic habitat where the eggs were laid, and do not feed or fly until they are adults. The amount time spent in the pupal stage is temperature dependent, but is generally two to three days (Capinera et al. 2008). Adult mosquitoes live in terrestrial habitats, but females need to stay near a water source to lay eggs and tend to thrive in water with organic matter. Longevity is temperature dependent, and extreme high or low temperatures can decrease the average mosquito lifespan and activity level (Andreadis et al. 2014). The life of an adult *Culex pipiens* can be as long as 130 days. Female mosquitoes overwinter in covered areas where the temperature tends to be warmer and stable compared to an exposed habitat, this includes caves, sewer systems, and underground cellars. During this time reproductive growth is temporarily suspended in diapause. Vertical transmission from mother to offspring can occur when an infected female mosquito transmits WNV to her eggs (Anderson et al. 2008).

The population dynamics of Culex mosquitos play an important role in WNV transmission cycle. In Illinois the population density of *Culex restuans* peaks in late spring and *Culex pipiens* peaks in late summer. The time at which the population density is equal for both species is called the crossover date. The crossover date varies from year to year, but can be predicted using a degree day model based on the number of days the maximum temperature exceeds 27^{o} C (Kunkel et al. 2006). In spring *Culex restuans* is responsible for bird to mosquito to bird amplification of WNV, and in summer *Culex pipiens* is responsible for maintaining the WNV transmission cycle between mosquitos and birds. Culex pipiens is also responsible for much of the mosquito to human transmission of WNV (Kunkel et al. 2006, Ciota et al. 2013).

2. <u>Transmission Hosts</u>

Certain types of birds may be more effective reservoirs than others, and therefore play a more substantial role in the viral transmission cycle. In 2003 Komar et al. infected 25 different species of birds with the 1999 strain of WNV and measured levels of viremia; 17 species became infected with WNV and tested positive for cloacal or oral viral shedding. Five bird species acquired WNV through oral transmission and four species through contact transmission. In this experiment viremia was also measured to evaluate each species potential as a reservoir in the WNV transmission cycle. The five most competent species were: blue jays, common grackles, house finches, American crows, and the house sparrows (Komar et al. 2003). Birds that have become infected with WNV and survived appear to develop immunity to WNV for the remainder of their lifetime, approximately two years (Perez-Ramirez et al. 2014). It is common to observe low seasonal WNV rates for about two years following a year with particularly high rates. A study in California measured WNV antibodies in birds and found an inverse association between reported human WNV incidence rates and seroprevalence of WNV antibodies (Kwan et al. 2012).

In Illinois blue jays, American crows, grackles, starlings, robins, cardinals, sparrows, finches, hawks and owls are common WNV reservoir (IDPH 2016b, Lampman et al. 2013). Most of these birds migrate away in the fall and return in the spring.

B. Symptoms and Diagnosis

Most WNV infections in humans are subclinical; about 80% of people infected do not develop any symptoms (CDC 2015). For symptomatic infections the incubation period ranges from two to 14 days, but can take longer in the immunocompromised (CDC 2015). In symptomatic cases of WNV infection, symptoms are flu-like: fever, headache, nausea, vomiting, and weakness (Hayes et al. 2005). Less than one percent of infections become neuroinvasive with meningitis, encephalitis, or acute flaccid paralysis. WNV meningitis symptoms include fever, neck stiffness, and headaches. WNV encephalitis symptoms include fever along with more severe neurologic symptoms including altered mental status, seizures, or tremors. Patients with WNV acute flaccid paralysis usually have paresis (weakness) or paralysis in one limb, and the infection can progress to respiratory paralysis. Patients with WNV acute flaccid paralysis may also have WNV meningitis or encephalitis. There are three laboratory tests to confirm a diagnosis of WNV. Diagnosis of WNV is confirmed by testing blood or cerebrospinal fluid for WNV specific immunoglobulin M (IgM) antibodies, a positive test indicates a recent infection but a positive test for WNV specific immunoglobulin G (IgG) antibodies indicates a past infection. Methods using reverse transcriptase-polymerase chain reaction and immunohistochemistry can also detect WNV in cerebrospinal fluid, blood, or tissues (CDC 2015). People with symptomatic WNV infections may have adverse long term health effects including tremors, postpolio syndrome, persistent fatigue, and problems with memory or concentration (Sejvar et al. 2008, Cook et al. 2010). A WNV cohort study in Houston, Texas found that 40% of cohort members had chronic kidney disease that was associated with having NWNV 4-9 years prior (Nolan et al. 2012).

All ages and races are equally affected by WNV, but people over the age of 50 are more likely to develop neuroinvasive disease. Many of the severe cases are associated with age or underlying medical conditions such as hypertension, diabetes mellitus, or immunosuppression (CDC 2013a). In Toronto seroprevalence of WNV specific IgM antibodies was measured in 833 organ transplant patients, and estimated a 40% risk of WNV infections to become neuroinvasive in organ transplant patients (Komar et al. 2004). There is no vaccine or preventive medicine for WNV. Treatment of a WNV infected patient is limited to supportive care for symptoms.

C. Surveillance and Prevention

Mosquito surveillance is useful to detect areas with high mosquito infection rates. Surveillance of WNV infections among mosquitoes consists of setting out mosquito traps in different areas for a period of time then collecting the mosquito trap. There are different types of mosquito traps, the CDC has designed both gravid traps and light traps. Gravid traps are designed to collect gravid (pregnant) female mosquitoes and light traps collect both males and females. After the traps are collected mosquitos are separated by species in pools of 50-100 mosquitoes and each pool is tested for WNV (CDC 2013a). Infections in mosquitoes are reported based on the percent of mosquito pools that test positive for WNV, rather than the percent of mosquitoes.

Human WNV infection is a nationally reportable disease in the US. Currently ArboNET serves as a passive surveillance system for arboviral infections including WNV. ArboNET was developed in 2000 and is managed by the CDC as a response to the 1999 WNV outbreak in New York. Physicians, veterinarians, and labs can report confirmed or suspected cases of WNV or other arboviral illnesses to their local health department. The health department then submits the report electronically to ArboNET. WNV is underreported, likely due to the number of subclinical and mild infections with non-specific symptoms, as well the low rate of laboratory testing of potential cases by clinicians. However, cases of neuroinvasive West Nile Virus (NWNV) are thought to be nearly 100% reported (DeGroot et al. 2014).

WNV prevention efforts consist of mosquito control at both the community and personal level, such as mosquito abatement, and the use of insect repellant, long sleeved tops, and long pants. Households can drain water out of unintentional water containers including buckets and spare tires.

D. West Nile Virus in Illinois

In September of 2001 American crows were the first birds in Illinois that tested positive for WNV, but the first human cases were not reported until 2002. That year nearly every county in Illinois reported at least one case of human WNV infection. By the end of 2002 Illinois had reported 884 cases; this was the highest number of cases of any state in the US for 2002 and the largest number of cases in a given year for Illinois to date (IDPH, 2016b). Since the 2002 emergence of WNV in Illinois new human WNV cases in

Illinois are reported annually, but spatial and temporal patterns vary. Most cases occur in the late summer months from July to September with the largest number of cases in August. In Illinois the primary vector of concern is *Culex pipiens*, but *Culex tarsalis* are thought to play a significant role in WNV transmission in areas along the Mississippi river. *Culex pipiens* have short life spans, about 12 days, and generally do not fly more than two miles from their nest. The most common WNV reservoirs in Illinois are crows, grackles, starlings, robins, cardinals, sparrows, and finches.

E. Land Cover

Culex mosquitoes have been shown to favor certain landscapes, particularly areas where stagnant water is common. The association between land cover and human WNV incidence may be regionally specific. In Cook County, IL WNV incidence in mosquitoes, birds, and horses was directly associated with water and grass area, and inversely associated with urban developed area (Liu et al. 2008). Studying the 2002 Northeast Illinois WNV outbreak Ruiz et al. found an association with physiographic region and WNV incidence in humans; people living in the Chicago Lake Plain region had higher incidence rates than people living outside of the region (Ruiz et al. 2004). Croplands with irrigation systems were associated with an increased risk of WNV in a study conducted in north-central Colorado (Eisen et al. 2010). Areas near wetlands and forested areas were associated with a higher incidence rate of mosquito WNV infections in South Dakota, and higher elevation was associated with a lower incidence rates (Chuang & Wimberly 2012).

F. Weather

Temperature and precipitation can have an impact on both the vectors and hosts for WNV. Laboratory experiments measured the EIP of WNV in several species of mosquitoes including *Culex pipiens* and *Culex tarsalis.* The EIP decreased with increasing temperatures in both species and no viral transmission was observed below 14.3°C (Reisen et al. 2006). In the cooler fall temperatures *Culex pipiens* are more likely to feed on plant sugars than blood in preparation of hibernation. Certain weather conditions are ideal to see an increase the population density of mosquitoes, and abundance is expected to be associated with increased WNV risk. Roiz et al. looked at the influence of weather on the abundance of mosquitoes for several species in different areas of Mediterranean wetlands. *Culex pipiens* abundance was found to be positively associated with winter rainfall (Roiz et al. 2014). Extreme high temperatures may reduce mosquito activity and result in lower rates of WNV transmission to humans. Drought induced amplification may occur when extreme dry weather limits water sources for mosquitoes and birds, and results in closer contact of mosquito habitats, birds, and humans. Drought induced amplification of mosquito WNV infection rates has been documented in New Jersey, and amplification of bird and human infection rates in Florida (Johnson et al. 2013, Shaman et al. 2010).

G. Climate Change and West Nile Virus

Climate change is expected to alter weather patterns which can impact the incidence of West Nile Virus via vector, reservoir, and viral evolution pathways. Chen et al. used predictive modeling techniques in several moderate to severe climate change scenarios to predict the effect of climate change on spatial and temporal patterns of WNV incidence in prairies of Alberta, Manitoba, and Saskatchewan, Canada. The authors concluded that by the 2050s there would be a northward expansion of available habitats for *Culex tarsalis* and a lengthened transmission season (Chen et al. 2013). The estimated influence of climate change on vector populations may not be uniform across all regions. Projected temporal and spatial patterns of *Culex quinquefasciatus* populations are different for Southwest US compared to Southeast US due to general regional climate differences; the impacts of climate change may take longer to become noticeable in the hot and dry climate of the southwest states (Morin et al. 2013). The impact of climate change has potential to alter the evolution of the virus; shorter viral seasons may lead to slower evolution of the virus and longer viral seasons may speed the process up (Bertolotti et al. 2007). Bird migration patterns are also likely to be altered as a result of climate change, and as a WNV reservoir this will affect WNV transmission rates. It could also lengthen the season for WNV transmission by lengthening the season for mosquito abundance and delaying bird migration (Paz et al. 2015).

H. Urbanization

Population density of Culex mosquitoes is associated with population density of humans. In some studies, urbanization was a significant predictor of human and mosquito WNV infections. Medlock & Vaux (2014) recently described how urban wetland development in the United Kingdom (UK), a sustainable strategy for drainage, has increased the presence of mosquito populations. An examination of the 2002 Northeastern Illinois outbreak of WNV found spatial clustering of census tracts with high incidence rates of WNV. A variety of sociodemographic factors were shown to have significant associations with census tracts that had at least one WNV case including age, race, income, housing age, and mosquito abatement district (Ruiz et al. 2004). Another study supported these results in their findings that population over 65, census tract below poverty line, and total household income were directly associated with WNV cases in Cook County, IL (Liu et al. 2008). The dynamics of WNV transmission in an urban setting may be different than rural areas for several reasons. The higher population density puts humans in closer contact with both birds and mosquitoes; Culex mosquitoes do not fly for more than a few minutes at one time. Urban settings have micro-environments that are ideal for *Culex pipiens* breeding including: swimming pools, rain barrels, and catch basins. In addition, human activity such as cutting down grass in dry detention basins has recently been associated with increased risk of human WNV infections (Mackay et al. 2016).

The interaction of urbanization and weather in relation to the rates of WNV infections is not known. Increased winter temperatures were associated with an increase in survival of overwintering mosquitoes in California (Nelms et al. 2013). Urban areas have more potential overwintering habitats compared to rural areas, including underground sewer systems. In some regions increased precipitation has been shown to be associated with an increase of mosquito WNV infection (DeGroote et al. 2014, Platonov et al. 2014). Urban areas have more impervious surfaces than rural areas and may have a higher amount of standing water after heavy precipitation, this can change the population dynamics of mosquitoes. This association may not be as strong in urban areas; Culex mosquitoes prefer habitats with stagnant water which are readily accessible in urban areas with bird baths, rain barrels, and flower pots. Urban areas provide Culex mosquitoes an abundance of these ideal microenvironments. Urban areas also maintain the close proximity of birds, Culex mosquitos, and humans which is an essential component of the WNV transmission cycle.

I. Geographically Specific

Some studies have developed models to predict rates or cases of WNV in a specific geographic area. These models appear to be region specific. In Russia an increase in average spring and summer temperature are expected to lead to increased incidence, a decrease in mean temperature in the winter months led to decreased incidence. Precipitation had no significant effect and the authors thought this might have been due to an abundance of natural lakes, rivers, and marshlands (Platonov et al. 2014). In contrast, a study in South Africa by Uejio et al. found an increase in rates of WNV infection associated with increased precipitation (Uejio et al. 2012). Rosà et al. described an increased mean winter temperature that lead to an earlier start and an increase in the length of mosquito season in Northern Italy (Rosà et al. 2014). Shaman et al. (2010) observed that a wet spring followed by a hot dry summer is associated with a higher incidence human WNV cases in the eastern plains of Colorado; in the western mountains of Colorado, a dry spring and dry summer was a better predictor of human WNV cases, although this was a weaker correlation. Young & Jensen used a machine learning program that built numerous models predicting WNV risk for the whole US based on weather and land cover and other data using remote sensor technology; they concluded that a single model with covariates of precipitation and deciduous forest area could be appropriate for the whole US, but with a caveat that regional biases existed (Young & Jensen 2013). Another study split the US into eight regions and identified various landscape, demographic, and climate variables. Different types of weather were found to predict WNV rates in the different regions; in the North Central Region that includes Illinois the average minimum temperature from November to February was significantly directly associated to having at least one human case of WNV, and precipitation was not significant. In contrast, in the Rocky Mountain region average minimum temperature in February, average minimum temperature in May, and precipitation anomalies in April and May were significantly associated with having at least one human WNV case (DeGroot et al. 2014).

J. Research Hypothesis

Previous research discussed earlier shows evidence for associations between mosquito WNV infection rates, temperature, precipitation, and land cover. How strongly these associations translate to human WNV infections and the association between population density and human WNV infections is less clear. Additionally, there are differences in risk factors and reporting biases for WNV and NWNV. Current literature on human WNV predominantly combines all types of WNV and NWNV in one group for data analysis.

In general this study tests the hypothesis that an increase of NWNV rates will be observed in years with warmer temperatures, increased total precipitation, and in counties with a high percent of land cover that is associated with a high population density of Culex mosquitoes. Nearly all NWNV cases are reported but only a small percent of other WNV cases are; including only NWNV cases will reduce the error due to underreporting, which may be a systematic error, random error, or both. The objective of this study is to explore the relationship between urbanization, weather, land cover, and annual county incidence rates of human NWNV infections in Illinois. A conceptual model shown as Figure 1 conveys the fundamental idea that was considered in developing these hypotheses.

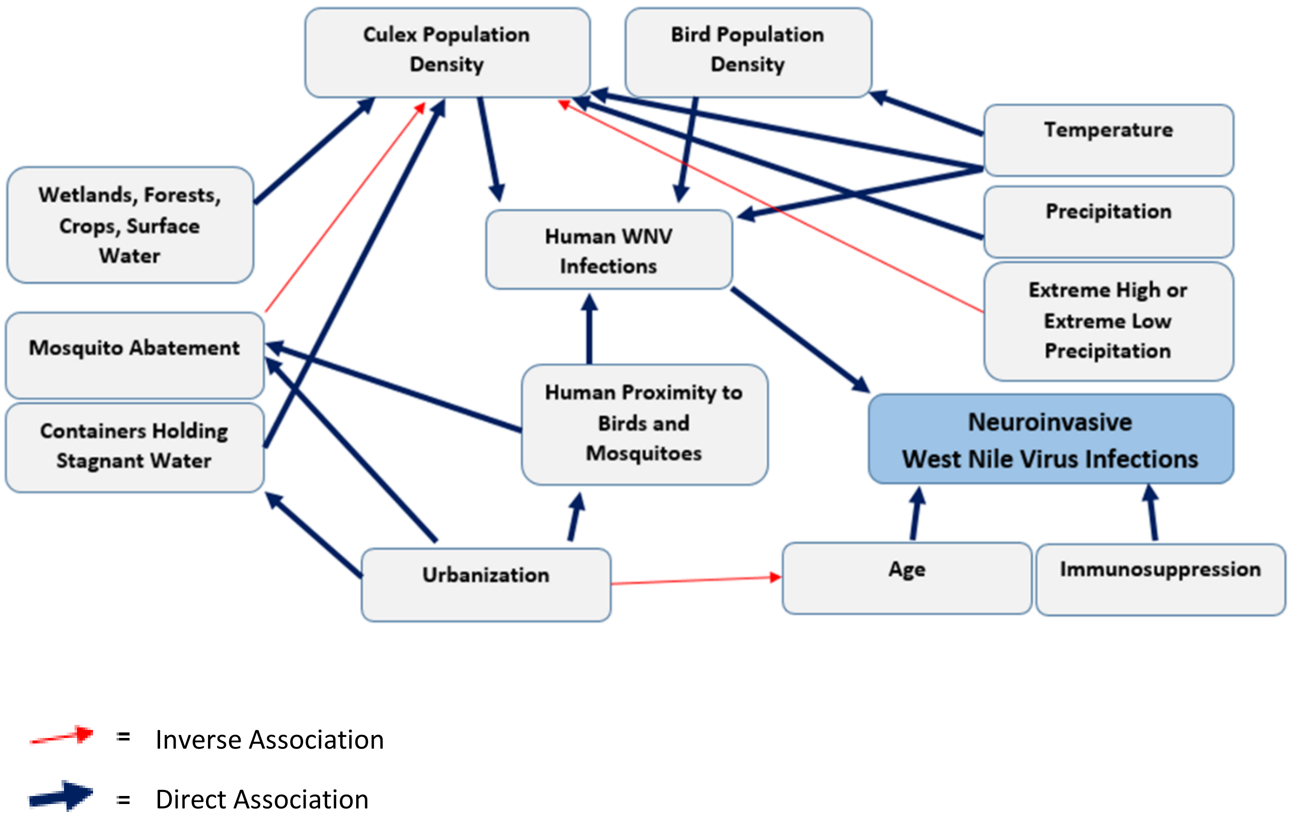
Hypothesis One: There is a direct positive association between higher NWNV rates and counties with a higher percent area of: wetlands, forest, developed land, or surface water.

Hypothesis Two: Warmer average temperatures in winter, spring, and summer will be associated with increased annual NWNV incidence rates.

Hypothesis Three: Higher levels of total precipitation will be associated with increased rates of NWNV, except for extreme precipitation conditions which will have an inverse association.

Hypothesis Four: Urban and rural areas will have different significant predictors for NWNV rates, possibly due to differences in human activity and the built environment. Temperature and precipitation will have a stronger significant measure of association in rural counties compared to urban counties. Wetland, forest, developed land, and surface water will have a stronger significant measure of association in urban counties compared to rural counties.

Figure 1. Conceptual model for the relationship between urbanization, land cover, weather, and neuroinvasive West Nile Virus infection rates



CHAPTER II. METHODS

A. Study Area

The study area includes all 102 counties in Illinois, the time period is 2002-2013. Data for all variables were aggregated to the county level.

B. Data Sources

The WNV dataset was obtained from Illinois Department of Public Health (IDPH) WNV Surveillance for years 2002-2013. The data obtained was de-identified by IDPH. This analysis was determined not to be human subject research by University of Illinois Institutional Review Board. Information collected during the years 2002-2003 was slightly different from data collected from 2004-2013 and were acquired in separate files. The data elements included in each dataset are summarized in Table I.

County level population data were obtained from the 2010 US Census for Illinois. Urbanization was evaluated using two standard classifications: Core Based Statistical Areas (CBSA) and Rural-Urban Continuum Code (RUCC). CBSAs are defined by the US Office of Management and Budget. Counties that have a core urban population of at least 50,000 with adjacent counties that are economically integrated with the core urban area are classified as being part of a metropolitan statistical area. Counties economically integrated with an urban core population of 10,000 to less than 50,000 are classified as being part of a micropolitan statistical area. Counties not part of a metropolitan or micropolitan statistical area are classified as rural. RUCC's are classified by the United States Department of Agriculture (USDA) and based on CBSA. In this classification method metropolitan statistical areas are divided into three groups based on population size. Counties not included in a metropolitan statistical area are divided into six groups based on population size and adjacency to a metropolitan area. County classification of CBSA was obtained from the US Census website

TABLE I. VARIABLES IN THE WEST NILE VIRUS SURVEILLANCE DATASETS FROM ILLINOIS DEPARTMENT OF PUBLIC HEALTH; COMPARISON OF 2002-2003 AND 2004-2013 DATASETS

Surveillance Dataset Variable Names		
2002-2003 Data	**2004-2013 Data**	**Description**
Neuroinvasive	Neuroinvasive	Yes or No
Gender	Gender	Male or Female
Age Range	Age Range	10 Year Increments
Race	Race	American Indian or Alaskan Native, Asian, Black or African American, Other, or White
Ethnicity	Ethnicity	Hispanic or Latino, Not Hispanic and not Latino
Deceased	Deceased	Yes or No
Case Status	Case Status	Probable or Confirmed
	Disease	WNV or WNF (West Nile Virus or West Nile Fever)
	Patient Seen in ER	Yes or No
	Admitted to Hospital	Yes or No
	Human Lab Tests Conducted	Yes or No
	Serogroup	Asymptomatic, Meningitis, Encephalitis, Acute Flaccid Paralysis, or Other

(https://www.census.gov/geo/maps-data/maps/statecbsa.html), and county Rural-Urban codes were obtained from the USDA website (http://www.ers.usda.gov/data-products/rural-urban-continuum-codes.aspx).

Weather data was obtained from the Midwest Regional Climate Center (MRCC) and the National Oceanic and Atmospheric Administration (NOAA). The majority of weather data was obtained through the Building Resilience Against Climate Effects (BRACE) project Dr. Jyotsna Jagai who compiled weather data from MRCC to obtain monthly averages for temperature and precipitation. Temperature and precipitation data were not available for four counties for any year in this study (Alexander, DuPage, Effingham, McDonough, and Williamson Counties). The remaining 98 counties reported temperature and/or precipitation for at least some months from January 2002 to December 2013. Due to missing values additional monthly and daily temperature data was obtained from both NOAA and MRCC for the four counties for which temperature and precipitation data were missing. Data for county monthly average temperature was requested from NOAA through their Climate Data Online website, and daily average temperature was available for immediate download on the MRCC website (https://www.ncdc.noaa.gov/cdo-web/, http://mrcc.isws.illinois.edu/CLIMATE/). The following variables were obtained in these datasets: minimum daily temperature, standard deviation of minimum daily temperature, maximum daily temperature, standard deviation of maximum daily temperature, mean daily temperature, standard deviation of mean daily temperature, and total precipitation. Mean daily temperature and total precipitation was aggregated to both the month and season for further analysis. The months of December, January, and February were combined for winter temperature averages and total precipitation. The months of March, April, and May were combined for spring averages and totals. The months of June, July, August were combined for summer averages and totals. The months of September, October, and November were combined for fall temperature averages and total precipitation.

Between 26.3% and 27.5% of monthly county temperature data and 9.6% of monthly county precipitation data were missing. Temperature data from 2002-2013 for 11 Illinois counties were not available, and several counties had greater than ten percent missing for average monthly temperature. A list of counties with missing temperature and precipitation is provided in Table II. Temperature was imputed as the average of the surrounding counties. If bordering counties had missing temperature, the average temperature for three bordering counties was taken from the following: one northern bordering county, one southern bordering county, and one eastern or western bordering county. Mason County was one exception where the average of two bordering counties was used, one bordering on the west and the other borders on the east. A variable was created to identify imputed versus reported temperatures. The accuracy of this imputation method was tested by randomly selecting 20 counties without any missing temperature data. The same imputation methods were followed for these 20 counties and a percent difference of imputed to actual temperatures was calculated. This analysis showed that for all key temperature variables imputation provided accurate measures. Unlike temperature, precipitation is highly variable on a local level in Illinois, and therefore missing precipitation data were not imputed.

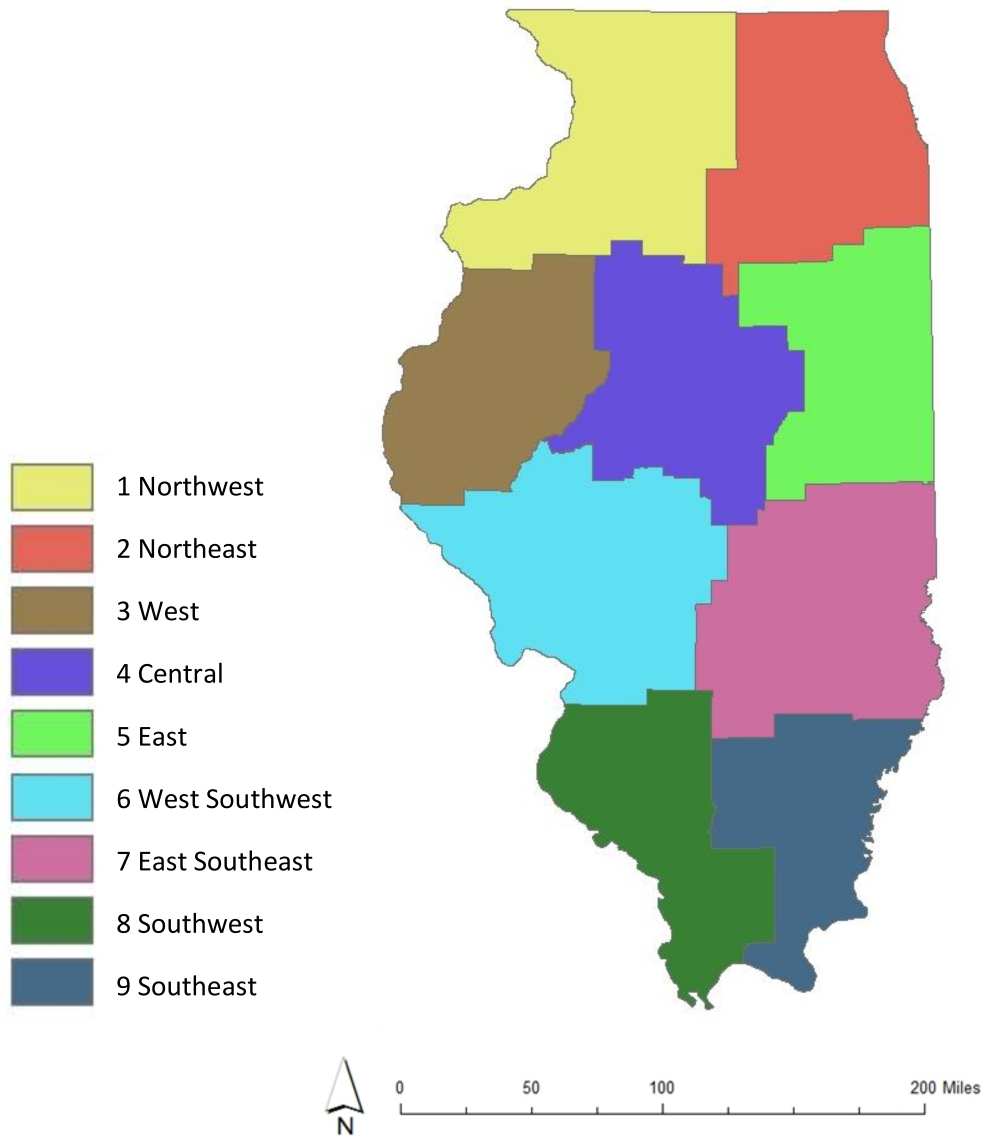
To simplify the results for weather the univariate statistics for temperature and precipitation were summarized and presented by climate divisions. There are nine defined climate divisions in Illinois pictured in Figure 2, climate division data was obtained from NOAA (http://www.esrl.noaa.gov/psd/data/usclimdivs/boundaries.html). Temperature and precipitation data for individual counties was used for bivariate and multivariable regression data analysis.

Land cover and elevation was downloaded from Illinois Geospatial Data Clearinghouse website (https://clearinghouse.isgs.illinois.edu/data/land-cover/usda-nass-cropland-data-layer-illinois-2007, https://clearinghouse.isgs.illinois.edu/data/elevation/surface-elevation-301-foot-digital-elevation-model-dem). Data for land cover and elevation came in the form of a raster file that was uploaded into ArcGIS 10.2.2 and used to obtain fractions of county area for a given land cover classification. Land cover was grouped by the classification codes defined by United States Geological Survey (USGS) and described in the National Land Cover Database (NLCD). ArcGIS 10.2.2.3552 was used to process land cover and elevation data into county level variables that were used for the analysis. Land cover was categorized by croplands, open water, developed open space, low intensity developed, medium intensity developed, high intensity developed, barren land, deciduous forest, herbaceous grasslands, herbaceous wetlands, and woody wetlands. Values for each land cover variable represented the

TABLE II. COUNTIES IN ILLINOIS WIHT MISSING TEMPERATURE AND PRECIPITATION DATA FROM 2002-2013

Temperature	
Missing All	**Missing >10%**
Brown	Fulton
Cass	Hamilton
Clark	Marshall
Cumberland	Menard
DeWitt	Monroe
Gallatin	Putnam
Mason	Saline
McDonough	St. Clair
Piatt	Wabash
Tazewell	
Williamson	
Precipitation	
Missing All	**Missing Few (less than 5%)**
Calhoun	DuPage
Hamilton	Monroe
Johnson	Piatt
Kendall	Saline
	Stark

Figure 2. Map of the nine climate divisions in Illinois



GCS: North American 1983
PCS: Illinois State Plane NAD 1983

percent of county land area covered by each land cover category. Developed land is defined as a mixture of vegetation and constructed materials and further classified by percent impervious surface. Open space developed, low intensity developed, medium intensity developed, and high intensity developed land consists of less than 20%, 20% to 49%, 50% to 79%, or 80% and above impervious surfaces, respectively. Total percent developed land was calculated by obtaining the sum of high intensity developed, medium intensity developed, low intensity developed, and open space developed land. The USGS groups several of these specific land cover types into a broader category and those broader categories were used as well. Total percent wetland was calculated by obtaining the sum of herbaceous wetlands and wooded wetlands. Total forest was calculated by obtaining the sum of deciduous forest and evergreen forest. County level elevation included highest elevation in feet, lowest elevation in feet, and elevation range in feet.

C. Statistical Analysis

Statistical analysis was performed with SAS 9.4. ArcGIS 10.2.2.3552 for visualization of land cover, temperature, precipitation, and NWNV rates. Distribution of characteristics of NWNV cases was summarized by age range, gender, race, ethnicity, and status. Missing values for 'status' were assumed to be alive. Cases were aggregated by county merged with US Census data in order to calculate rates of NWNV for each county. Rates were calculated as the number of NWNV cases divided by the total county population and expressed as per 100,000 people. A 12-year cumulative incidence rate for NWNV was calculated as well as yearly incidence rates for each county.

D. Univariate

Frequency counts and percentages were calculated for any categorical variables including gender, age, ethnicity, race, case status, if the patient was deceased, RUCC, and CBSA. Univariate statistics were obtained for continuous temperature, land cover, and census variables including mean, median, standard deviation, skewness, kurtosis, and missing data. A Shapiro-Wilk p-value of greater than 0.05 was used to define normality for continuous variables. Continuous variables that were not normally distributed were evaluated for normality after a natural log transformation. Continuous variables including NWNV incidence rates were additionally analyzed as dichotomous variables with cut points at the mean, median, one standard deviation above the mean or median, or one standard deviation below the mean or median. Temperature and precipitation were dichotomized in several variables with various cut points. See Tables XIX-XXI, Appendix A for a summary univariate statistics for each variable.

Variable name, type, and distribution is described in Table III. Most of the land cover variables followed a lognormal distribution. Precipitation was highly right skewed but was not lognormal, and county average temperature was approximately normally distributed.

E. <u>Temporal and Spatial Considerations</u>

Land cover assumed to be static throughout the time period of this study. For bivariate analysis with land cover the 12 year NWNV incidence rates for counties in Illinois were calculated and compared to the static data for land cover. Annual county NWNV incidence rates were compared to average daily temperature measurements and total precipitation for each season.

A. <u>Bivariate</u>

Scatterplots were used to evaluate the relationship between NWNV rates and land cover variables, and also between NWNV rates and weather variables. Scatterplots were created with the log of annual county NWNV incidence rates on the y axis versus all continuous variables for temperature and precipitation. Scatterplots were generated for the log of 12 year NWNV incidence rates versus population, rates, weather, land cover, and elevation variables. Visualizing the relationship between

NWNV rates and the variables in a scatterplot did not indicate a clear threshold nor did it show a distinct linear or quadratic relationship. Two bivariate analyses were conducted: simple linear regression with a log transformed outcome, and 2 X 2 contingency tables with two dichotomous variables. Caution was used interpreting p-values in the bivariate associations; there were over twenty bivariate measures of association for the same dependent variable, and it is possible that associations could be statistically significant at 0.05 due to chance alone.

Simple linear regression was performed using the natural log transformed 12 year NWNV incidence rates with each non-transformed continuous land cover variable. Simple linear regression was also performed using the natural log transformed annual NWNV incidence rates and each continuous weather variable. A quadratic relationship was assessed with mean seasonal temperature squared and mean precipitation squared. The β estimate, standard error, p-value, and r^2 were recorded for each association.

A bivariate analysis using two dichotomous variables was also performed; 2 by 2 tables were created to compare counties with high 12-year incidence rates of NWNV to counties with low 12-year incidence rates of NWNV with variables described in land cover and elevation. Odds ratios and Pearson χ^2 p-values were recorded for each bivariate association.

Weather variables were dichotomized and compared to a dichotomous annual county NWNV incidence rate. County weather data was analyzed for an association with annual county NWNV incidence rate based on both monthly data and seasonal categories for average mean daily temperature, mean daily minimum temperature, mean maximum temperature, and total precipitation. Winter weather was defined as temperature and precipitation in the months of December, January, and February. Spring

TABLE III. VARIABLE NAME, TYPE, AND DISTRIBUTION FOR WEST NILE VIRUS SURVEILLANCE, LAND COVER, AND WEATHER DATASETS FOR ILLINOIS 2002-2013

Variable Description	Variable Type	Distribution
Surveillance Dataset		
Neuroinvasive	Dichotomous	
Age Range	Ordinal, 9 Categories	
Ethnicity	Nominal, 2 Categories	
Case Status	Probable or Confirmed	
Gender	Nominal, 2 Categories	
Race	Nominal, 7 Categories	
Land Cover Dataset		
Fraction Cropland	Proportion	Left Skewed
Fraction Non-Crop Agriculture	Proportion	Lognormal
Fraction Open Water	Proportion	Lognormal
Fraction Developed Open Space	Proportion	Lognormal
Fraction Low Intensity Developed	Proportion	Lognormal
Fraction Medium Intensity Developed	Proportion	Lognormal
Fraction High Intensity Developed	Proportion	Lognormal
Developed	Proportion	Lognormal
Fraction Barren Land	Proportion	Lognormal
Fraction Deciduous Forest	Proportion	Lognormal
Fraction Evergreen Forest	Proportion	Lognormal
Fraction Herbaceous Grassland	Proportion	Lognormal
Fraction Wooded Wetlands	Proportion	Lognormal
Fractions Herbaceous Wetlands	Proportion	Lognormal
Highest Elevation in Feet	Continuous	Lognormal
Elevation Rang in Feet	Continuous	Lognormal
Lowest Elevation in Feet	Continuous	Lognormal
Area in Square Miles	Continuous	Lognormal
Total Population	Continuous	Left Skewed
Total Number of People Over 50	Continuous	Lognormal
Percent of People Over 50	Proportion	Left Skewed
Population Density	Proportion	Lognormal
Weather Dataset		
Mean Maximum Temperature	Continuous	approximately normal
Mean Minimum Temperature	Continuous	approximately normal
Meant Temperature	Continuous	approximately normal
Total Precipitation	Continuous	Lognormal

weather was defined as temperature and precipitation in the months March, April, and May. Summer weather was defined as temperature and precipitation in the months June, July, and August. Fall weather was defined as temperature and precipitation in the months September, October, and November. The average for mean daily temperature were approximately normal, but total precipitation was highly skewed to the right. Each average monthly minimum, maximum, and mean temperature variable was dichotomized three different ways; one with a cut point at the mean, one with a cut point one standard deviation below the mean, and one with a cut point one standard deviation above the mean. Because precipitation was highly skewed, the cut point was based on the median total precipitation instead of the mean values. Each monthly total precipitation variable was dichotomized three different ways; one with a cut point at the median, one with a cut point 0.5 standard deviation below the median, and one with a cut point one standard deviation above the median. Contingency tables with two dichotomous variables were created comparing high monthly average temperature to low monthly average temperature and precipitations compared to the NWNV rate for the year. In addition, 2 by 2 contingency tables were created comparing high seasonal average temperature to low seasonal average temperature and precipitations compared to the NWNV rate for the year.

Several types of variables representing urbanization were assessed to understand which variable may, this included CBSA, RUCC, population density, and percent developed land. A bivariate analysis was conducted with 12 year NWNV incidence rates and each urbanization variable. Population density and percent developed land are both continuous variables; each were analyzed using bivariate analysis simple linear regression was conducted with natural log transformed 12 year NWNV incidence rates as the outcome. CBSA and RUCC are categorical variables, the bivariate analysis consisted of contingency table with a test for trend.

Most cases occurred in 2002 and in Cook County. To understand the influence this has on the measure of association, the bivariate analysis was repeated without cases from the year 2002, and then again without cases from Cook County for any year. Collinearity was assessed in a Spearman correlation matrix and obtaining variance inflation factor in a linear model.

B. Modeling Approach

There are no distinct cut-off points for percent land cover, temperature, or precipitation in the current literature with relation to NWNV rates, therefore these variables were analyzed as continuous variables without transformation in multivariable modeling. There were several statistical difficulties taken into consideration before beginning multivariable model building including, zero-inflation, over-dispersion, serial dependence, and multi-collinearity.

Over 80% of the annual county NWNV incidence rates were equal to zero indicating the data was zero-inflated. An ordinary regression model does not differentiate between the predictors of occurrence of an event (a county having a non-zero number of NWNV cases) and the predictors of the value of an event (the number of cases of NWNV in a county where one or more cases occurred). However, often these are two discrete associations, with different predictors associated with the presence vs. the absence of an event occurring and the number of occurrences. Additionally, annual county NWNV incidence rate had a mean of 0.519 per 100,000 and a standard deviation of 1.97, indicating the data is over-dispersed. Serial autocorrelation was measured with the Durbin-Watson statistic calculated at 2.03; this is above the critical value of 1.69 and indicates that there is no significant serial autocorrelation with annual rates of NWNV. Because of the diagnostic information described above, a zero-inflated negative binomial (ZINB) regression model was used. ZINB regression creates two models, one predicting rates or counts greater than zero and another predicting zero rates or counts. Each model can have different covariates.

Model building began by first identifying potential land cover, weather, and urbanization predictors of NWNV rates for the state of Illinois as a whole, and then stratified by urban/suburban areas and rural areas. Statistical evaluation of covariates, as well as a priori knowledge, was used to determine inclusion of covariates in the final model. There are nearly twenty potential covariates based on the conceptual model in Figure 1. Several were partially collinear which would result in unstable estimates with a large standard error if those variables were included in the same model. A backward selection process has a one in twenty chance of producing spurious significant relationships when alpha is set at 0.05. To prevent this and to assess for collinearity one variable at a time, a stepwise model building approach was used. The year 2002 was controlled for based on a priori knowledge that this was an outbreak year with the first recorded human WNV cases, and there may be factors that contributed to the first outbreak differently than subsequent years. After identifying significant associations, confounding was assessed based on a ten percent change in the β-coefficient. Three sensitivity analyses were conducted; one excluded counties with imputed temperature, one excluded cases from the year 2002, and one excluded cases from Cook County, the most populous county in the State, which accounts for a majority of NWNV cases. A two-sided p-value less than 0.05 was considered statistically significant. No evidence of multicollinearity among the final independent variables was indicated (based on evaluation of standard errors, and evaluation of variance of inflation and tolerance tests).

CHAPTER III. RESULTS

The distribution of characteristics for NWNV surveillance data is reported in Table IV. The majority of NWNV cases is in the age group ≥50 years, and gender distribution of NWNV cases was nearly equal with 49.4% female and 50.6% male. Race and ethnicity had a substantial amount of missing values which may have contributed to the differences in distribution compared to the overall distribution in Illinois. Missing values for patient status were assumed to be alive, 10.9% of NWNV cases in Illinois from 2002-2013 were fatal.

The variation by year in NWNV case count can be seen in Figure 2 as graph of total annual NWNV case count for Illinois from 2002-2013. In 2002, there were the most recorded cases of 2002 NWNV (N=553); this was also the first year with any recorded cases of human WNV in Illinois. From 2003 to 2013 the number of human NWNV cases has fluctuated with 2012 having the second highest case count of 187. Years 2005 and 2012 had a considerable increase in the number of reported cases compared to the previous year, and 2009 had the fewest recorded cases of NWNV in this time frame.

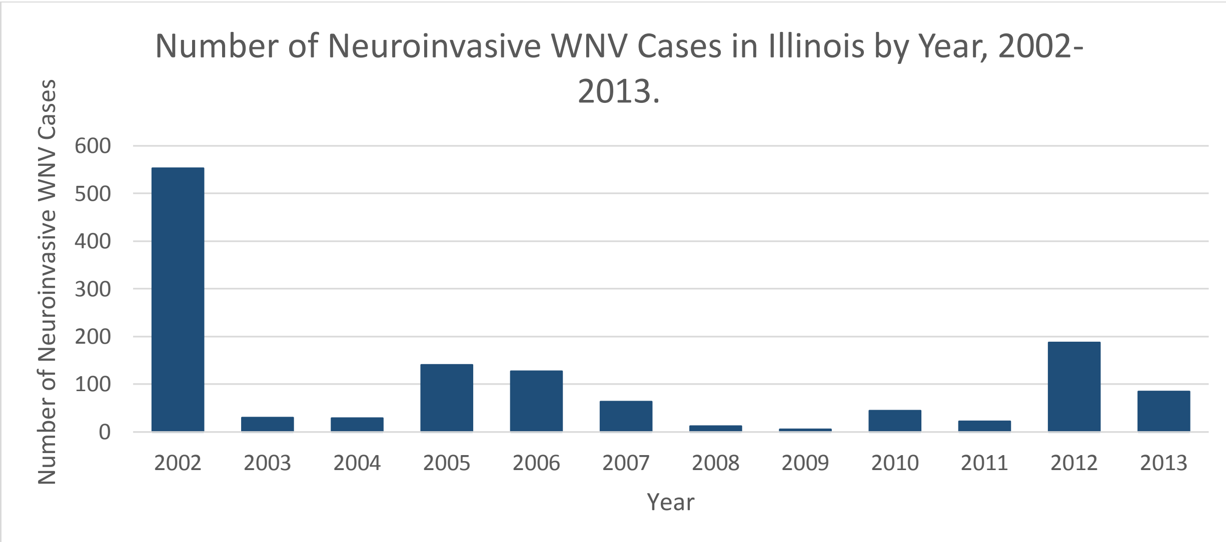
Counties in northeast Illinois generally have at least one reported case of NWNV annually, but the rates are not necessarily higher in comparison to other counties in Illinois. Figure 3 shows spatial and temporal variability of county NWNV incidence rates. In a given year there is variability of rates among counties in Illinois; some counties have higher rates while other counties have low rates. But most counties do not consistently have high annual NWNV incidence rates.

The 12-year cumulative incidence of NWNV for all of Illinois from 2002-2013 was 5.4 per 100,000. Rates in rural counties may be inflated compared to counties with a larger population due to having a smaller denominator when calculating county incidence rates. Table V shows the 12-year cumulative incidence of NWNV by CBSA; the average and median rates of counties in a CBSA is provided as well as average rates from aggregated data for all counties in a CBSA. Counties grouped as not classified in a

TABLE IV. DISTRIBUTION OF CHARACTERISTIC FOR PATIENTS WITH NEUROINVASIVE WEST NILE VIRUS INFECTIONS FROM ILLINNOIS SURVEILLANCE 2002-2013 COMPARED TO DISTRIBUTION OF CHARACTERISTICS OF ILLINOIS POPULATION FROM THE 2010 UNITED STATES CENSUS

	Neuroinvasive West Nile Virus Surveillance % (n)	**Total Illinois Population (2010 Census)** %
Age		
<20 Years Old	3.9 % (51)	27.30%
20-49 Years Old	28.8% (375)	41.50%
>50 Years Old	67.3% (875)	31.30%
Gender		
Female	49.4% (643)	51.00%
Male	50.6% (658)	49.00%
Race*		
Asian	1.4% (18)	5.20%
Black or African American	10.4% (135)	15.40%
White	76.5% (995)	73.40%
Other	5.0% (65)	3.50%
Missing	6.7% (88)	
Ethnicity* +		
Hispanic or Latino	6.1% (44)	21.50%
Not Hispanic or Latino	64.0% (459)	78.50%
Missing or Unknown	29.8% (214)	
Status*		
Deceased	10.9% (134)	
Alive or Missing	90.8% (1090)	
* >5% missing or unknown values		

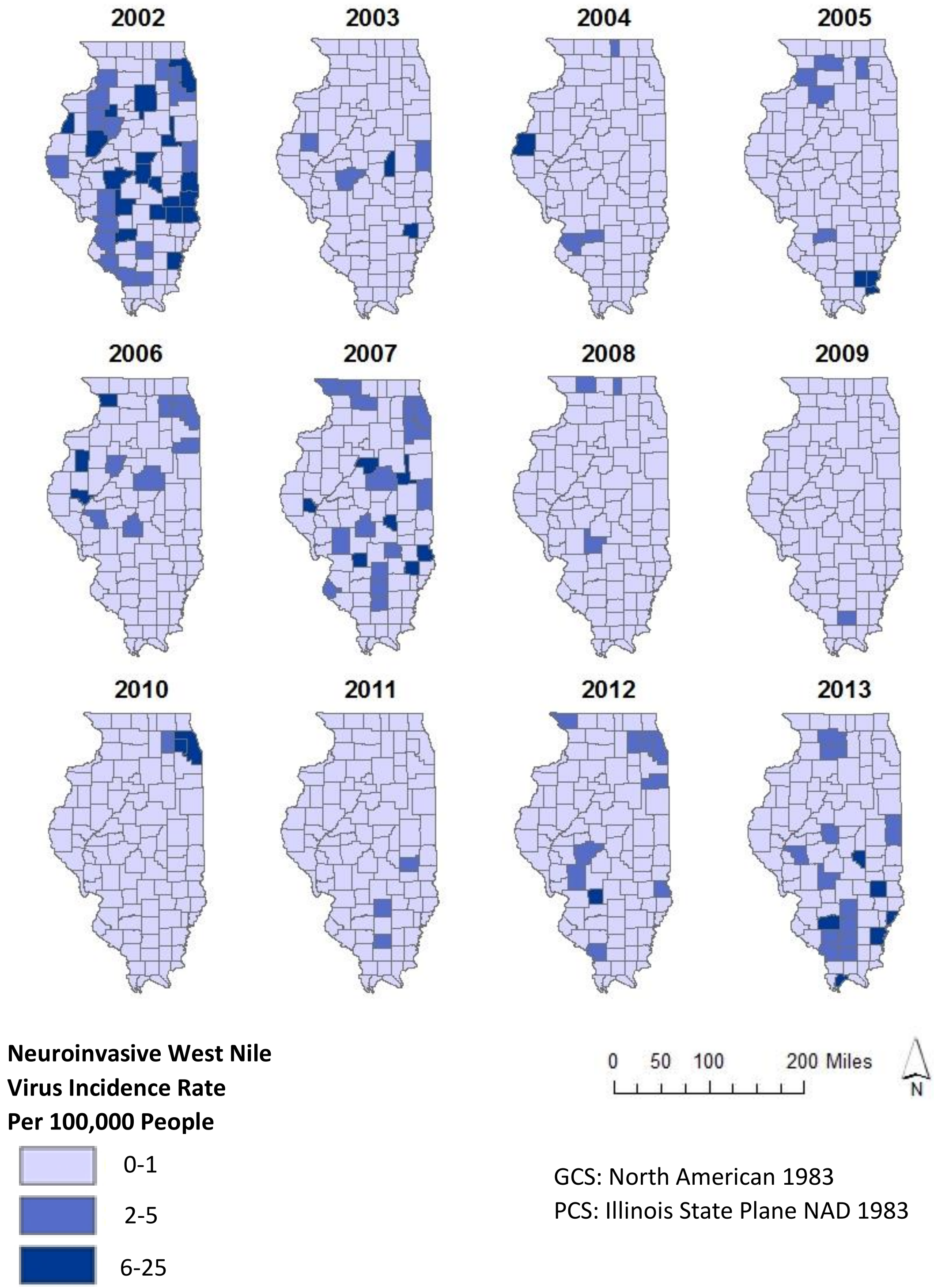
Figure 3. Number of NWNV Cases by Year for 2002-2013 in Illinois, from Illinois Department of Public Health West Nile Virus Surveillance



CBSA had the highest average county rates of NWNV (7.4 per 100,000), followed by counties in a metropolitan statistical area (6.2 per 100,000). In contrast, aggregated totals showed rates of NWNV were highest in metropolitan statistical areas, followed by counties not classified in a CBSA. Metropolitan counties had the lowest percent population over 50 with an average of 33.4%; rural counties had the highest percent population over 50, average 39.4%.

Average county percent for developed open space, barren land, low intensity developed, medium intensity developed, high intensity developed, open water, and non-crop agriculture were highest in counties in a metropolitan statistical area; there is a decreasing trend from urban to rural areas, this trend can be seen in Table VI. Average county percent for deciduous forest, evergreen forest, herbaceous grassland, and herbaceous wetland were lowest for counties in a metropolitan statistical area, with an increasing trend from urban to rural areas. Mean county percent of wooded wetlands

Figure 4. Maps of Neuroinvasive West Nile Virus incidence rate by year for Illinois counties, from 2002-2013



were highest in rural counties (1.3%), with counties in a metropolitan statistical area second (1.2%), percent wooded wetlands was lowest for counties in a micropolitan statistical area (1.0%). This is the only land cover variable that did not show an urban to rural trend.

The accuracy of the temperature imputation method was tested using temperature data from 20 counties. The same imputation methods were used and the results are reported in Table VII, Table VIII, and Table VIX which shows the actual and imputed temperatures for winter, spring, and fall as well as percent difference. All of the imputed temperatures were less than ten percent different from actual temperatures for all seasons, and 170 out of 180 imputations were less than five percent different. The average percent difference of all 20 counties for mean temperature and mean maximum temperature was less than one percent, mean minimum winter temperature has an average of 1.7% lower than the actual temperature.

Temperature distributions in both an individual climate division and for the whole state were distributed approximately normal, however, average total precipitation was always skewed to the right. Average county temperature is lowest in climate division 1, which is in the northern Illinois. Average county temperatures increase as the climate divisions move south. There is no North-South trend observed with precipitation, however, climate division 5 and 2 have the highest overall median total precipitation and climate division 6 has the lowest. The average mean monthly temp for all counties in a climate division is provided in Table X and the median total monthly precipitation is provided in Table XI. A map of the nine climate divisions in Illinois is pictured in Figure 2.

Figures 5-10 show the state average temperature and precipitation for a given season from 2002-2013 as well as the annual NWNV incidence rates for the state of Illinois. A crude association between temperature and NWNV rates can be seen in Figure 6 looking at the mean summer temperature with NWNV rates; other seasons and precipitation are not as straightforward. 2009, 2008, and 2011 had the

TABLE V. COUNTY CHARACTERISTICS AND RATES OF NEUROINVSIVE WEST NILE VIRUS FROM 2002-2013 ILLINOIIS WEST NILE VIRUS SURVEILLANCE AND 2010 UNITED STATES CENSUS, FOR WHOLE STATE AND BY CORE BASED STATISCICAL AREA

	All of Illinois		Counties in a Metropolitan Statistical Area		Counties in a Micropolitan Statistical Area		Counties Not Classified in a Core Statistical Area	
Regional 12-Year Cumulative Incidence of NWNV (per 100,000 people)	5.4		10.9		5.4		6.6	
Total Regional NWNV Cases	1301.0		1190.0		71.0		40.0	
Total Regional Population	12,830,632		10,907,952		1,314,261		608,419	
County	mean	median	mean	median	mean	median	mean	median
County NWNV Rate (per 100,000 people)	6.2	5.2	6.2	5.2	4.9	4.9	7.4	6
County NWNV Case Count	12.8	1	36.06	5	2.2	1.5	1.1	1
Population Density (People per Mile^2)	196.1	48.8	503.3	192.8	66.3	63.5	34.3	30.3
County Population	125790	27315	330544	113449	41070.7	36550	16443.8	16233
Percent of Population ≥50	36.8%	37.7%	33.4%	33.7%	37.4%	37.7%	39.2%	39.4%
County Area (Mile^2)	545	511	550.5	486	597.6	576.5	494.7	444

TABLE VI. MEAN AND MEDIAN PERCENT OF COUNTY AREA WITH LAND COVER CLASSIFICATION FOR ILLINOIS COUNTIES 2002-2013, FOR WHOLE STATE AND BY CORE BASED STATISTICAL AREA

	All of Illinois		Counties in a Metropolitan Statistical Area		Counties in a Micropolitan Statistical Area		Counties Not Classified in a Core Statistical Area	
Land cover	mean	median	mean	median	mean	median	mean	median
Percent Developed Open Space	6.7%	6.4%	7.8%	6.6%	6.2%	6.3%	6.2%	6.2%
Percent Barren	0.08%	0.04%	0.14%	0.07%	0.06%	0.04%	0.04%	0.07%
Percent Low Intensity Developed	4.4%	2.8%	8.3%	5.1%	3.2%	3.0%	2.0%	1.8%
Percent Medium Intensity Developed	1.10%	0.35%	0.0276	0.0103	0.00529	0.00434	0.0022	0.0021
Percent High Intensity Developed	0.41%	0.08%	1.07%	0.36%	0.15%	0.14%	0.05%	0.04%
Percent Open Water	1.9%	1.3%	2.2%	1.6%	1.8%	0.8%	1.7%	1.4%
Percent Cropland	51.0%	54.0%	51.6%	52.7%	55.0%	57.2%	47.8%	53.0%
Percent Non-Crop Agriculture	0.030%	0.005%	0.074%	0.007%	0.017%	0.007%	0.011%	0.003%
Percent Deciduous Forest	19.00%	17.00%	13.68%	11.06%	18.54%	17.25%	24.17%	20.13%
Percent Evergreen Forest	0.140%	0.001%	0.002%	0.0004%	0.070%	0.002%	0.312%	0.002%
Percent Herbaceous Grassland	13.0%	11.0%	10.8%	11.1%	13.2%	11.0%	15.6%	14.4%
Percent Woody Wetlands	1.2%	0.6%	1.2%	0.4%	1.0%	0.4%	1.3%	0.9%
Percent Herbaceous Wetlands	0.37%	0.15%	0.27%	0.09%	0.35%	0.14%	0.47%	0.18%
Highest Elevation (Feet)	801.5	805.5	842.8	830	810.9	798.5	756.5	740
Elevation Range (Feet)	316.6	290	308.4	310	323.1	300	318.32	260
Lowest Elevation (Feet)	484.9	462.5	534.4	530	487.8	480	438.2	421

lowest annual rates of NWNV for the state of Illinois; the same years also had 3 or more seasons with temperatures below average. 2012 was the second highest year for NWNV; in 2012 winter, spring, and summer all had above average seasonal temperatures compared to the 12 year average. 2002 was the first year WNV was found in the human population in Illinois, but infected birds and mosquitoes were found in Illinois in 2001. The year 2002 had above average seasonal temperatures for winter and summer, which is ideal for mosquito population growth and may have contributed to the outbreak. Years 2005 and 2012 had a considerable increase in case count compared to the previous year; both years had relatively warm winter, spring, and summer compared to the averages. 2009 had the fewest recorded cases of NWNV; this year had colder than average winter, spring, summer and summer. 2003, 2004, 2008, and 2011 also had a small amount of WNV cases; in these years at least 2 seasons between winter, spring, and summer were colder than average.

A. Bivariate Results

Scatterplots are provided in Figures 12-40, Appendix B. Many counties had zero rates that formed a distinct line near the bottom of the scatterplot. Scatterplots comparing the natural log of NWNV rates to each land cover variable did not show a clear linear or non-linear trend. No obvious trend was observed in scatterplots comparing the natural log of NWNV rates to weather.

The results of simple linear regression with the natural log of NWNV rates as an outcome for each land cover type is provided in Table XII. Percent low intensity developed land was significant. Open space developed, medium intensity developed, and high intensity developed land was near significant. The β-coefficient for all developed land showed a similar measure of effect and was grouped into one category called ‘Developed Land,’ this was significant. Herbaceous and wooded wetlands were significant individually and in the combined ‘Wetland’ group.

TABLE VII. RESULTS OF TEMPERATURE IMPUTATION METHOD VERIFICATION FOR MEAN WINTER TEMPERATURES 2002-2013

	Mean Minimum Temperature (F)			Mean Temperature (F)			Mean Maximum Temperature (F)		
	Actual	Imputed	Percent Difference	Actual	Imputed	Percent Difference	Actual	Imputed	Percent Difference
Carroll	15.00	14.52	3.2%	24.18	24.30	-0.5%	33.40	34.14	-2.2%
Clinton	23.87	24.07	-0.8%	32.06	32.27	-0.7%	40.34	40.57	-0.6%
DeKalb	16.00	16.89	-5.5%	23.71	25.08	-5.8%	31.36	33.25	-6.0%
Edwards	25.66	27.01	-5.3%	34.03	34.31	-0.8%	42.65	42.21	1.0%
Hancock	18.51	19.89	-7.5%	28.00	28.38	-1.4%	37.47	36.90	1.5%
Henry	17.04	17.66	-3.6%	25.42	25.87	-1.8%	33.78	34.13	-1.0%
Jackson	25.66	25.52	0.5%	35.24	34.66	1.6%	44.83	43.91	2.0%
Jasper	23.68	23.26	1.8%	32.07	32.13	-0.2%	40.56	41.00	-1.1%
Kankakee	18.89	18.80	0.4%	27.11	26.82	1.0%	35.34	34.84	1.4%
Lake	16.89	17.01	-0.7%	24.70	25.16	-1.9%	32.51	33.27	-2.3%
Logan	20.13	20.56	-2.2%	28.67	29.03	-1.2%	37.23	37.48	-0.7%
Marion	23.51	24.27	-3.2%	32.13	32.72	-1.8%	40.75	41.20	-1.1%
Massac	28.64	28.28	1.2%	37.19	36.86	0.9%	45.78	45.46	0.7%
Morgan	20.54	21.65	-5.4%	29.94	30.29	-1.2%	39.40	38.95	1.2%
Perry	26.66	25.20	5.5%	35.38	33.92	4.1%	44.12	42.66	3.3%
Pike	20.59	21.17	-2.8%	29.54	29.92	-1.3%	38.67	38.69	-0.1%
Putnam	17.94	18.43	-2.7%	26.71	26.54	0.7%	35.46	34.59	2.4%
Shelby	22.59	23.10	-2.3%	30.85	31.29	-1.5%	39.09	39.50	-1.1%
Union	26.07	26.19	-0.5%	36.04	36.20	-0.5%	46.34	46.57	-0.5%
Woodford	17.61	18.51	-5.1%	26.96	26.92	0.2%	36.34	35.32	2.8%

TABLE VIII. RESULTS OF TEMPERATURE IMPUTATION METHOD VERIFICATION FOR MEAN SPRING TEMPERATURES 2002-2013

	Mean Minimum Temperature (F)			Mean Temperature (F)			Mean Maximum Temperature (F)		
	Actual	Imputed	Percent Difference	Actual	Imputed	Percent Difference	Actual	Imputed	Percent Difference
Carroll	37.48	36.91	1.5%	48.90	48.82	0.2%	60.32	60.74	-0.7%
Clinton	45.90	45.90	0.0%	55.47	55.47	0.0%	65.00	65.00	0.0%
DeKalb	38.15	38.18	-0.1%	48.29	49.00	-1.5%	58.44	59.82	-2.4%
Edwards	46.07	44.28	3.9%	56.73	54.54	3.9%	67.40	64.98	3.6%
Hancock	40.72	42.31	-3.9%	52.22	52.64	-0.8%	63.84	63.05	1.2%
Henry	39.56	40.35	-2.0%	50.44	50.87	-0.8%	61.35	61.54	-0.3%
Jackson	45.94	45.45	1.1%	57.30	56.69	1.1%	68.65	68.01	0.9%
Jasper	44.34	43.77	1.3%	54.82	54.84	0.0%	65.36	65.92	-0.9%
Kankakee	40.00	39.75	0.6%	50.76	50.45	0.6%	61.53	61.14	0.6%
Lake	37.16	37.67	-1.4%	46.94	47.99	-2.2%	56.73	58.34	-2.8%
Logan	41.38	41.96	-1.4%	52.68	53.13	-0.9%	63.97	64.32	-0.5%
Marion	44.30	45.18	-2.0%	54.81	55.47	-1.2%	65.32	65.79	-0.7%
Massac	47.57	47.37	0.4%	58.10	57.71	0.7%	68.59	68.11	0.7%
Morgan	40.96	42.90	-4.7%	52.97	53.97	-1.9%	64.97	65.04	-0.1%
Perry	47.20	45.55	3.5%	57.64	56.17	2.6%	68.08	66.83	1.8%
Pike	42.20	42.58	-0.9%	53.12	53.38	-0.5%	64.14	64.23	-0.1%
Putnam	39.50	40.26	-1.9%	51.07	51.20	-0.3%	62.62	62.12	0.8%
Shelby	43.64	44.13	-1.1%	54.22	54.75	-1.0%	64.82	65.43	-0.9%
Union	45.24	45.17	0.1%	57.51	57.60	-0.2%	69.94	70.19	-0.4%
Woodford	39.48	40.04	-1.4%	51.17	51.23	-0.1%	62.98	62.45	0.8%

TABLE IX. RESULTS OF TEMPERATURE IMPUTATION METHOD VERIFICATION FOR MEAN SUMMER TEMPERATURES 2002-2013

	Mean Minimum Temperature (F)			Mean Temperature (F)			Mean Maximum Temperature (F)		
	Actual	Imputed	Percent Difference	Actual	Imputed	Percent Difference	Actual	Imputed	Percent Difference
Carroll	59.60	58.86	1.2%	71.29	71.24	0.1%	82.98	83.63	-0.8%
Clinton	66.67	66.67	0.0%	76.34	76.34	0.0%	85.97	85.97	0.0%
DeKalb	61.58	60.75	1.3%	71.54	71.55	0.0%	81.55	82.36	-1.0%
Edwards	66.29	66.34	-0.1%	77.18	77.18	0.0%	88.05	87.93	0.1%
Hancock	62.60	64.63	-3.2%	74.08	74.83	-1.0%	85.57	85.04	0.6%
Henry	61.96	63.08	-1.8%	72.52	73.16	-0.9%	83.09	83.31	-0.3%
Jackson	65.92	65.82	0.1%	77.33	76.86	0.6%	88.75	87.91	0.9%
Jasper	64.50	64.16	0.5%	75.46	75.28	0.2%	86.42	86.40	0.0%
Kankakee	63.08	61.62	2.3%	73.28	72.20	1.5%	83.47	82.78	0.8%
Lake	60.87	60.54	0.6%	70.78	71.11	-0.5%	80.70	81.69	-1.2%
Logan	61.76	62.95	-1.9%	73.00	73.97	-1.3%	84.26	84.99	-0.9%
Marion	65.01	65.31	-0.5%	75.70	75.88	-0.2%	86.40	86.44	0.0%
Massac	66.83	66.70	0.2%	77.32	77.31	0.0%	87.83	87.91	-0.1%
Morgan	61.68	63.66	-3.2%	73.66	74.65	-1.3%	85.63	85.66	0.0%
Perry	66.01	65.86	0.2%	76.69	76.64	0.1%	87.39	87.41	0.0%
Pike	63.11	64.00	-1.4%	74.26	74.71	-0.6%	85.43	85.43	0.0%
Putnam	61.81	62.34	-0.9%	73.22	73.09	0.2%	84.61	83.83	0.9%
Shelby	64.33	64.55	-0.3%	74.99	75.25	-0.3%	85.66	85.98	-0.4%
Union	65.99	65.99	0.0%	77.16	77.19	0.0%	88.37	88.42	-0.1%
Woodford	61.37	62.04	-1.1%	72.89	72.94	-0.1%	84.42	83.84	0.7%

TABLE X. MEAN MONTHLY TEMPERATURE FOR ALL OF ILLINOIS AND BY CLIMATE DIVISION, 2002-2013

		Climate Division								
Temperature (ºF)	**State**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**
Mean Temp January	27.18	22.33	23.54	25.01	26.31	25.30	28.36	28.92	32.58	32.04
Mean Temp February	29.15	25.09	25.34	27.08	27.59	25.83	30.19	31.52	34.54	33.89
Mean Temp March	41.87	37.58	37.86	40.30	41.44	40.51	42.71	43.85	46.18	46.01
Mean Temp April	52.69	48.88	48.71	52.10	52.66	51.94	53.71	54.09	56.13	56.36
Mean Temp May	62.08	59.14	58.35	62.50	61.17	61.04	63.58	63.95	64.54	64.37
Mean Temp June	71.37	68.89	68.95	72.05	72.19	69.52	71.96	72.58	72.89	73.29
Mean Temp July	74.89	74.05	73.24	73.64	74.45	74.66	75.49	74.59	78.25	75.54
Mean Temp August	73.88	71.78	71.98	73.00	73.51	72.65	73.84	75.20	77.29	75.36
Mean Temp September	66.45	62.61	65.00	66.33	66.68	65.28	67.72	67.71	68.12	68.87
Mean Temp October	53.20	51.25	51.93	52.77	53.70	51.53	54.81	53.48	53.84	55.31
Mean Temp November	42.81	40.17	40.71	41.97	40.37	42.06	43.29	43.98	45.67	46.85
Mean Temp December	31.14	26.90	27.67	29.33	30.23	29.54	32.03	32.92	35.49	35.74

TABLE XI. MEDIAN TOTAL MONTHLY PRECIPITAITON FOR ALL OF ILLINIOS AND BY CLIMATE DIVISION, 2002-2013

		Climate Division								
	State	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**
Precipitation (inches)	Median	Median	Median	Median	Median	Median	Median	Median	Median	Median
Total Precipitation April	6.03	7.66	7.95	4.65	6.82	8.67	4.43	5.98	5.295	4.64
Total Precipitation May	5.25	7.98	9.22	4.81	6.43	8.01	3.67	4.07	3.7	3.43
Total Precipitation June	4.81	3.93	5.75	4.21	5.30	5.49	4.01	4.98	4.84	4.37
Total Precipitation July	5.03	5.44	6.73	3.91	5.36	6.55	3.89	5.15	4.52	4.81
Total Precipitation August	4.42	3.71	4.78	3.22	5.23	5.45	4.09	5.28	4.77	4.49
Total Precipitation September	4.46	4.01	5.44	3.36	4.44	6	3.65	4.95	4.96	4.81
Total Precipitation October	3.49	2.69	3.58	2.03	3.43	4.1	3.135	4.25	4.63	4.19

Figure 5. Graph of mean winter temperature and annual NWNV incidence rate for Illinois by year, 2002-2013

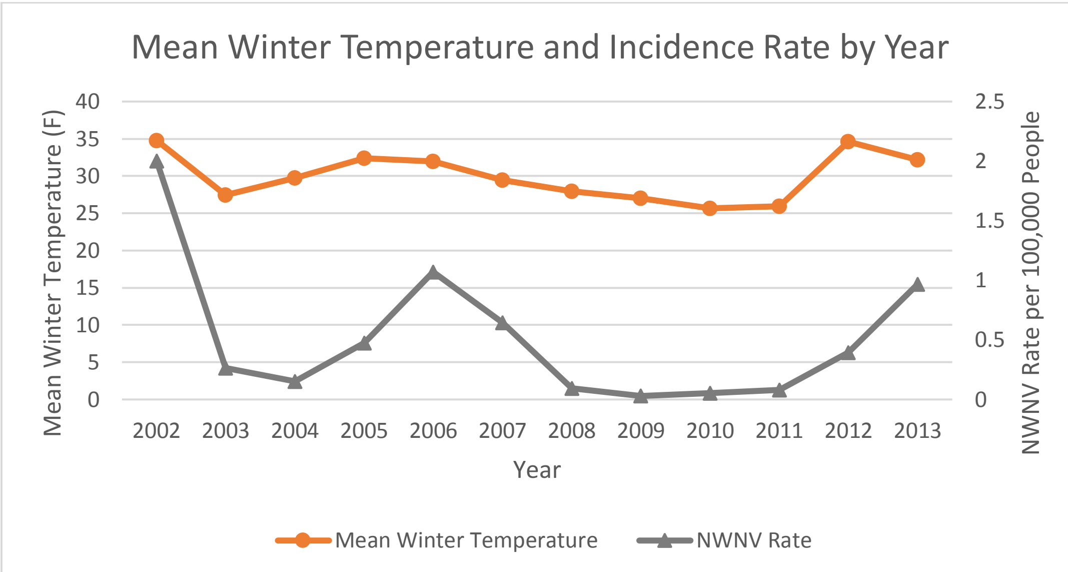


Figure 6. Graph of mean spring temperature and annual NWNV incidence rate for Illinois by year, 2002-2013

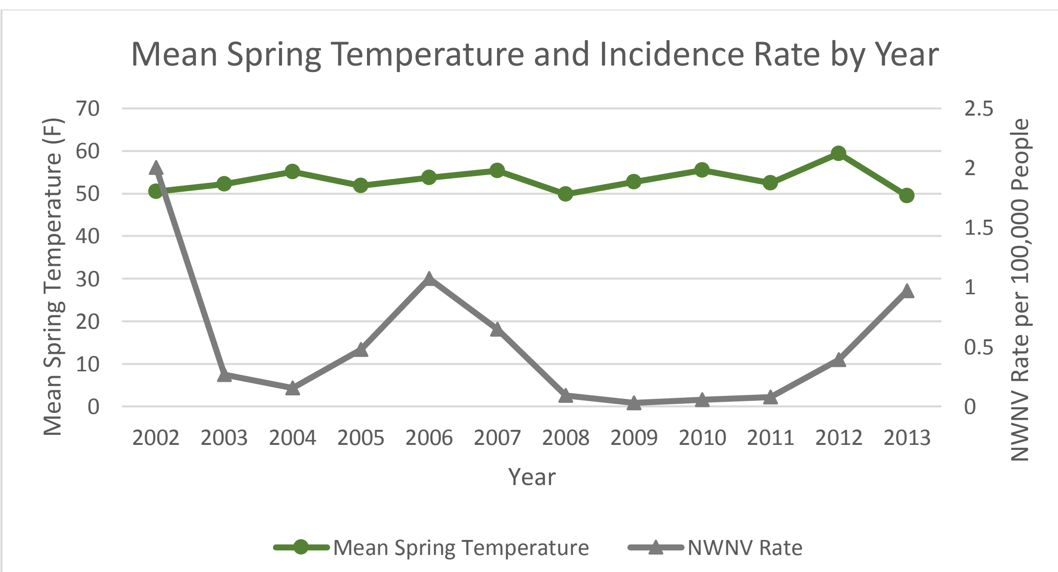


Figure 7. Graph of mean summer temperature and annual NWNV incidence rate for Illinois by year, 2002-2013

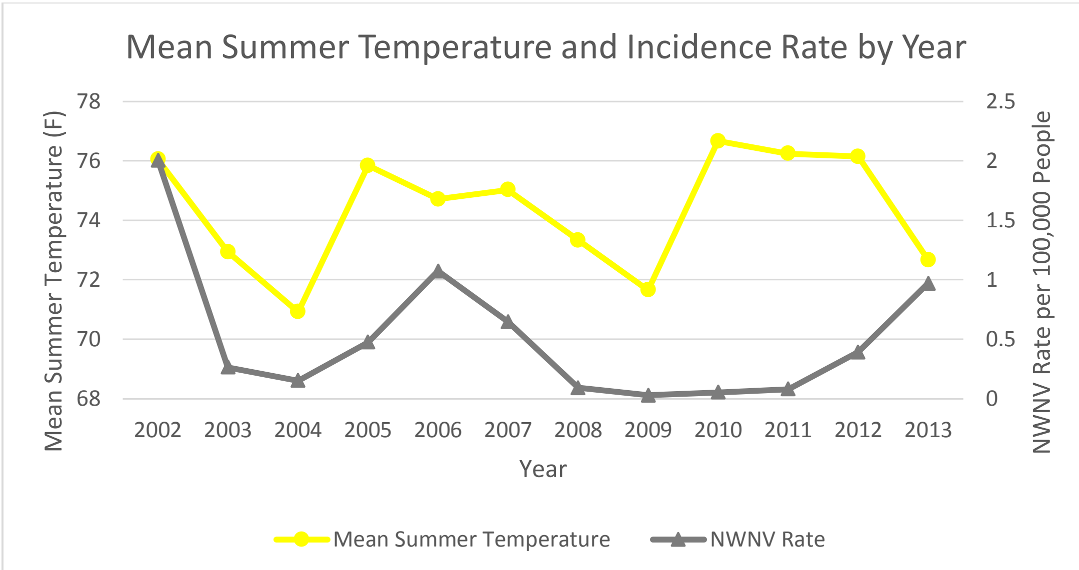


Figure 8. Graph of total fall precipitation and annual NWNV incidence rate for Illinois by year, 2002-2013

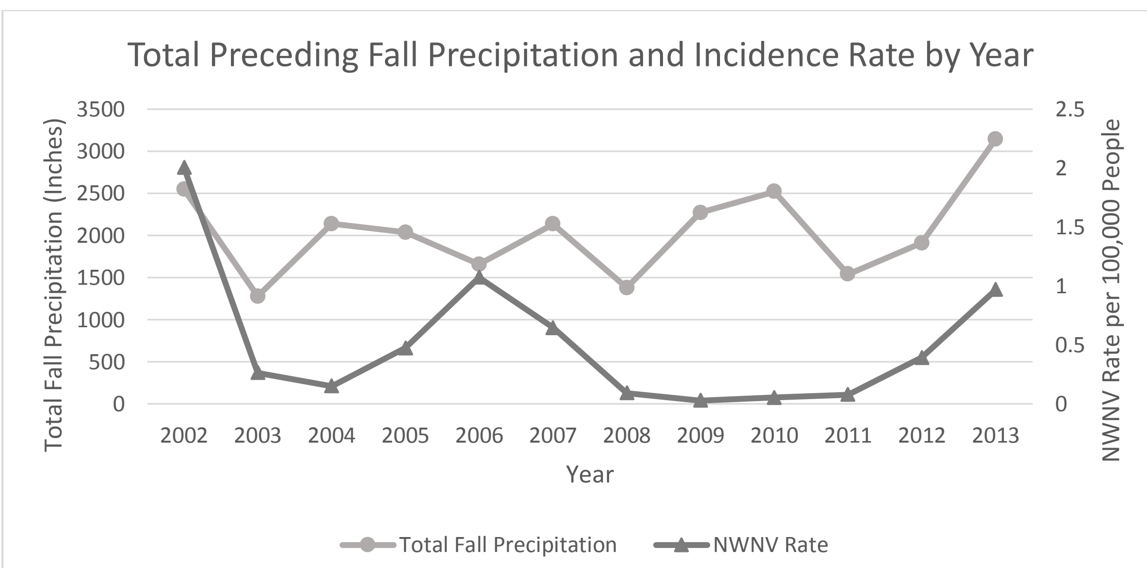


Figure 9. Graph of total spring precipitation and annual NWNV incidence rate for Illinois by year, 2002-2013

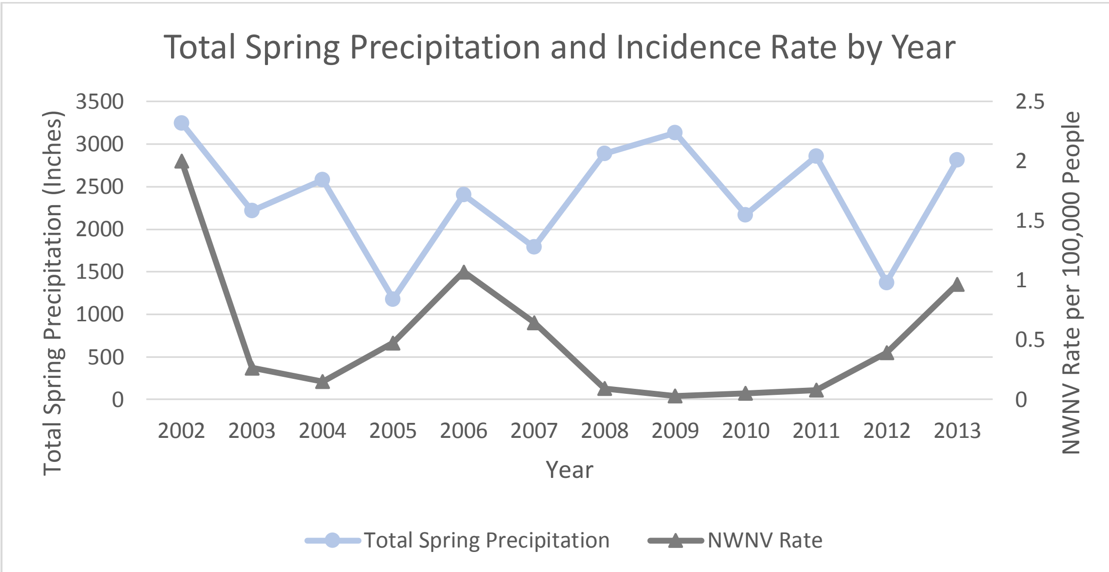
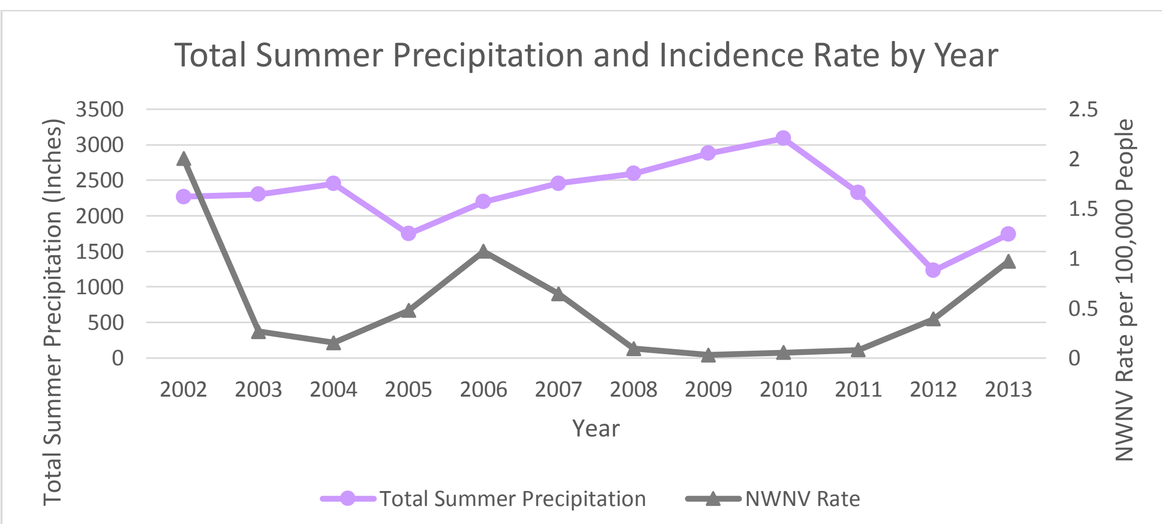


Figure 10. Graph of total summer precipitation and annual NWNV incidence rate for Illinois by year, 2002-2013



Odds ratios were also calculated using a dichotomous outcome variable based on 12 year NWNV incidence rate and dichotomous land cover, these results are provided in Tables XXII-XXIII, Appendix C. Having a 12 year incidence rate of more than 3 per 100,000 was significantly negatively associated with a high percent of county area with herbaceous wetland or wooded wetlands. Having a high amount of developed open space was moderately associated with increased 12-year incidence rates of NWNV.

The results of simple linear regression with the natural log of NWNV rates as an outcome with a continuous weather variable is provided in Table XIII, this analysis also looked at each weather variable squared. Summer temperature appears to potentially have a quadratic relationship with NWNV rates, but fall precipitation and winter temperature appear to have a linear relationship. Spring and summer

temperatures appear to have a potential quadratic relationship with NWNV rates, but the p-value for the linear temperatures had lower p-values. Odds ratio from a 2 X 2 contingency table analysis using two dichotomous variables is provided in Tables XXII-XXIII, Appendix C.

B. <u>Model Results</u>

Initial model results for ZINB regression are presented in Table XIV. Significant covariates for NWNV rates were RUCC, percent of population over 50, mean minimum winter temperature, mean summer temperature, and mean summer temperature squared. The model was controlled for cases that occurred in the first year; year-to-year trend was not significant and was highly influenced by the large number of cases in 2002. There were no additional confounding variables that changed any of the β estimates more than 10%. In this model RUCC was treated as a nominal variable and the referent group was RUCC9. Counties in suburban and urban areas with RUCC 1-4 were all significant and showed

TABLE XII. RESULTS OF SIMPLE LINEAR REGRESSION MEASURING THE ASSOCIATION OF LAND COVER VARIABLES WITH 12 YEAR NEUROINVASIVE WEST NILE VIRUS INCIDENCE RATES

	β Coefficient	**Standard Error**	**p-value**	**R^2**
Percent Cropland	0.000	0.005	0.965	0.000
Percent Non-Crop Agricultural Land	0.247	1.006	0.807	0.001
Percent Open Water	-0.048	0.055	0.391	0.007
Percent Developed Open Space	0.081	0.046	0.081	0.030
Percent Low Intensity Developed Land*	0.033	0.016	0.047	0.039
Percent Medium Intensity Developed Land	0.063	0.033	0.058	0.036
Percent High Intensity Developed Land	0.130	0.072	0.074	0.032
Percent Barren Land	1.191	0.903	0.191	0.017
Percent Deciduous Forest	-0.008	0.008	0.306	0.011
Percent Evergreen Forest	-0.119	0.146	0.419	0.007
Percent Herbaceous Grassland	-0.009	0.014	0.532	0.004
Percent Wooded Wetlands*	-0.159	0.067	0.020	0.053
Percent Herbaceous Wetlands*	-0.512	0.158	0.002	0.095
Combined Land Cover Groups				
Percent Developed*	0.018	0.009	0.038	0.043
Percent Wetland*	-0.144	0.051	0.006	0.075
Percent Wetland and Grassland	-0.016	0.013	0.215	0.015
Percent Forest	-0.008	0.008	0.299	0.011
*p-value <0.05				

TABLE XIII. RESULTS OF SIMPLE LINEAR REGRESSION MEASURING THE ASSOCIATION OF WEATHER VARIABLE WITH ANNUAL INCIDENCE RATES OF NEUROINVASIVE WEST NILE VIRUS

	β-estimate	S.E. X	p-value	β-estimate X^2	SE X^2	X^2 p-value	r^2
FALL							
Total Fall Precipitation	0.003	0.001	0.019				0.0070
Total Fall Precipitation Squared	0.000	0.000	0.968	0.000	0.000	0.289	0.0077
WINTER							
Mean Maximum Winter Temperature	0.014	0.003	<.0001				0.0225
Mean Maximum Winter Temperature Squared	0.027	0.033	0.419	0.000	0.000	0.699	0.0226
Mean Minimum Winter Temperature	0.015	0.003	<.0001				0.0208
Mean Minimum Winter Temperature Squared	0.048	0.050	0.012	-0.001	-0.001	0.080	0.0233
Mean Winter Temperature	0.015	0.003	<.0001				0.0224
Mean Winter Temperature Squared	0.022	0.030	0.467	0.000	0.000	0.829	0.0224
Total Winter Precipitation	0.000	0.002	0.972				0.0000
Total Winter Precipitation Squared	0.003	0.005	0.535	0.000	0.000	0.501	0.0004
SPRING							
Mean Maximum Spring Temperature	-0.007	0.003	0.022				0.0220
Mean Maximum Spring Temperature Squared	0.077	0.041	0.063	-0.001	0.000	0.043	0.0076
Mean Minimum Spring Temperature	-0.010	0.004	0.005				0.0063
Mean Minimum Spring Temperature Squared	0.078	0.056	0.172	-0.001	0.001	0.122	0.0083
Mean Spring Temperature	-0.010	0.003	0.004				0.0070
Mean Spring Temperature Squared	0.073	0.060	0.310	-0.001	0.001	0.238	0.0081
Total Spring Precipitation	0.001	0.001	0.165				0.0017
Total Spring Precipitation Squared	0.000	0.002	0.865	0.000	0.000	0.710	0.0018
SUMMER							
Mean Maximum Summer Temperature	0.004	0.003	0.214				0.0013
Mean Maximum Summer Temperature Squared	0.042	0.047	0.379	0.000	0.000	0.454	0.4310
Mean Minimum Summer Temperature	0.008	0.005	0.121				0.0708
Mean Minimum Summer Temperature Squared	0.368	0.183	0.044	-0.003	0.001	0.030	0.0497
Mean Summer Temperature	0.006	0.005	0.238				0.0009
Mean Summer Temperature Squared	0.497	0.202	0.014	-0.003	0.001	0.016	0.0056
Total Summer Precipitation	0.000	0.001	0.779				0.0001
Total Summer Precipitation Squared	-0.004	0.003	0.081	0.000	0.000	0.129	0.0003

TABLE XIV. MULTIVARIABLE MODEL RESULTS MEASURING THE ASSSOCITATION OF URBANIZATION, WEATHER, AND LAND COVER WITH ANNUAL INCIDENCE RATES OF NEUROINVASIVE WEST NILE VIRUS IN ILLINOIS FROM 2002-2013

	β Estimate	Standard Error	p-value
Intercept	-118.40	46.26	0.011
RUCC 8	-0.18	0.77	0.81
RUCC 7	-0.21	0.54	0.70
RUCC 6	-0.43	0.54	0.42
RUCC 5	-0.12	0.97	0.90
RUCC 4	-1.24	0.57	0.028
RUCC 3	-1.15	0.57	0.043
RUCC 2	-1.37	0.57	0.017
RUCC 1	-1.12	0.59	0.059
RUCC 9 - Reference	0	0	.
Percent of Population Over 50	0.11	0.02	<.0001
Mean Minimum Winter Temperature	0.17	0.03	<.0001
Mean Summer Temperature	3.11	1.25	0.013
Mean Summer Temperature Squared	-0.02	0.008	0.011
First Year	0.32	0.98	0.75
Dispersion	0.63	0.17	
Zero Model	β Estimate	Standard Error	p-value
Intercept	-8.7017	1.9088	<.0001
RUCC 8	0.1017	0.9486	0.9146
RUCC 7	-0.1726	0.6668	0.7958
RUCC 6	0.0067	0.6563	0.9918
RUCC 5	1.4935	0.984	0.1291
RUCC 4	-1.8854	0.7112	0.008
RUCC 3	-1.1102	0.6812	0.1032
RUCC 2	-1.8401	0.737	0.0125
RUCC 1	-1.5341	0.7368	0.0373
RUCC 9 - Reference	0	0	.
Percent of Population Over 50	0.2885	0.046	<.0001
First Year	-1.2328	1.9315	0.5233
Goodness-of-fit			
Deviance	1591.69		
Log Likelihood	-795.85		

similar measures of effect, but counties in RUCC 5-8 were not significantly. Counties in RUCC 1-4 were grouped and RUCC 5-8 were removed from the model. The final model for the state of Illinois is presented in Table XV.

Table XV shows results of ZINB models stratified by urbanization. Counties in the group defined as urban and suburban counties are part of either a metropolitan or micropolitan CBSA; counties defined as rural are not grouped in either CBSA. Percent developed land was significant for both urban and suburban areas as well as rural areas, but the strength and direction of the relationship for each is different. In urban and suburban counties the percent of population over the age of 50 and mean winter temperature were significantly and positively associated with annual incidence rates of NWNV; these were not significantly associated in rural counties. The only significant association with NWNV rates in rural counties was developed land.

Three sensitivity analyses were conducted comparing the ZINB model results for the state of Illinois; the results for these are presented in Table XVII and Table XVIII. One sensitivity analysis looked at results without data from the year 2002, and the other looked at results without data from Cook County. The β estimates did not significantly change for any of the covariates in either sensitivity analysis, the confidence intervals of each β estimate overlap.

TABLE XV. FINAL MODEL RESULTS FOR THE ASSOCIATION OF URBANIZATION, WEATHER, AND LAND COVER WITH ANNUAL INCIDENCE RATES OF NEUROINVASIVE WEST NILE VIRUS IN ILLINOIS FROM 2002-2013

	β Estimate	Standard Error	p-value
Intercept	-121.04	45.93	0.008
RUCC 1 - 4	-0.93	0.20	<.0001
RUCC 5-9 – Reference	0	0	.
Percent of Population Over 50	0.10	0.02	<.0001
Mean Minimum Winter Temperature	0.17	0.03	<.0001
Mean Summer Temperature	3.18	1.24	0.010
Mean Summer Temperature Squared	-0.02	0.01	0.009
First Year	0.38	0.98	0.70
Dispersion	0.65	0.17	
Zero Model	β Estimate	Standard Error	p-value
Intercept	-7.2881	1.2765	<.0001
RUCC 1 - 4	-1.6597	0.2425	<.0001
RUCC 5-9 – Reference	0	0	.
Percent of Population Over 50	0.2528	0.0337	<.0001
First Year	-1.341	1.9495	0.4916
Goodness-of-fit			
Deviance	1602.84		
Log Likelihood	-801.42		

Figure 11. Counts and Rates of Neuroinvasive West Nile Virus by Illinois County, and County Core Based Statistical Area Classification, 2002-2013

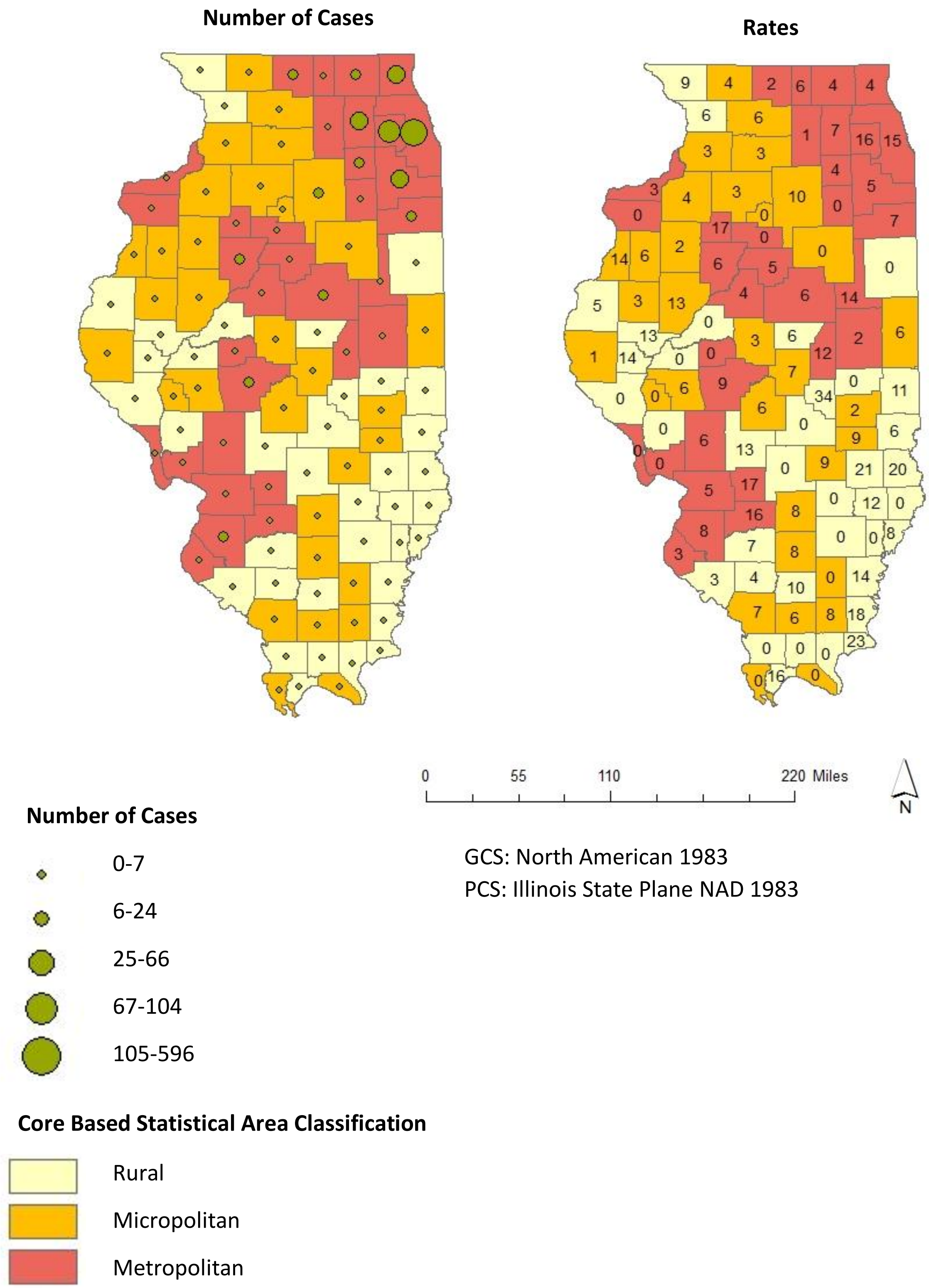


TABLE XVI. MODEL RESULTS FOR THE ASSOCIATION OF URBANIZATION, WEATHER, AND LAND COVER WITH ANNUAL INCIDENCE RATES OF NEUROINVASIVE WEST NILE VIRUS IN THE STATE OF ILLINOIS FROM 2002-2013, STRATIFIED BY URBAN AND RURAL COUNTIES

Urban and Suburban Counties			
	β Estimate	Standard Error (SE)	p-value
Intercept	-8.94	1.11	<.0001
Percent of Population Over 50	0.14	0.024	<.0001
Percent Developed Land	0.01	0.005	0.028
Mean Winter Temperature	0.14	0.02	<.0001
First Year	0.22	1.04	0.83
Dispersion	0.79	0.22	
Zero Model	β Estimate	SE	p-value
Intercept	-4.6724	2.4737	0.0589
Percent of Population Over 50	0.1825	0.0629	0.0037
Population Density	-0.0106	0.004	0.008
First Year	-18.5592	34976.53	0.9996
Goodness-of-fit			
Deviance	1084.31		
Log Likelihood	-542.15		
Rural Counties	β Estimate	SE	p-value
Intercept	4.02	0.45	<.0001
Percent Developed Land	-0.22	0.05	<.0001
First Year	-5.59	15.94	0.73
Dispersion			
Zero Model	β Estimate	Standard Error	p-value
Intercept	4.98	1.1	<.0001
Percent Developed Land	-0.29	0.12	0.015
Goodness-of-fit			
Deviance	444.85		
Log Likelihood	-222.42		

TABLE XVII. SENSITIVITY ANALYSIS COMPARING MODEL RESULTS USING THE FULL DATASET, A DATASET EXCLUDING COUNTIES WITH IMPUTED TEMPERATURES FOR 2002-2013

	All Years and All Counties			**Excluding Imputed Temperatures**		
	β Estimate	Standard Error	p-value	β Estimate	Standard Error	p-value
Intercept	-121.04	45.93	0.008	-179.033	55.4259	0.0012
RUCC 1 - 4	-0.93	0.20	<.0001	-0.8344	0.2192	0.0001
RUCC 5-9 – Reference	0	0	.	0	0	.
Percent of Population Over 50	0.10	0.02	<.0001	0.101	0.0215	<.0001
Mean Minimum Winter Temperature	0.17	0.03	<.0001	0.203	0.0288	<.0001
Mean Summer Temperature	3.18	1.24	0.010	4.7497	1.4906	0.0014
Mean Summer Temperature Squared	-0.02	0.01	0.009	-0.0327	0.01	0.0011
First Year	0.38	0.98	0.70	0	0	.
Dispersion	0.65	0.17		0.363	0.1223	
Zero Model	β Estimate	Standard Error	p-value	-16.92	2.71	<.0001
Intercept	-7.2881	1.2765	<.0001	-1.31	0.30	<.0001
RUCC 1 - 4	-1.6597	0.2425	<.0001	1.44	0.85	0.09
RUCC 5-9 – Reference	0	0	.	29.49	4.74	<.0001
Percent of Population Over 50	0.2528	0.0337	<.0001	0.12	0.03	<.0001
First Year	-1.341	1.9495	0.4916	-0.01	0.01	0.27
Goodness-of-fit				0.00	0.00	
Deviance	1602.84					
Log Likelihood	-801.42			1167.00		

TABLE XVIII. SENSITIVITY ANALYSIS COMPARING MODEL RESULTS USING THE FULL DATASET, A DATASET EXCLUDING 2002, AND A DATASET EXCLUDING COOK COUNTY

	All Years and All Counties			**Excluding 2002 Cases**			**Excluding Cook County**		
	β Estimate	Standard Error	p-value	β Estimate	Standard Error	p-value	β Estimate	Standard Error	p-value
Intercept	-121.04	45.93	0.008	-120.88	46.05	0.009	-114.96	46.89	0.014
RUCC 1 - 4	-0.93	0.20	<.0001	-0.93	0.20	<.0001	-0.94	0.21	<.0001
RUCC 5-9 – Reference	0	0	.	0.00	0.00	.	0.00	0.00	.
Percent of Population Over 50	0.10	0.02	<.0001	0.10	0.02	<.0001	0.10	0.02	<.0001
Mean Minimum Winter Temperature	0.17	0.03	<.0001	0.17	0.03	<.0001	0.17	0.03	<.0001
Mean Summer Temperature	3.18	1.24	0.010	3.17	1.24	0.011	3.01	1.27	0.017
Mean Summer Temperature Squared	-0.02	0.01	0.009	-0.02	0.01	0.009	-0.02	0.01	0.015
First Year	0.38	0.98	0.70	0.00	0.00	.	0.38	1.00	0.703
Dispersion	0.65	0.17		0.67	0.18		0.69	0.19	
Zero Model	β Estimate	Standard Error	p-value	β Estimate	Standard Error	p-value	β Estimate	Standard Error	p-value
Intercept	-7.29	1.28	<.0001	-7.26	1.28	<.0001	-6.76	1.29	<.0001
RUCC 1 - 4	-1.66	0.24	<.0001	-1.66	0.24	<.0001	-1.64	0.24	<.0001
RUCC 5-9 – Reference	0	0	.	0.00	0.00	.	0.00	0.00	.
Percent of Population Over 50	0.25	0.03	<.0001	0.25	0.03	<.0001	0.24	0.03	<.0001
First Year	-1.34	1.95	0.49	0.00	0.00		-1.35	1.92	0.484
Goodness-of-fit									
Deviance	1602.84			1597.63			1557.02		
Log Likelihood	-801.42			-798.82			-788.17		

CHAPTER IV. DISCUSSION

This study analyzed incidence rates of NWNV in 102 Illinois counties for 12 years and assessed the associations with land cover, weather, and urbanization variables. In the final multivariable zero-inflated negative binomial model annual NWNV rates were associated with warm winter and summer temperatures. NWNV rates were also directly associated with percent of population over the age of 50, and negatively associated with living in a county with a RUCC 1-4. The total population of counties in RUCC 1-4 ranged from 5,089 people in Calhoun County to 5,194,675 people in Cook County, based on 2010 Census data The percent of population over the age of 50, percent cropland, and mean winter temperature were important predictors of annual NWNV incidence rates for urban and suburban counties in Illinois. Percent population over the age of 50 was significant in the final models for the whole state and for urban counties, this is likely to be an indication of county susceptibility to NWNV due to having a larger older population. Tables XXV-XXVI, Appendix D show an increase in effect on NWV incidence (larger beta-coefficients) with increasing percent population over 50, 60, 70, and 80. Percent developed land was an important predictor of annual NWNV incidence rates for both urban and rural counties, but had a different directions of association. However, with 40 cases in this 12 year study period there is a lack of statistical power in rural counties that makes determining urban-rural differences in hypothesis 4 inconclusive.

A bivariate analysis with land cover and 12 year NWNV incidence rates revealed an inverse association with annual NWNV incidence rates and both herbaceous wetland and wooded wetlands, but these were not significant in the ZINB models. There were no significant land cover variables in the final model that included all counties in the state of Illinois. This is in agreement with a previous study that found temperature in winter, but not land cover, to be significantly associated with having high rates of human WNV infections in the North-Central US (DeGroot et al. 2014). These results are in contrast to what was expected in hypothesis 1 and previous studies that assessed the association of land cover with bird and mosquito WNV infections. The multivariable model measured the association between static land cover values with dynamic annual NWNV incidence rates; this is a limitation and may be attenuating the effect of land cover. Previous studies found an association of mosquito infection rates found with urban development, water area, and grass area, and another study that found mosquito infection rates associated with wetlands and forests (Liu et al. 2008, Chuang & Wimberly 2012). Differences seen in this study compared to previous research could be due to the small amount of NWNV cases compared to all WNV infections, resulting in a lack of power; these results also may differ due to a measure of human activity and human proximity to mosquitoes that is essential for WNV transmission to humans. Models stratified by urbanization showed that developed land was associated with NWNV infection rates for both urban and rural areas, but the direction of the association differed. Urban and suburban counties had a small but significant direct association with percent developed land and NWNV rates, there was an inverse association with percent developed land in rural counties and a stronger measure of effect. The inverse association between NWNV rates and developed land seen in rural counties could be explained if areas with developed land in rural counties are more likely to practice mosquito abatement compared to areas with a smaller percent of developed land. A low population density in rural counties combined with mosquito abatement could indicate the lack of proximity between mosquitoes and humans.

The final model showed increased risk of NWNV with warmer temperatures in winter and summer months with a quadratic relationship with summer temperatures. This is mostly in agreement with hypothesis 2 that anticipated a significant association between NWNV rates and warm spring temperatures. The quadratic relationship with summer temperature supports previous research that shows decreased rates when temperatures are exceptionally high (Reisen et al. 2008).

Contrary to hypothesis 3 precipitation was not a significant predictor in any of the final models. It is possible that all counties in Illinois generally get adequate rain for mosquito population growth. Previous studies in Russia that found precipitation to be a significant predictor of human WNV for some areas of Russia but not significant in areas that were generally rainy in comparison (Platonov et al. 2014). Extreme amounts of total seasonal precipitation may have been too rare of an occurrence to properly address in this study.

A. Strengths and Limitations

A large amount of literature on WNV focus on mosquito infection rates as outcomes; some consider bird, horse, or human WNV infections, and very few use NWNV infections. Human WNV infections are often subclinical or mild, and are vastly underreported. This underreporting may differ through both time and space and produce systematically biased results. This is one of few studies that explores human NWNV infections as an outcome; this outcome is expected to be more reliable because NWNV is expected to be nearly 100% reported.

The distribution of characteristics and mortality rate of NWNV in Table 1 are consistent with information from the CDC, with the exception of race and ethnicity which also had many missing values (6.9 % and 29.8%, respectively). It is possible that these are underreported in a non-random pattern if a person was more likely to skip this question when they felt like their race or ethnicity was not represented in any of the groups listed.

NWNV rates are estimated to be nearly 100% reported, however the fraction of neuroinvasive cases compared to all infections is small and most counties had 0 cases for most years. Additionally, age and immunosuppression are risk factors of developing neuroinvasive disease when infected with WNV. The results in this study do not simply reflect associations with land cover, weather, and urbanization on all

WNV cases. These results may indicate susceptibility of the population of a county to NWNV versus a measure of disease WNV incidence (not limited to NWNV). Rural counties in Illinois have a significantly older population than urban and suburban counties and rural counties may not have enough variability in population age to measure significant differences. Table XXIV, Appendix D provides statistics on percent county population over the age of 50, 60, 70 and 80 by rural or urban status. In the multivariable ZINB model, replacing percent of population over the age of 50 with ages of 60, 70, and 80 showed an increased measure of association with increased age; these results are provided in Tables XXV-XXVI, Appendix D.

Rates in rural counties may be inflated compared to urban counties since the total population is much smaller. The average rate decreased from 7.4 to 6.6 per 100,000 people when total NWNV case count of all rural counties was aggregated and divided by the sum of the populations for all rural counties; this was not observed in metropolitan or micropolitan areas.

The exact location of WNV transmission cannot be determined from the public health data available for analysis. This study made the assumption that a WNV infection was transmitted to the person in their county of residence. This may have caused some error if a person acquired WNV when traveling to a county outside of their residence.

Most NWNV cases occurred in Cook County, and many cases occurred in 2002. These factors may have a strong influence on the observed associations. However, the two different sensitivity analyses showed that the associations did not largely or meaningfully change when excluding data from 2002 or Cook County. Warm temperatures in 2002 created ideal conditions for increase Culex population density; this and lack of immunity both birds and humans played a role in the 2002 outbreak (IDPH, 2016a). Cook

County is the most populated in Illinois; while Cook County consistently had cases of NWNV it did not consistently have the highest rates compared to other urban counties.

Several limitations of this study were caused by the aggregation of time and space and fall under the umbrella of ecological fallacy. Incidence rates were reported at the county level and aggregated in time by year in order to calculate a yearly county incidence rate. Weather was reported at the county level and aggregated by season to obtain mean seasonal temperatures and total seasonal precipitation. Land cover characteristics were summed to obtain a percent of county area for each land cover type. This results in some loss of information on the distribution of the types of land cover throughout a county, this is an example of the modifiable areal unit problem. This information may be important if, for example, small areas of developed land and forest were scattered throughout a county and had a different effect on WNV transmission than a county where developed land and forest were contiguous. Moreover, individuals who developed NWNV in counties that had a high percent of forest (for example) may live quite far from the forested areas.

Human and avian immunity are important to consider because a non-fatal infection is expected result in lifelong immunity to subsequent WNV infections. 2002 was the first year with reported human WNV infections in Illinois, and at that time presumably all Illinois were susceptible to WNV infection, cases that occurred in the first year was controlled for in the final model. Years with high rates of WNV infections tend to be followed by years with lower rates of WNV infections and may be due to a large avian population with WNV immunity (Kwan et al. 2012).

Proximity of humans to mosquitoes and birds plays a critical role in transmission of WNV to humans, however, this study did not incorporate surveillance data that included mosquito or bird WNV infection rates. This could provide helpful information, but may add more complexity because surveillance methods for testing mosquito and bird WNV infection rates differs by county and time.

This study did not incorporate specific features of the built environment that offer ideal habitats for mosquitoes to reproduce and may play a role in WNV transmission to humans. *Culex pipiens* are usually found near stagnant water including bird baths, swimming pools, and spare tires; in the winter they try to hide in covered or hollowed areas where temperatures are more stable. This information could provide useful insight on the role of the built environment in WNV transmission to humans, but may change often and is data is not collected on a large scale.

B. Significance

The results presented in this study are specific to NWNV in Illinois, but the methods used in this study can be repeated for different geographic areas. These methods may also be used for other vector borne diseases where mosquitoes are the primary vector, results will vary because a virus may have a different mosquito species as a primary vector. Data for land cover, weather, and urbanization is available online and accessible to everyone at no cost. This allows a researcher to conduct a study following these methods that can be complete in a relatively quick time frame at little to no cost.

There is a growing interest in understanding how climate change will effect transmission of vector borne diseases. WNV is particularly important to study because it is the most common mosquito borne disease in the US, and Illinois is one of several states that has a history of high rates of WNV. An assessment was recently published by the US Global Change Research Program looking at the climate change impacts on human health, this assessment included a chapter on vector borne diseases and included WNV. The two most probable ways climate change may effect WNV transmission is longer WNV seasons and an expansion into small geographic areas where WNV is not endemic (Beard et al.

2016). Expansion may occur with changes to land cover over time influenced by climate change and urban development may create more suitable habitats for *Culex pipiens* mosquitoes. Warmer temperatures can result in both higher rates and a longer season of WNV due to an increase in population density of *Culex pipiens* and a longer period of time with increased mosquito activity. The National Climate Assessment projects warmer temperatures in all seasons for all of Illinois (Pryor et al. 2014).

The information gained from this study can be used to inform counties when they are at a higher risk of for a season with high rates of WNV. Communities have the opportunity to create a season specific WNV action plan that includes mosquito abatement and communication to residents. Communities can also construct communication systems to inform residents their level of risk for WNV each year and educate people on prevention strategies that they can do at home.

There are several opportunities for future studies to add to our understanding of WNV transmission in Illinois. Looking at changes in urbanization of counties over time would benefit our understanding of how urbanization effects WNV incidence rates. However, measureable county level changes in urbanization is relatively slow and this type of evaluation may need to include a longer study period. In the 12 years included in this study there were 16 counties that had change in RUCC, most changed by plus or minus 1. The impact of climate change on human WNV incidence can be evaluated by predicting future incidence rates of WNV in the context of climate change using state climate change projections and identifying areas that may be the most susceptible to changes in land cover. Obtaining WNV surveillance data on a smaller spatial scale is unlikely because it is protected health data, however, using a smaller spatial could provide us with more information on the distribution among different types of land cover that may play a significant role in WNV transmission. A shorter temporal scale could allow us to predict when to expect a lengthened WNV season. While current literature has a strong foundation of knowledge on factors associated with mosquito WNV infections, there is a need to understand how these relate to WNV transmission to humans. Obtaining specific data on features in the built environment, human activity, mosquito abatement, bird WNV surveillance, and mosquito WNV surveillance could provide more insight and support for specific factors that are associated to WNV transmission to humans.

APPENDICES

Appendix A

TABLE XIX. UNIVARIATE STATISTICS FOR CONTINUOUS VARIABLES FROM 2010 UNITED STATES CENSUS AND ILLINOIS WEST NILE VIRUS SURVEILLANCE 2002-2013

	Missing	Mean	Median	Standard Deviation	Skewness	Kurtosis
Rate and Population						
Percent of Population Over 50	0	0.37	0.38	0.05	-0.88	0.78
Total Population	0	125790.5	27315.5	527293.3	9.0	86.7
County Area (Square Miles)	0	545.03	511.00	223.06	0.71	0.19
Population Density	0	196.10	48.48	626.75	6.90	53.60
Annual NWNV Incidence Rate (per 100,000 people)	0	0.53	0.00	1.99	5.94	43.27
12 Year NWNV Incidence Rate (per 100,000 people)	0	0.00	0.00	0.00	1.41	2.72
*approx. normally distributed						

APPENDIX A (CONTINUED)

TABLE XX. UNIVARIATE STATISTICS FOR LAND COVER FROM 2007 ILLINOIS CROPLAND DATA LAYER

	Missing	Mean	Median	Standard Deviation	Skewness	Kurtosis
Percent Cropland	0	51.32	54.07	21.18	-0.58	-0.29
Percent Non-Crop Agricultural Land	0	0.03	0.01	0.10	5.85	37.17
Percent Open Water	0	1.90	1.30	1.89	2.10	5.53
Percent Developed Open Space	0	6.71	6.40	2.26	2.41	10.57
Percent Low Intensity Developed	0	4.39	2.82	6.26	4.68	24.96
Percent Medium Intensity Developed	0	1.14	0.35	3.14	6.30	44.36
Percent High Intensity Developed	0	0.41	0.08	1.43	7.23	58.03
Percent Barren	0	0.08	0.04	0.12	3.14	11.29
Percent Deciduous Forest	0	19.01	16.80	13.14	1.34	2.25
Percent Evergreen Forest	0	0.14	0.00	0.72	7.91	68.05
Percent Herbaceous Grassland	0	13.30	11.34	7.61	1.11	1.62
Percent Woody Wetland	0	1.18	0.63	1.52	2.33	7.21
Percent Herbaceous Wetland	0	0.37	0.15	0.63	3.12	10.94
Highest Elevation (Feet)	0	801.49	805.50	143.79	0.57	0.31
Lowest Elevation (Feet)	0	484.88	462.50	105.00	0.33	-0.71
*approx. normally distributed						

APPENDIX A (CONTINUED)

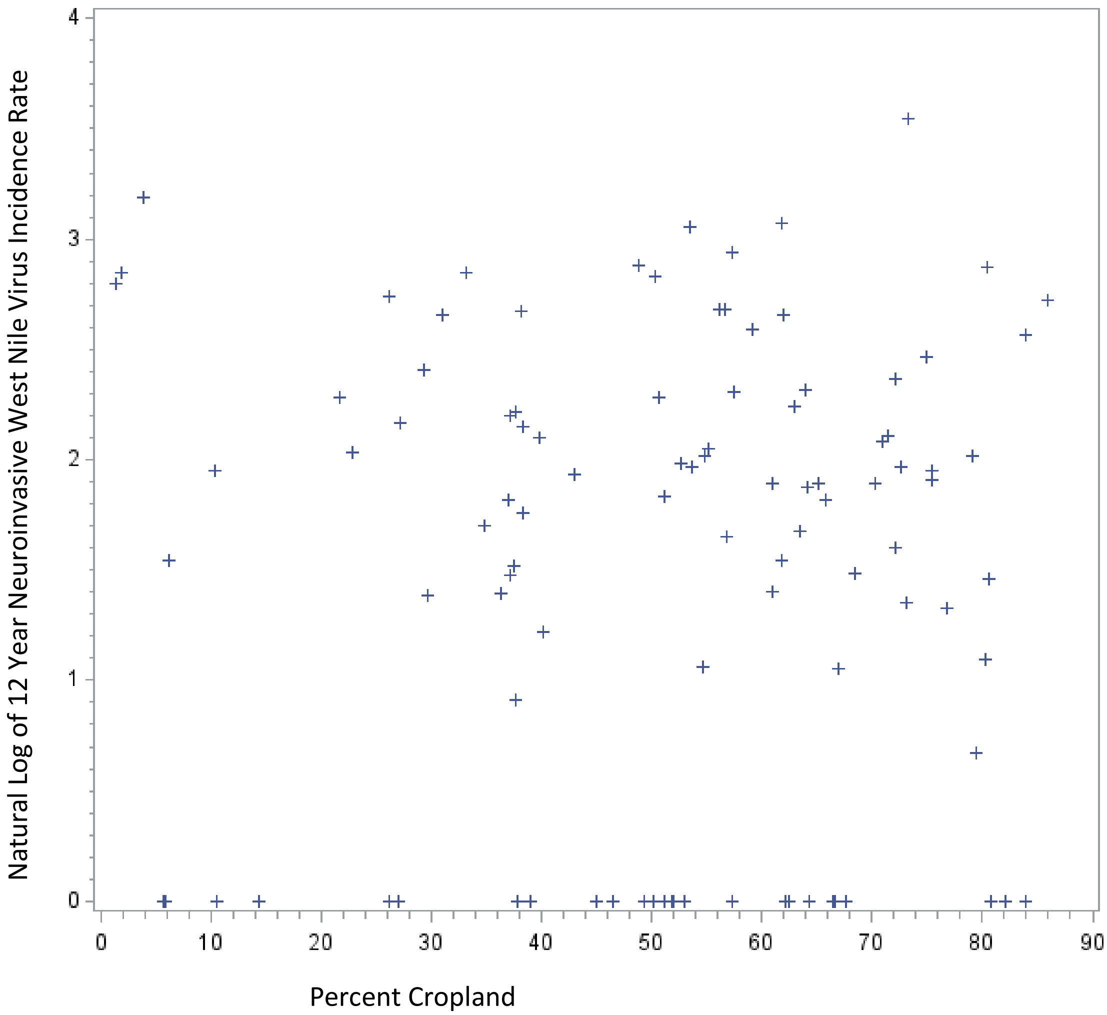
TABLE XXI. UNIVARIATE STATISTICS FOR SEASONAL WEATHER DATA FOR ILLINOIS COUNTIES 2002-2013

	Missing	Mean	Median	Standard Deviation	Skewness	Kurtosis
Total Precipitation Fall	84	21.20	17.29	14.44	1.71	4.97
Mean Minimum Temperature Winter	0	21.53	21.73	4.90	-0.02	-0.19
Mean Temperature Winter	0	29.97	30.10	5.00	-0.07	-0.64
Mean Maximum Temperature Winter	0	38.47	38.56	5.38	0.01	-0.68
Mean Minimum Temperature Spring	0	42.47	42.44	4.04	-0.03	-0.20
Mean Temperature Spring	0	53.20	53.18	4.27	-0.03	-0.01
Mean Maximum Temperature Spring	0	63.80	63.99	4.98	-0.47	1.41
Total Precipitation Spring	85	25.03	20.43	17.34	1.58	3.70
Mean Minimum Temperature Summer	0	63.64	63.58	2.74	0.08	-0.17
Mean Temperature Summer	0	74.36	74.53	2.85	-0.10	-0.25
Mean Maximum Temperature Summer	0	84.79	85.10	4.19	-2.14	10.65
Total Precipitation Summer	92	23.74	18.20	16.81	1.49	2.73
*approx. normally distributed						

Appendix B

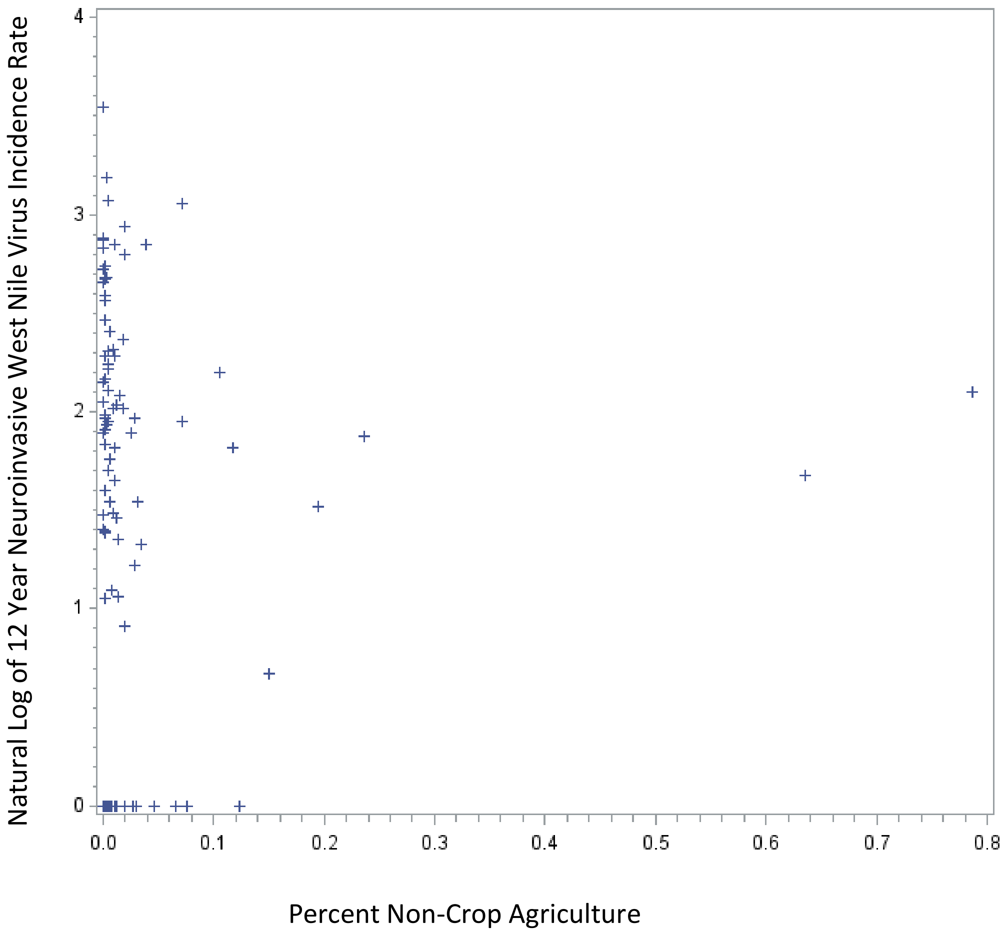
A. Land Cover Scatterplots

Figure 12. Scatterplot for Natural Log of 12 Year Neuroinvasive Incidence Rate by Percent Cropland



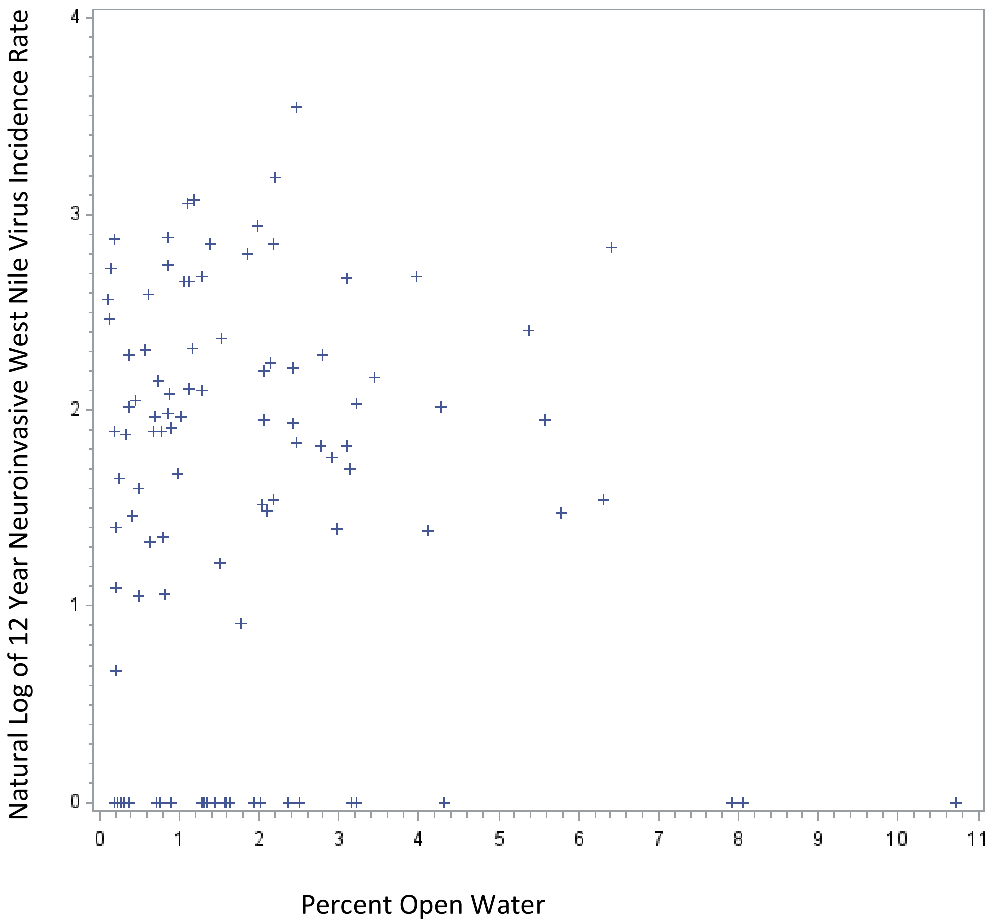
APPENDIX B (CONTINUED)

Figure 13. Scatterplot for Natural Log of 12 Year Neuroinvasive Incidence Rate by Percent Non-Crop Agriculture



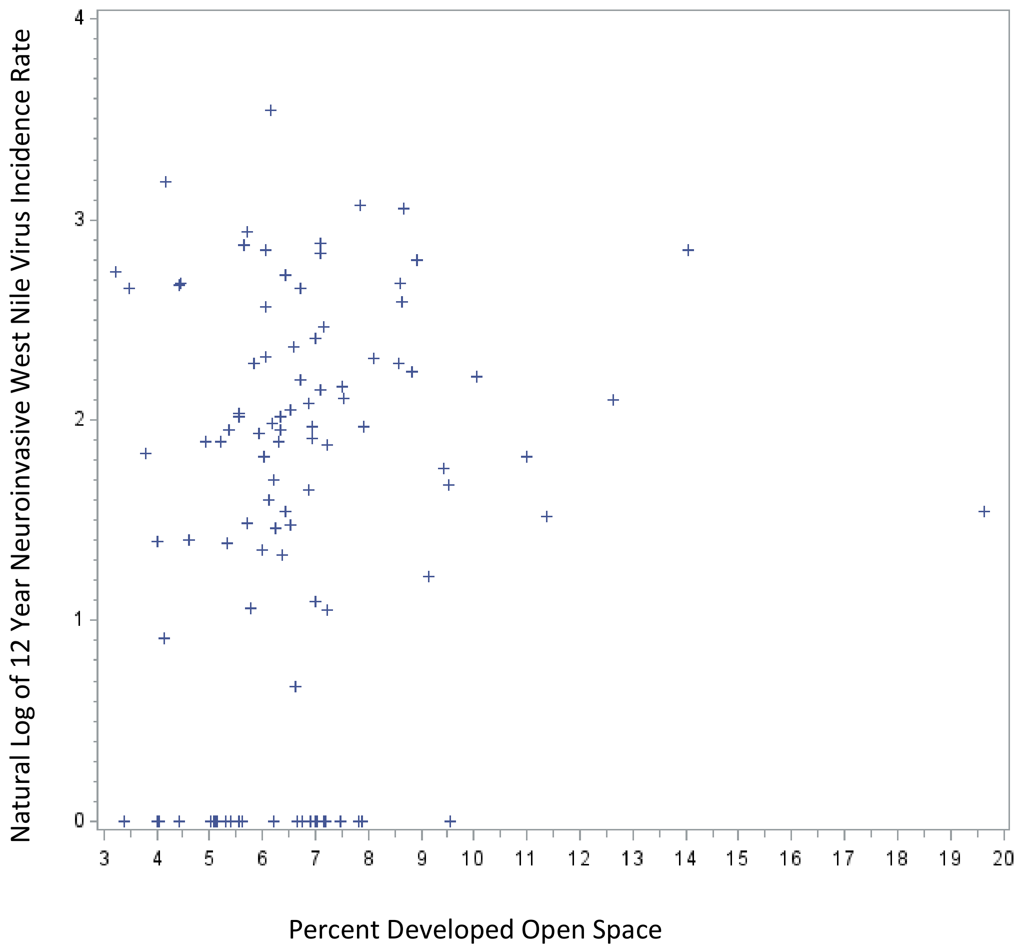
APPENDIX B (CONTINUED)

Figure 14. Scatterplot for Natural Log of 12 Year Neuroinvasive Incidence Rate by Percent Open Water



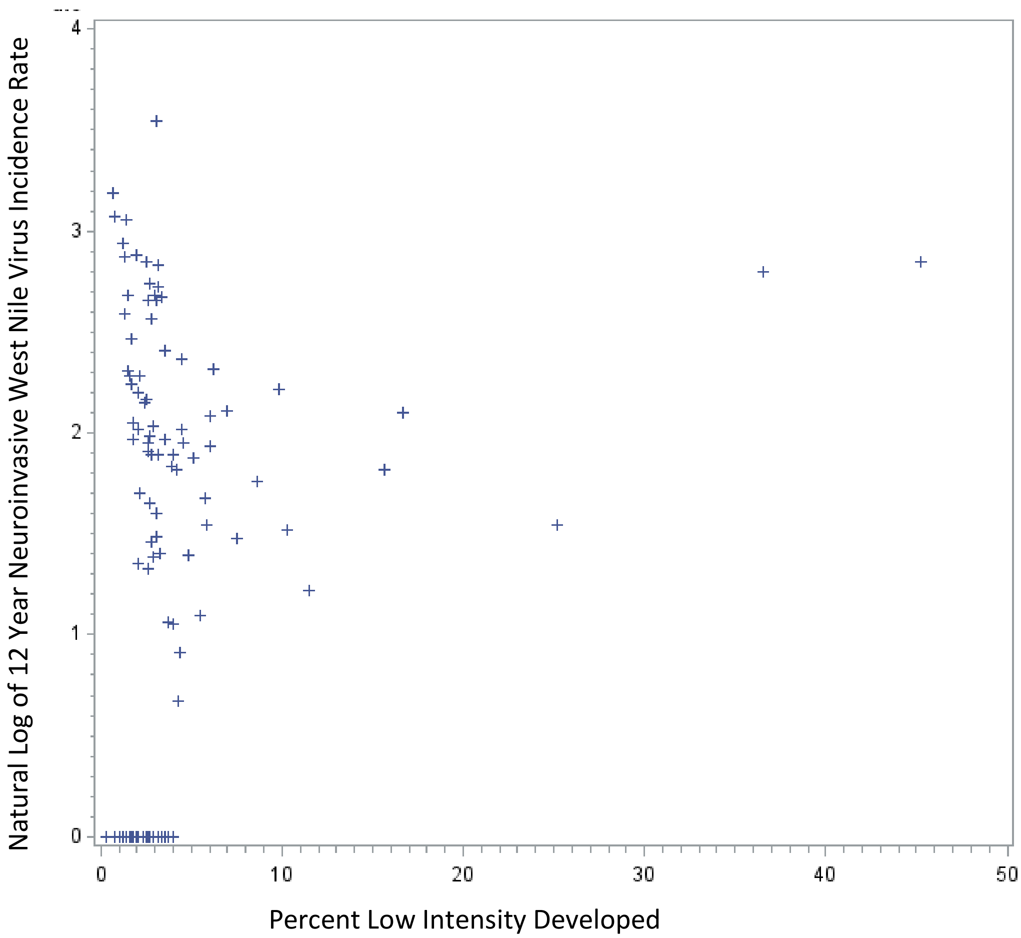
APPENDIX B (CONTINUED)

Figure 15. Scatterplot for Natural Log of 12 Year Neuroinvasive Incidence Rate by Percent Developed Open Space



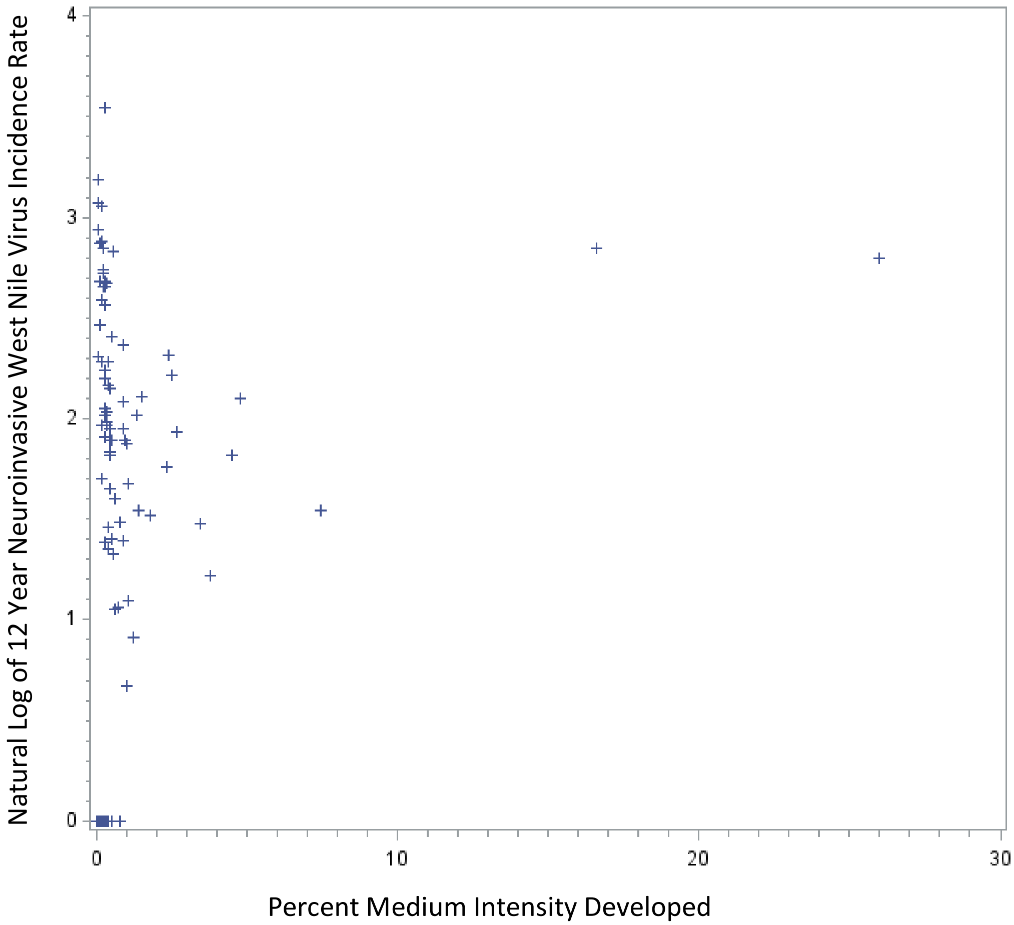
APPENDIX B (CONTINUED)

Figure 16. Scatterplot for Natural Log of 12 Year Neuroinvasive Incidence Rate by Percent Low Intensity Developed Land



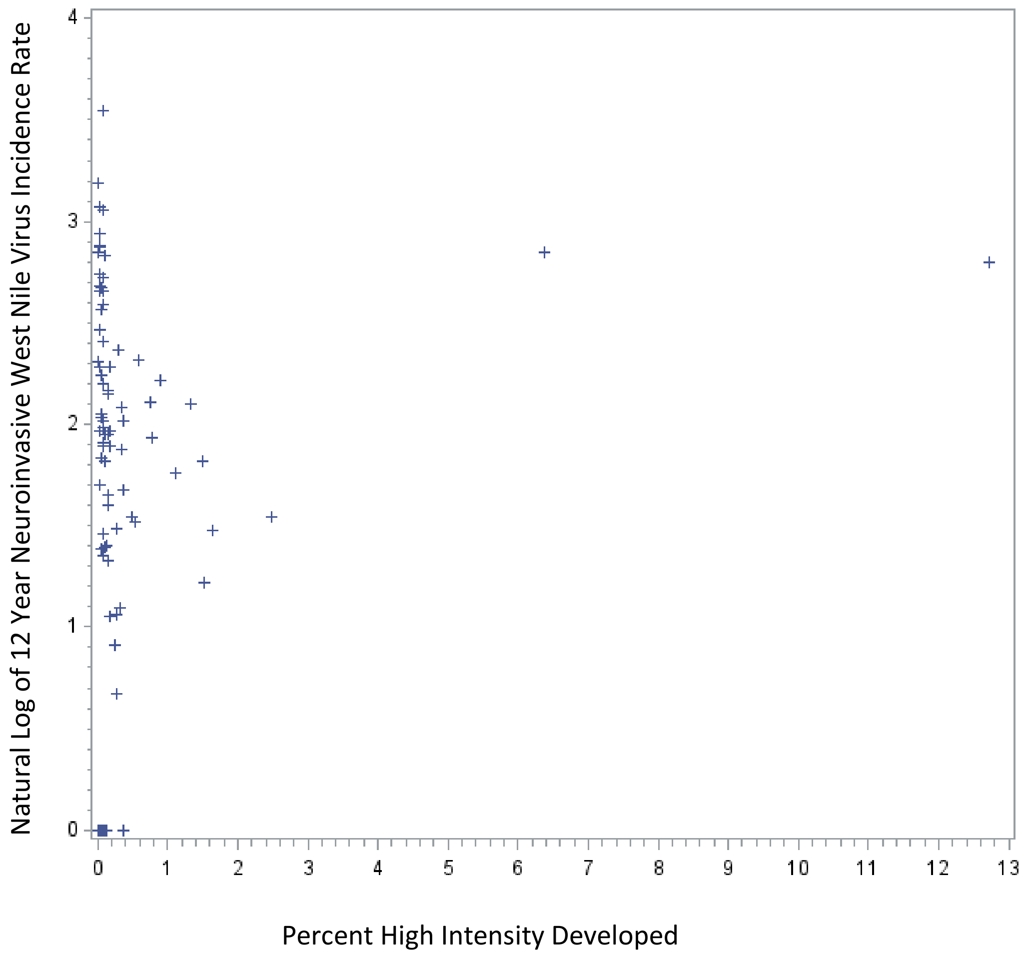
APPENDIX B (CONTINUED)

Figure 17. Scatterplot for Natural Log of 12 Year Neuroinvasive Incidence Rate by Percent Medium Intensity Developed Land



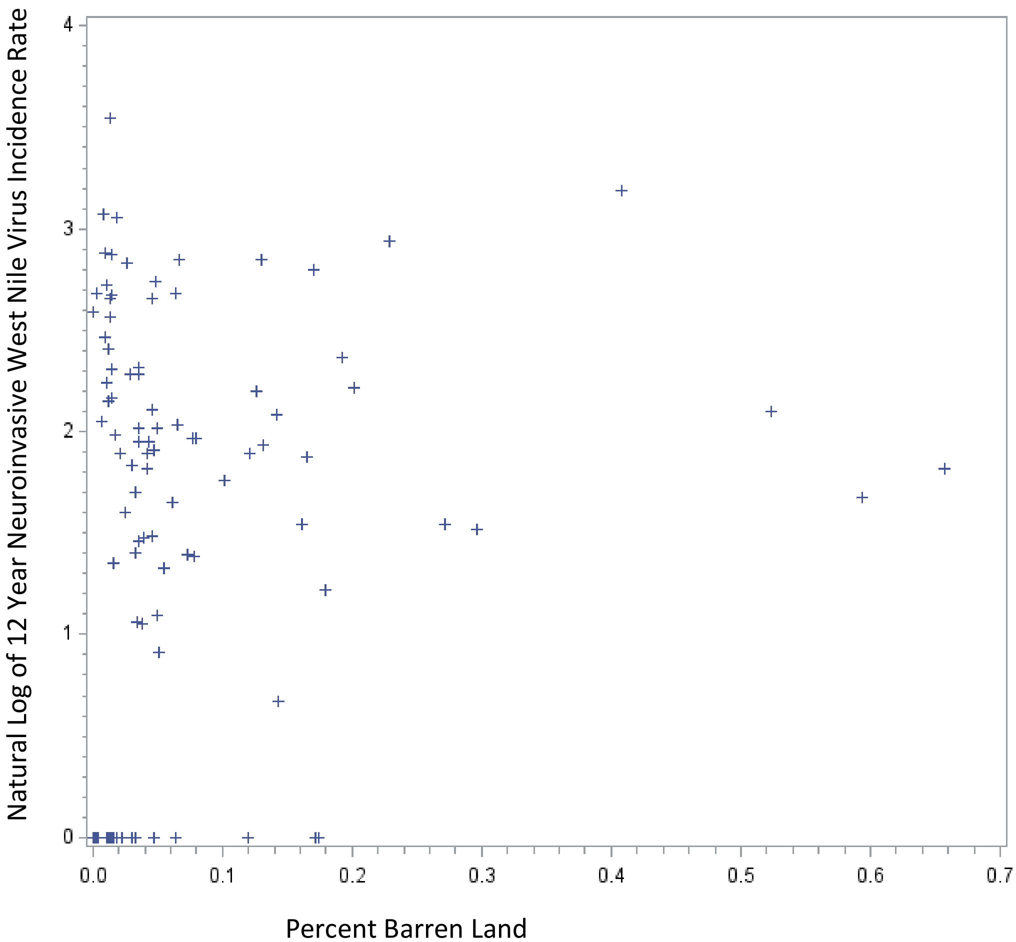
APPENDIX B (CONTINUED)

Figure 18. Scatterplot for Natural Log of 12 Year Neuroinvasive Incidence Rate by Percent High Intensity Developed Land



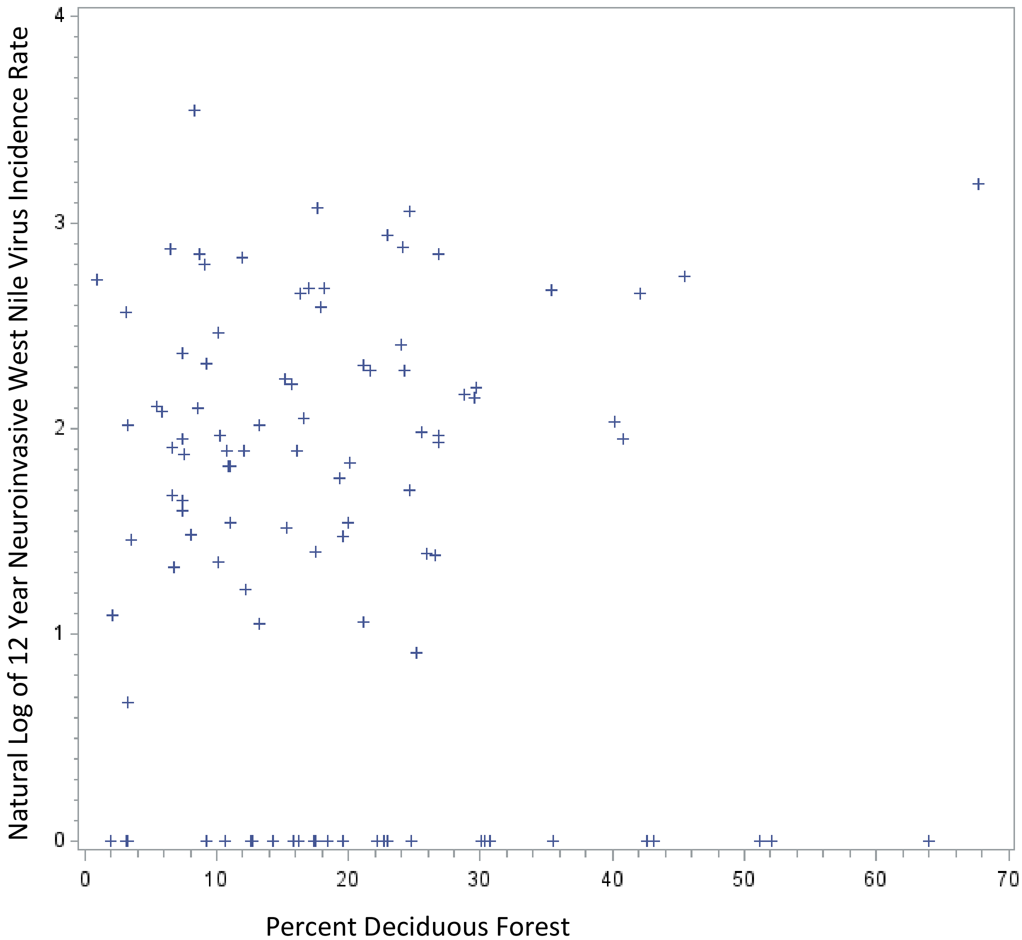
APPENDIX B (CONTINUED)

Figure 19. Scatterplot for Natural Log of 12 Year Neuroinvasive Incidence Rate by Percent Barren Land



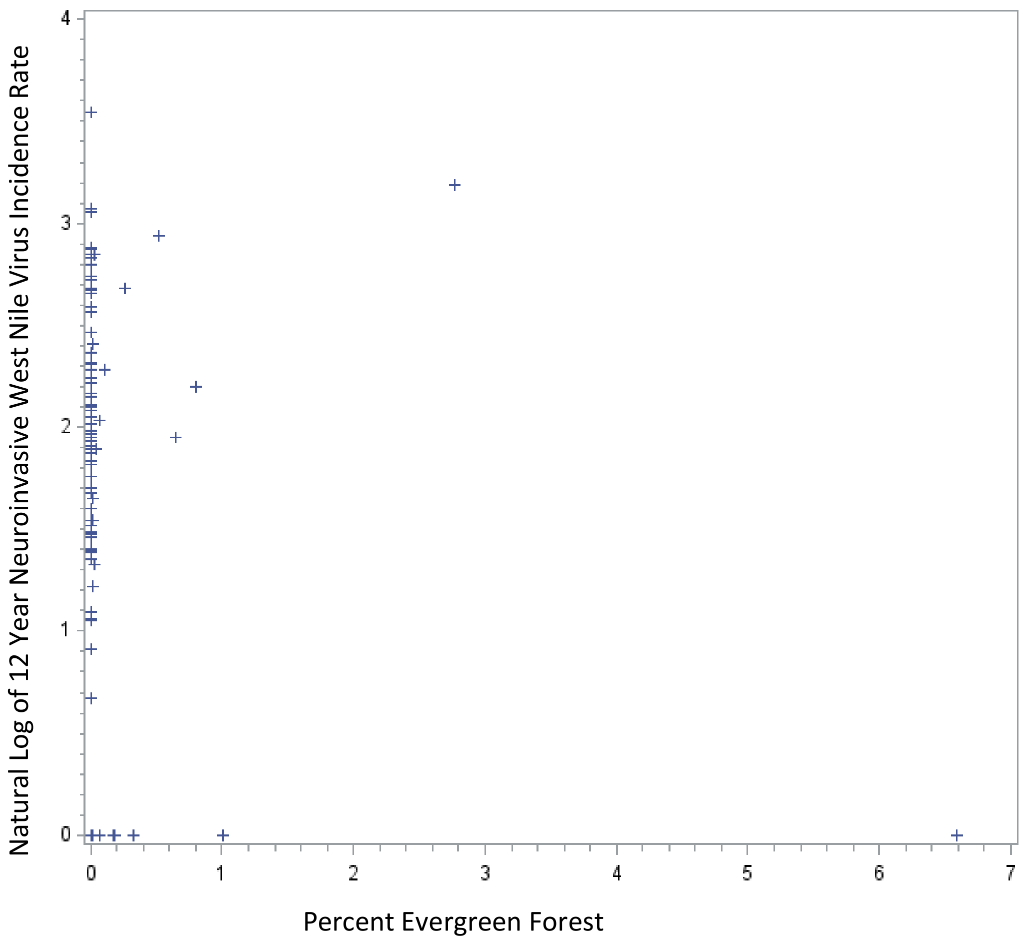
APPENDIX B (CONTINUED)

Figure 20. Scatterplot for Natural Log of 12 Year Neuroinvasive Incidence Rate by Percent Deciduous Forest



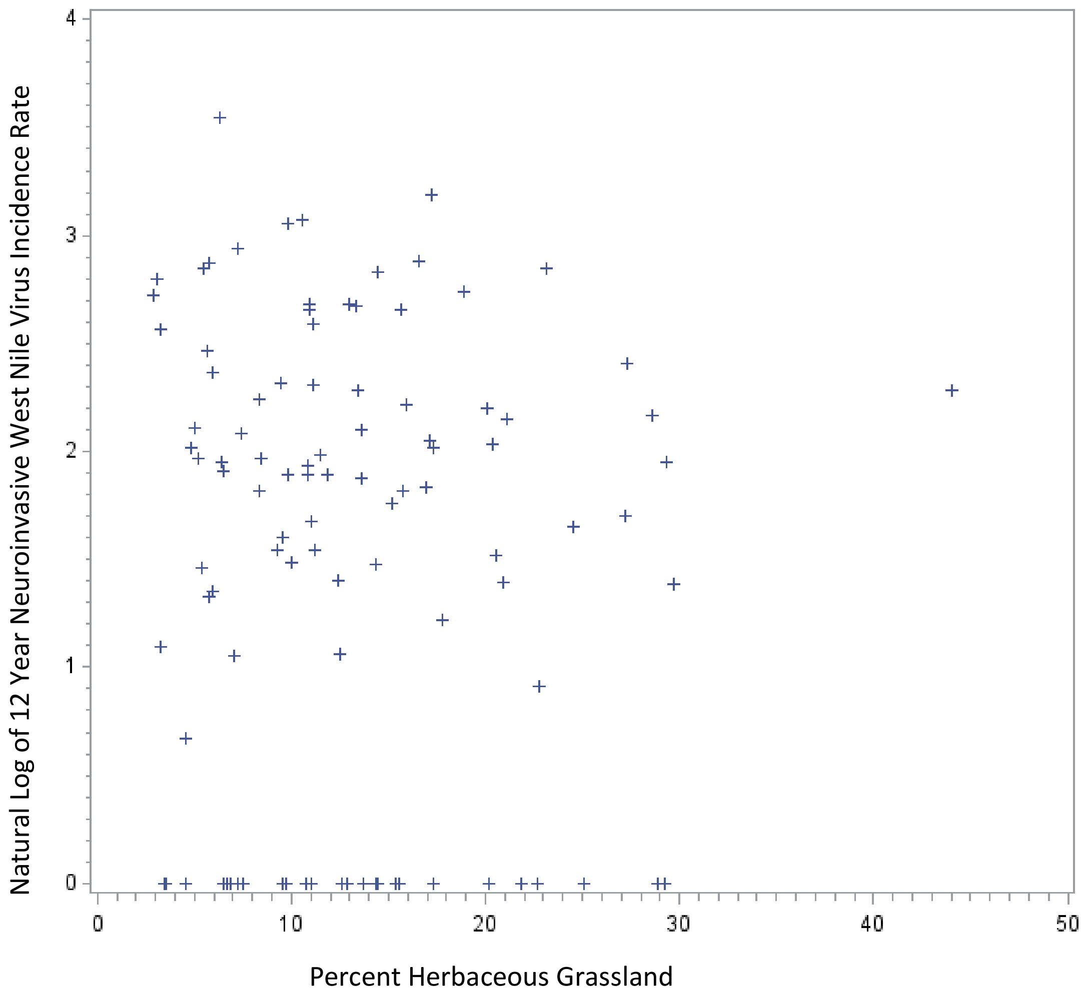
APPENDIX B (CONTINUED)

Figure 21. Scatterplot for Natural Log of 12 Year Neuroinvasive Incidence Rate by Percent Evergreen Forest



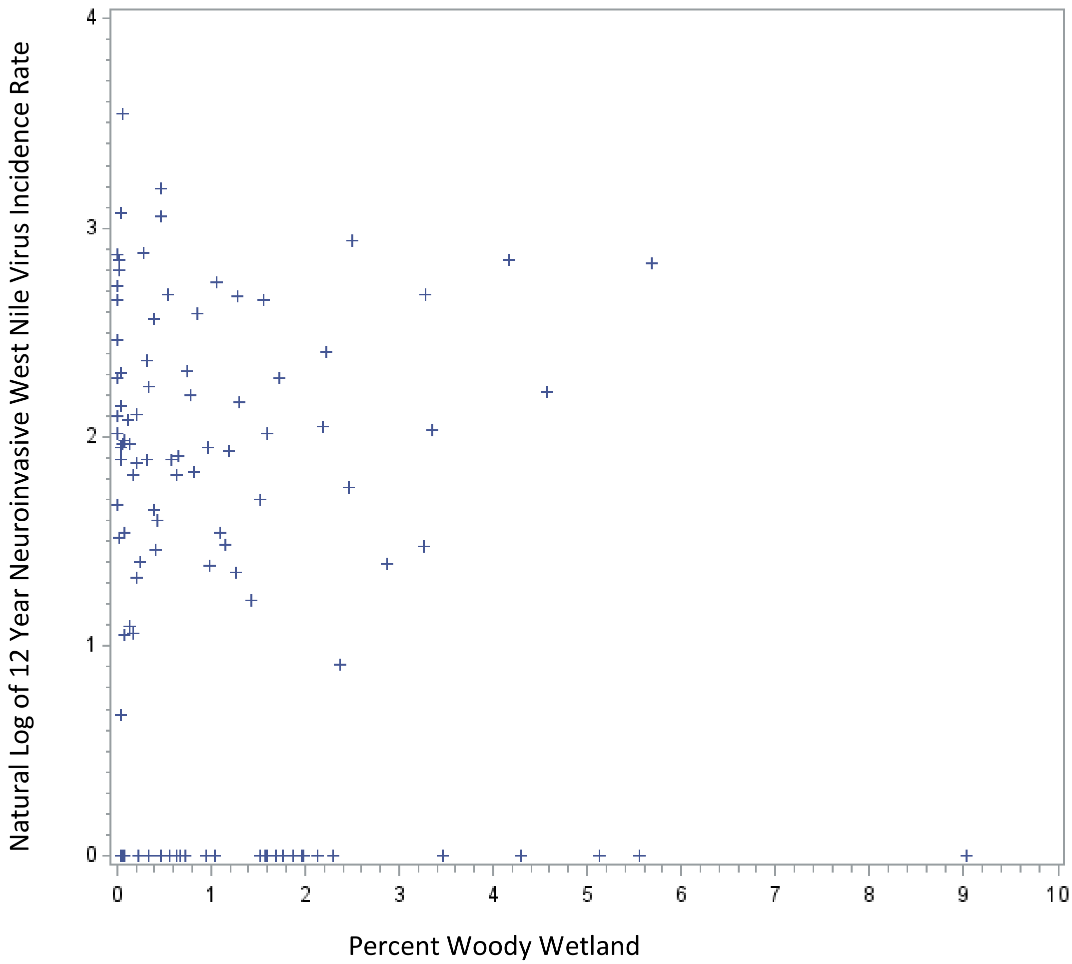
APPENDIX B (CONTINUED)

Figure 22. Scatterplot for Natural Log of 12 Year Neuroinvasive Incidence Rate by Percent Herbaceous Grassland



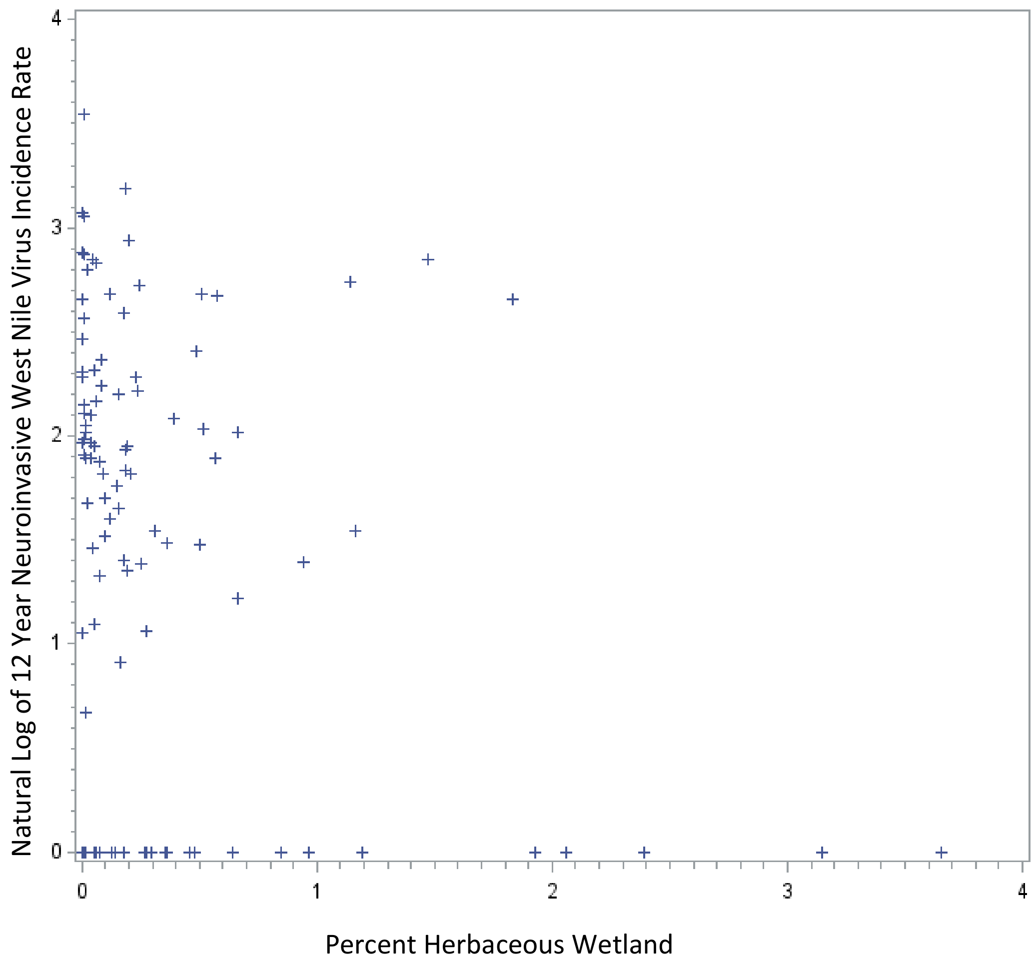
APPENDIX B (CONTINUED)

Figure 23. Scatterplot for Natural Log of 12 Year Neuroinvasive Incidence Rate by Percent Woody Wetland



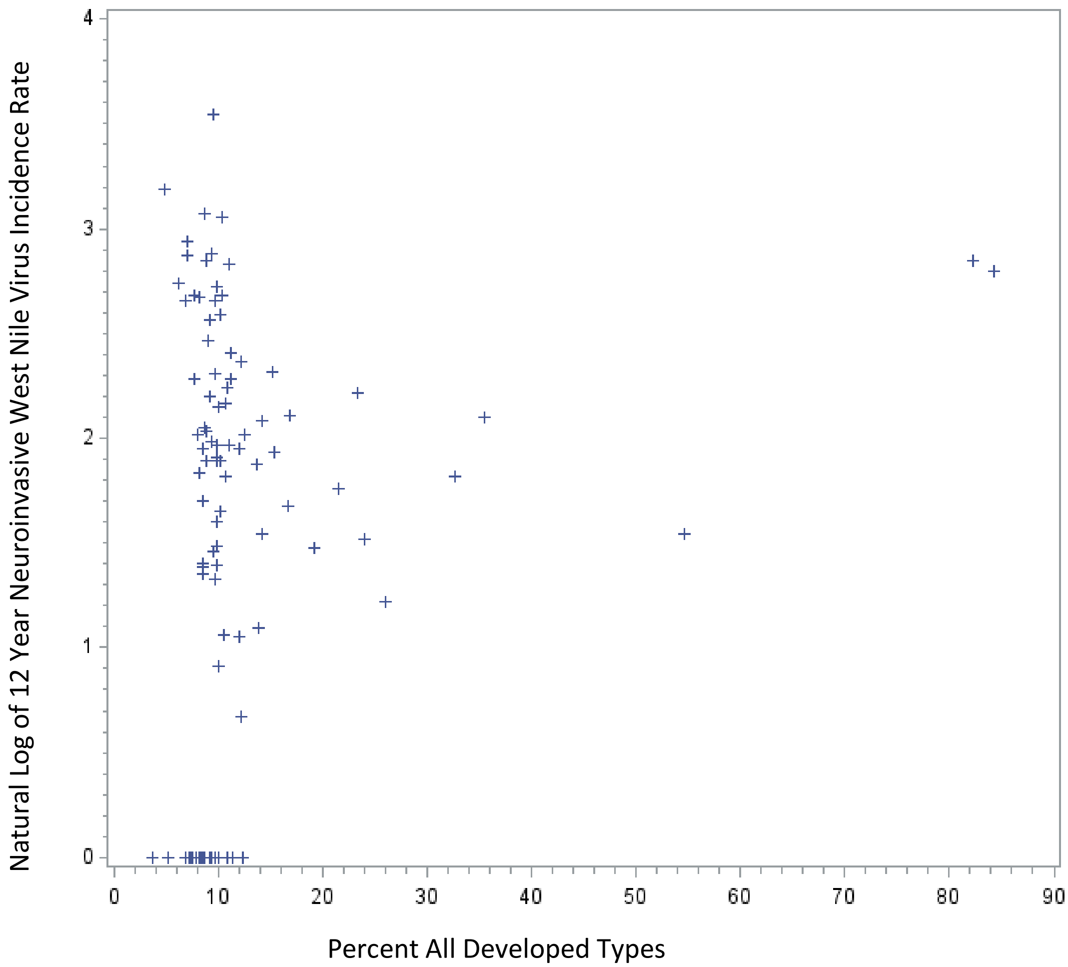
APPENDIX B (CONTINUED)

Figure 24. Scatterplot for Natural Log of 12 Year Neuroinvasive Incidence Rate by Percent Herbaceous Wetland



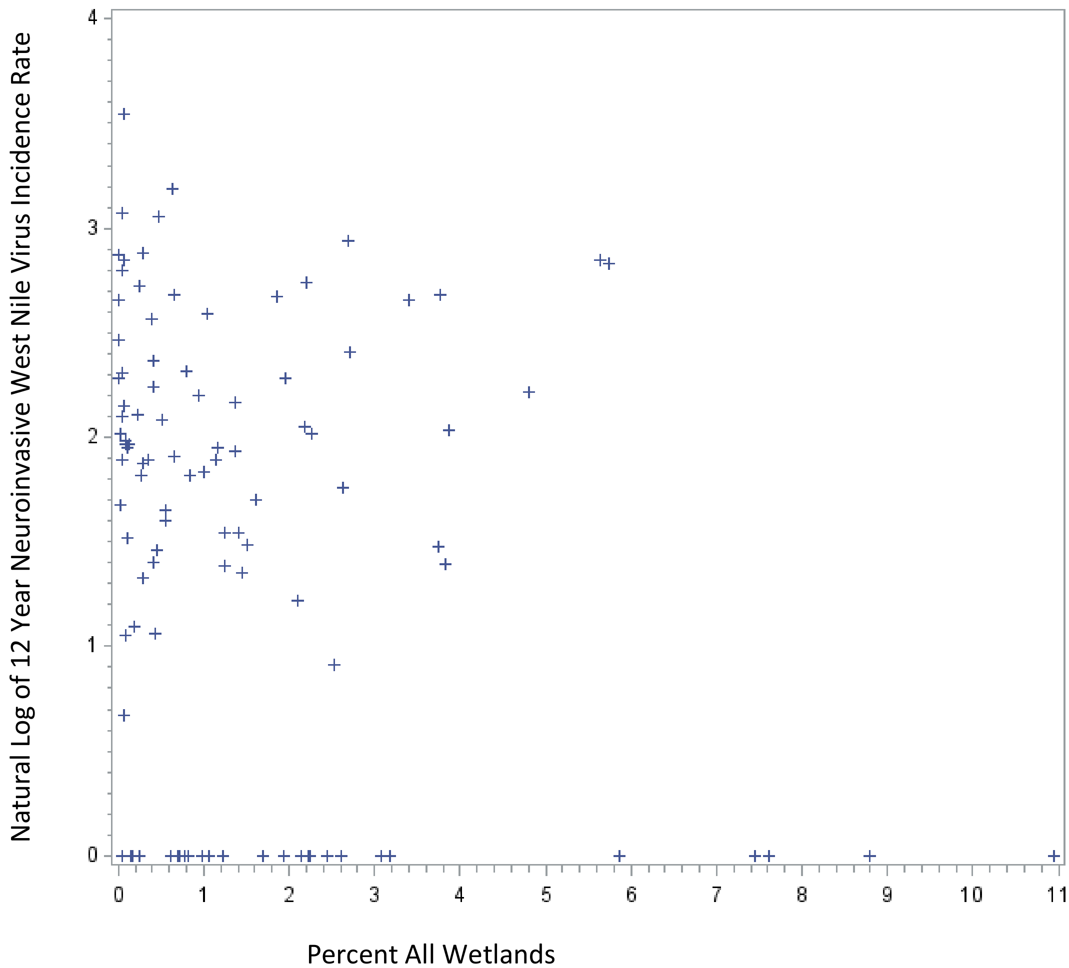
APPENDIX B (CONTINUED)

Figure 25. Scatterplot for Natural Log of 12 Year Neuroinvasive Incidence Rate by Percent All Developed Land



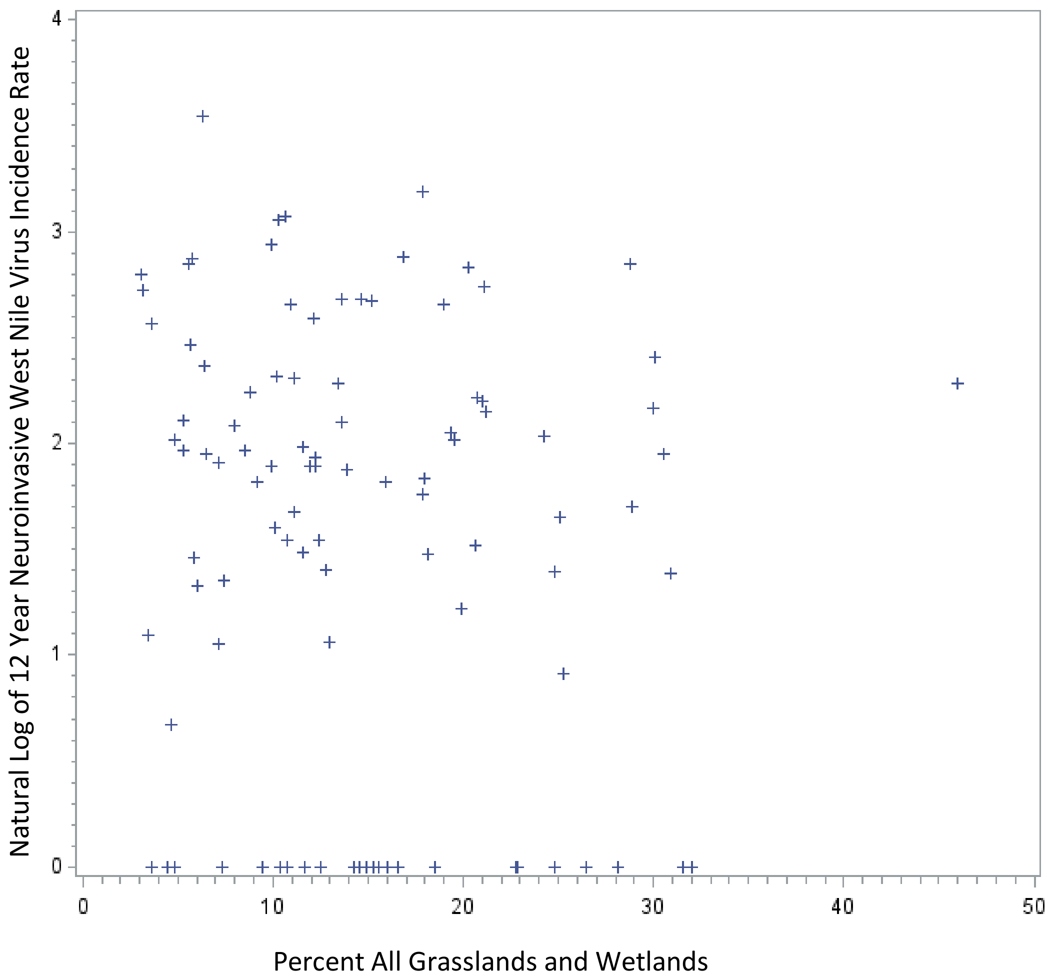
APPENDIX B (CONTINUED)

Figure 26. Scatterplot for Natural Log of 12 Year Neuroinvasive Incidence Rate by Percent Wetland



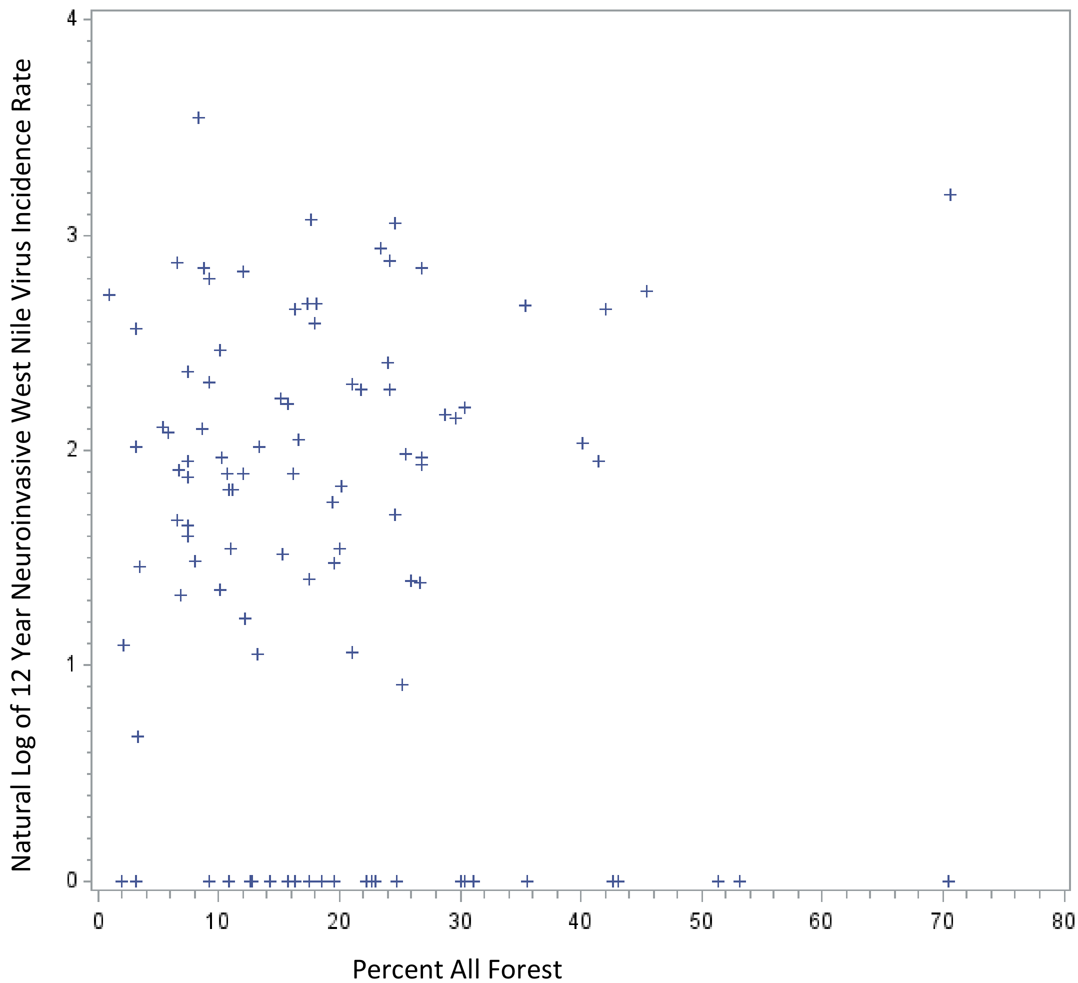
APPENDIX B (CONTINUED)

Figure 27. Scatterplot for Natural Log of 12 Year Neuroinvasive Incidence Rate by Percent All Wetland and Grassland



APPENDIX B (CONTINUED)

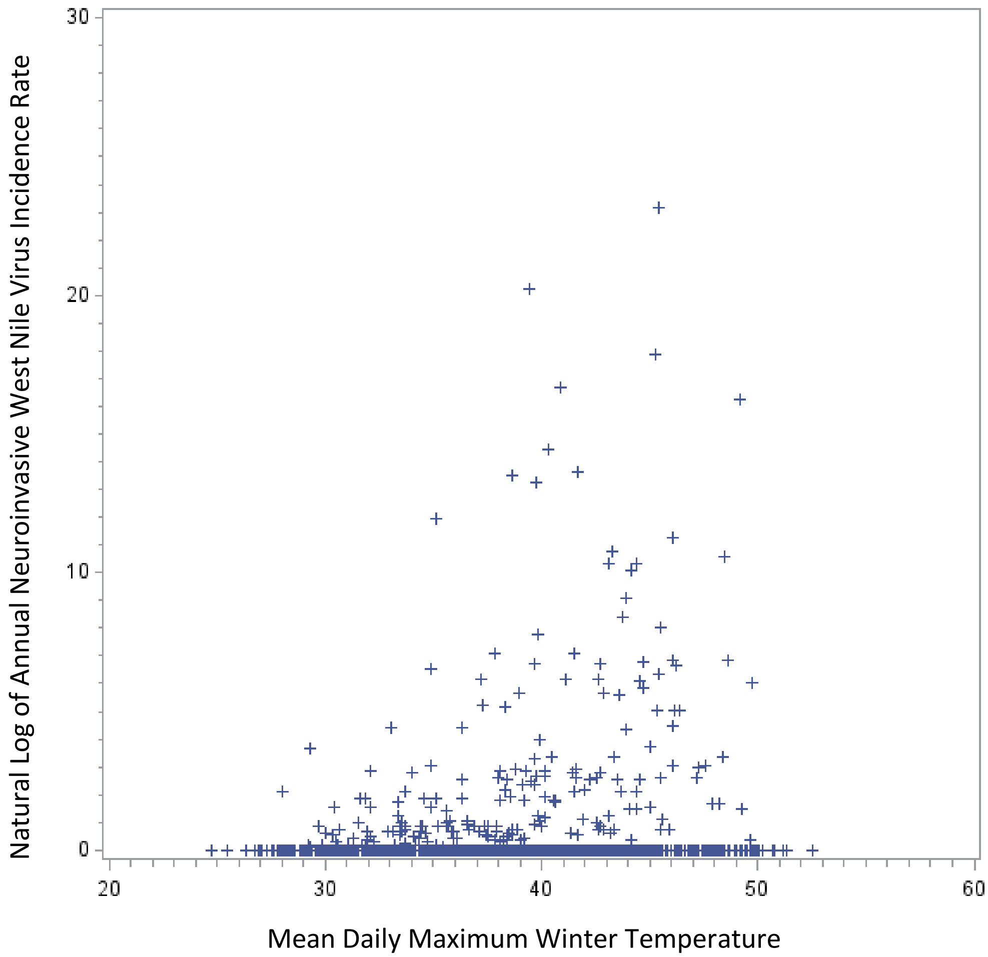
Figure 28. Scatterplot for Natural Log of 12 Year Neuroinvasive Incidence Rate by Percent All Forest



APPENDIX B (CONTINUED)

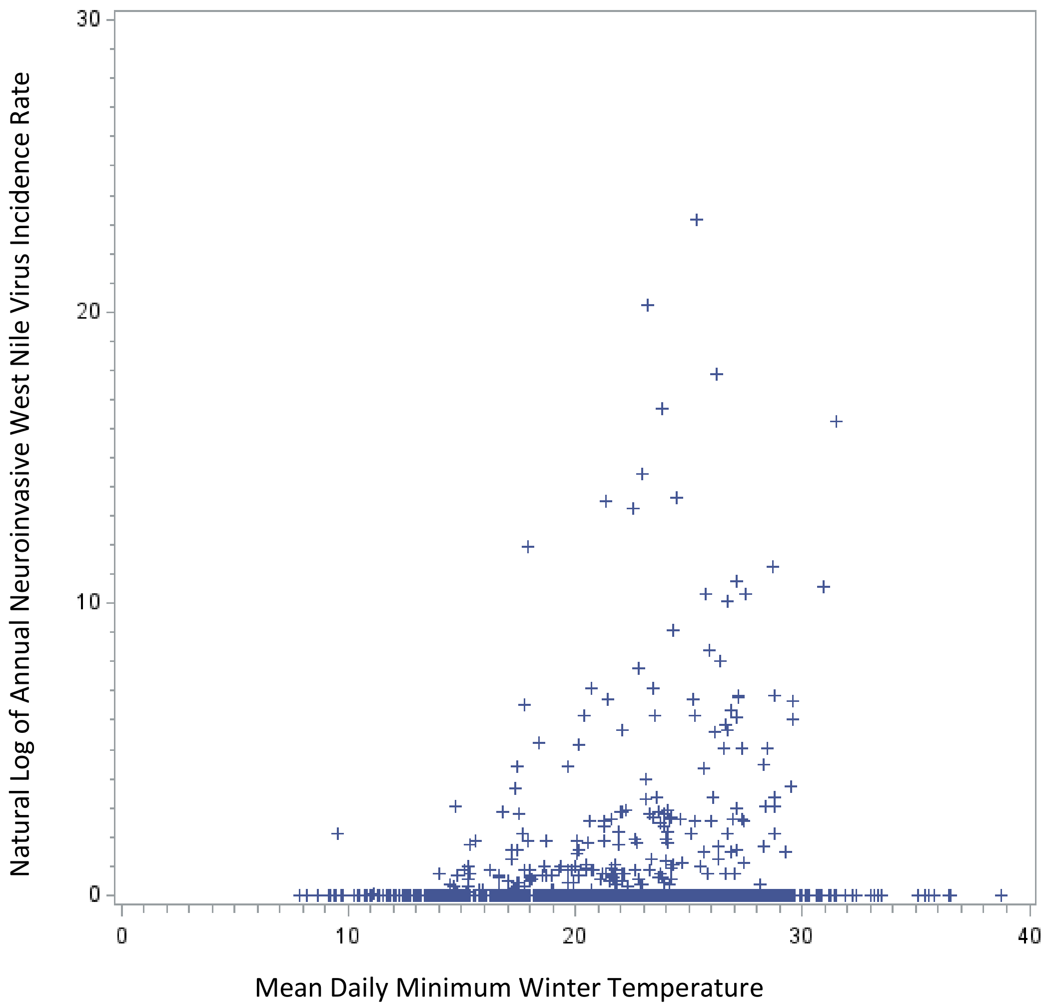
B. Weather Scatterplots

Figure 29. Scatterplot for the Natural Log of Annual Neuroinvasive West Nile Virus Incidence Rates by Mean Daily Maximum Winter Temperature



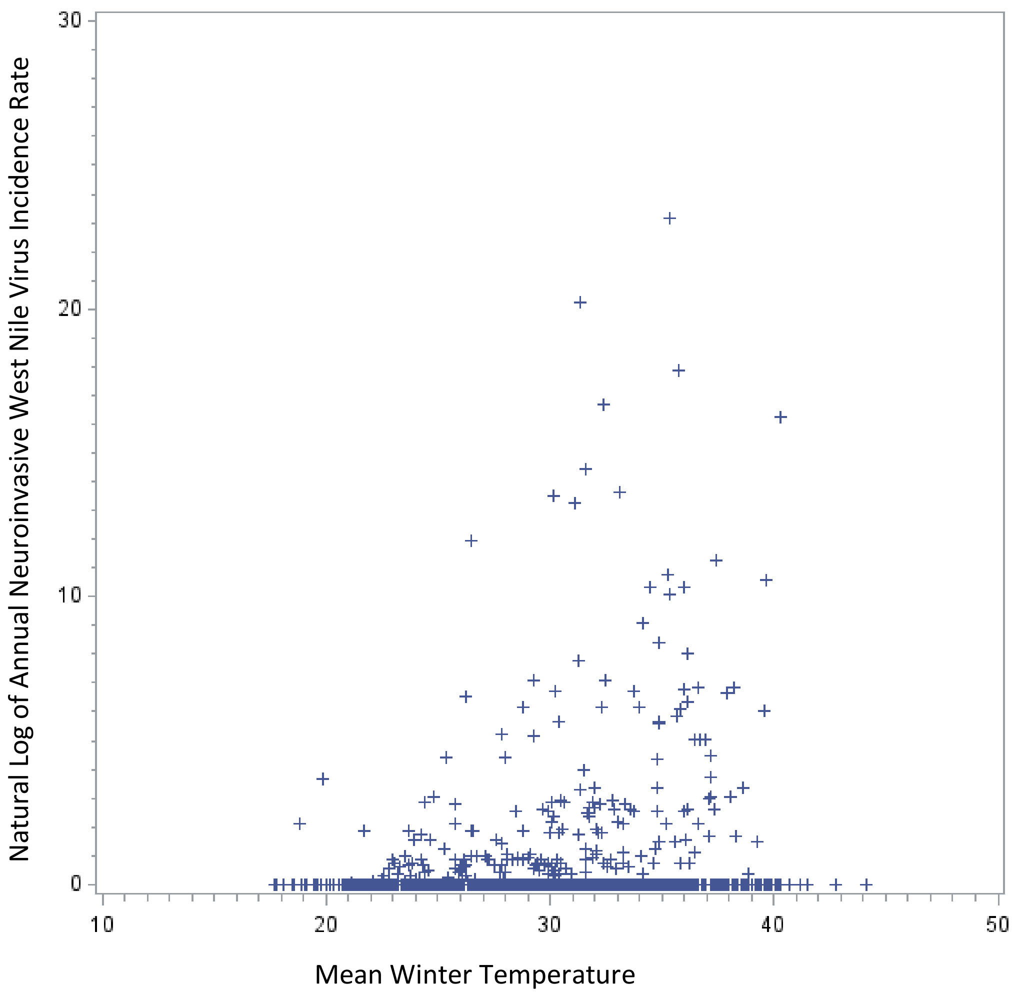
APPENDIX B (CONTINUED)

Figure 30. Scatterplot for the Natural Log of Annual Neuroinvasive West Nile Virus Incidence Rates by Mean Daily Minimum Winter Temperature



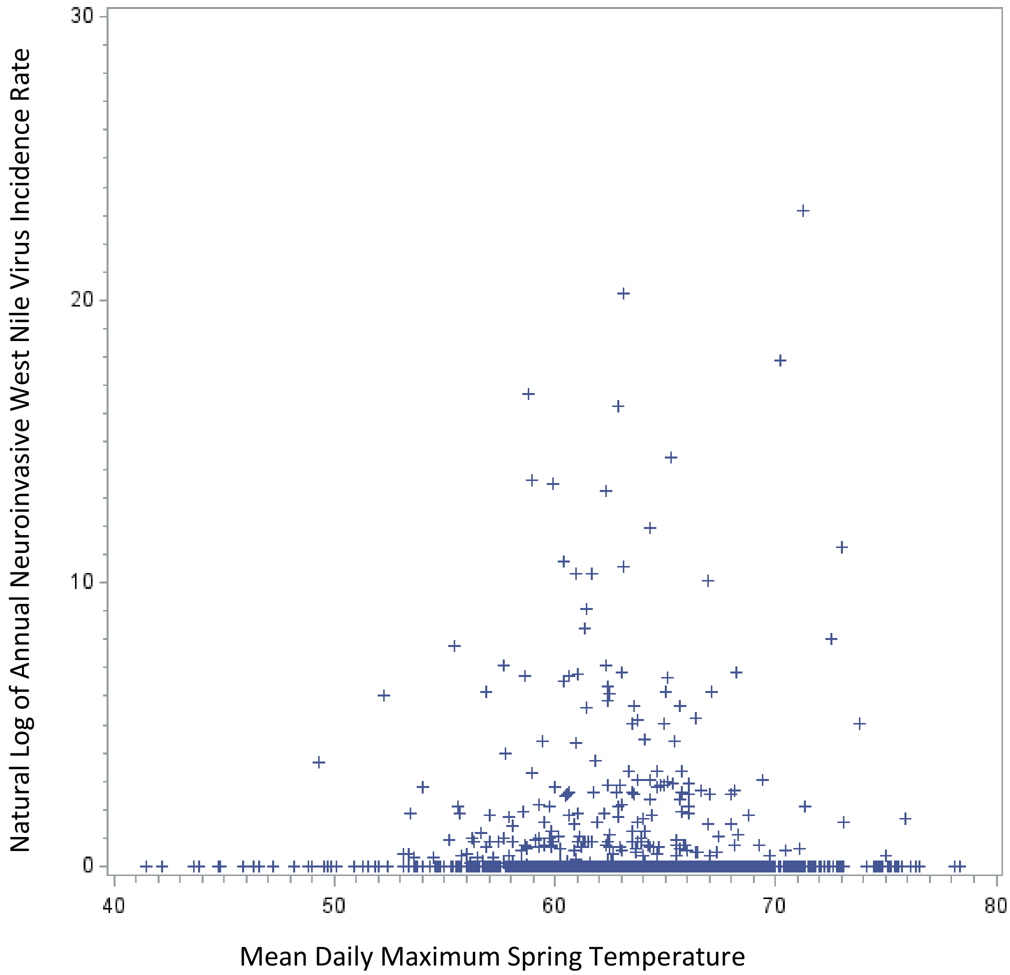
APPENDIX B (CONTINUED)

Figure 31. Scatterplot for the Natural Log of Annual Neuroinvasive West Nile Virus Incidence Rates by Mean Winter Temperature



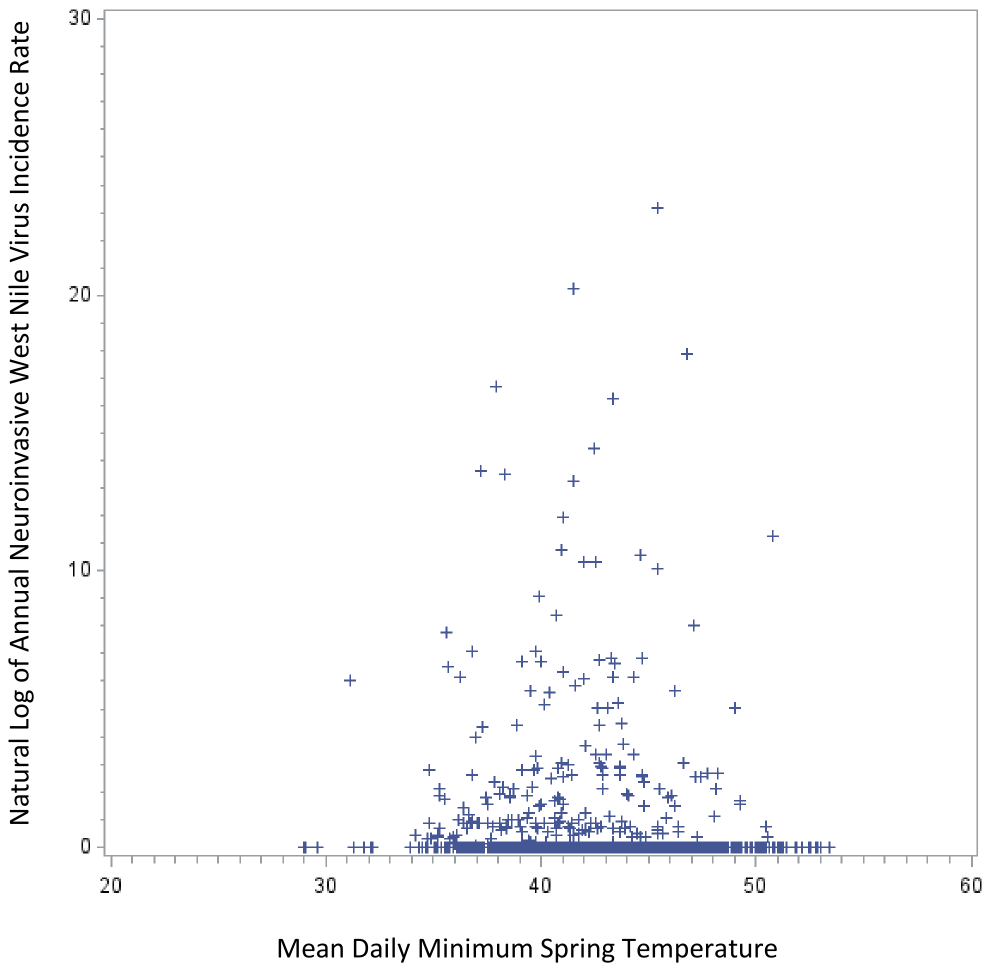
APPENDIX B (CONTINUED)

Figure 32. Scatterplot for the Natural Log of Annual Neuroinvasive West Nile Virus Incidence Rates by Mean Daily Maximum Spring Temperature



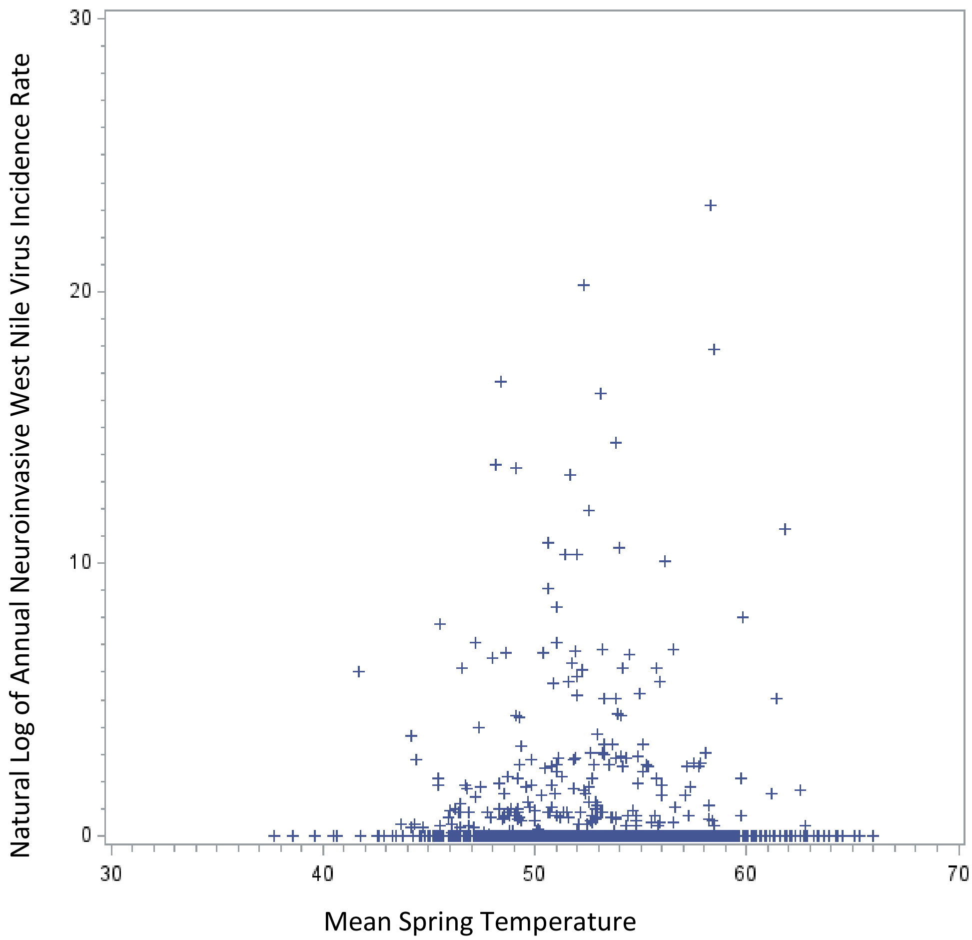
APPENDIX B (CONTINUED)

Figure 33. Scatterplot for the Natural Log of Annual Neuroinvasive West Nile Virus Incidence Rates by Mean Daily Minimum Spring Temperature.



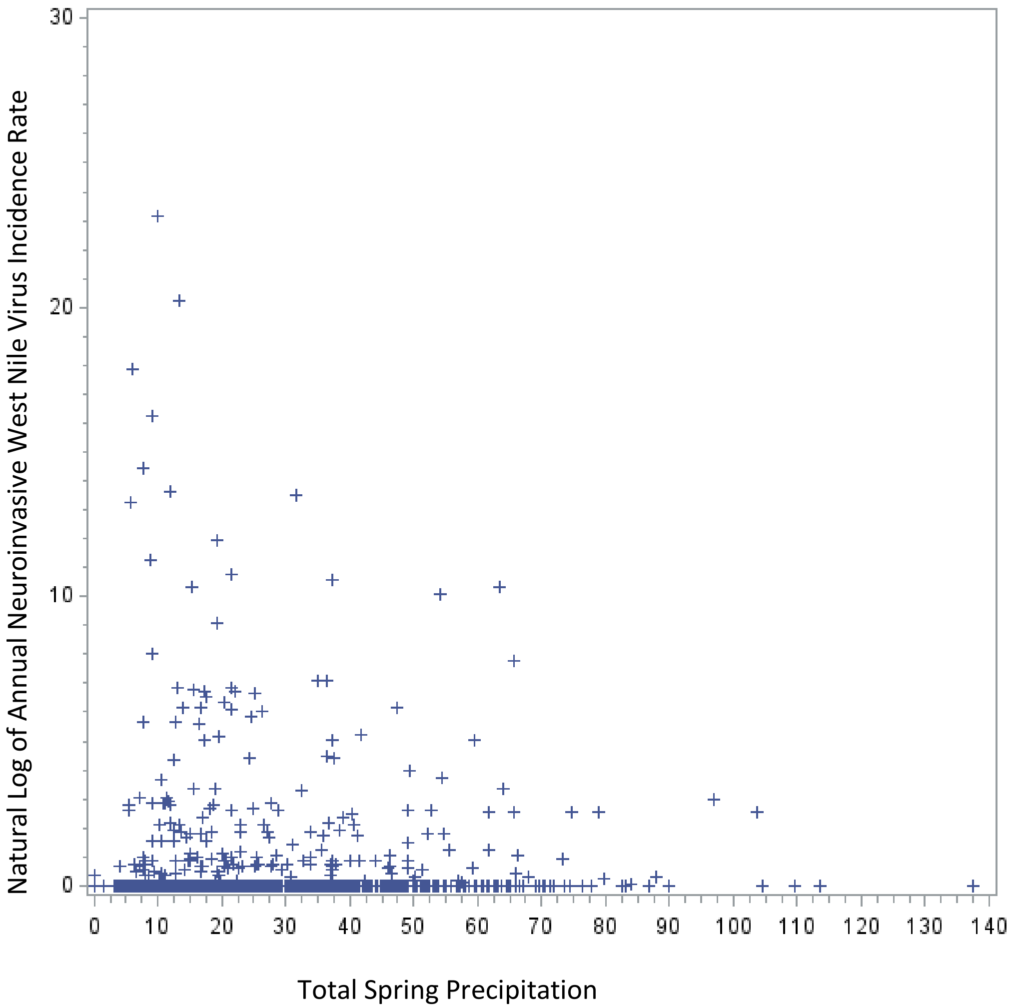
APPENDIX B (CONTINUED)

Figure 34. Scatterplot for the Natural Log of Annual Neuroinvasive West Nile Virus Incidence Rates by Mean Spring Temperature



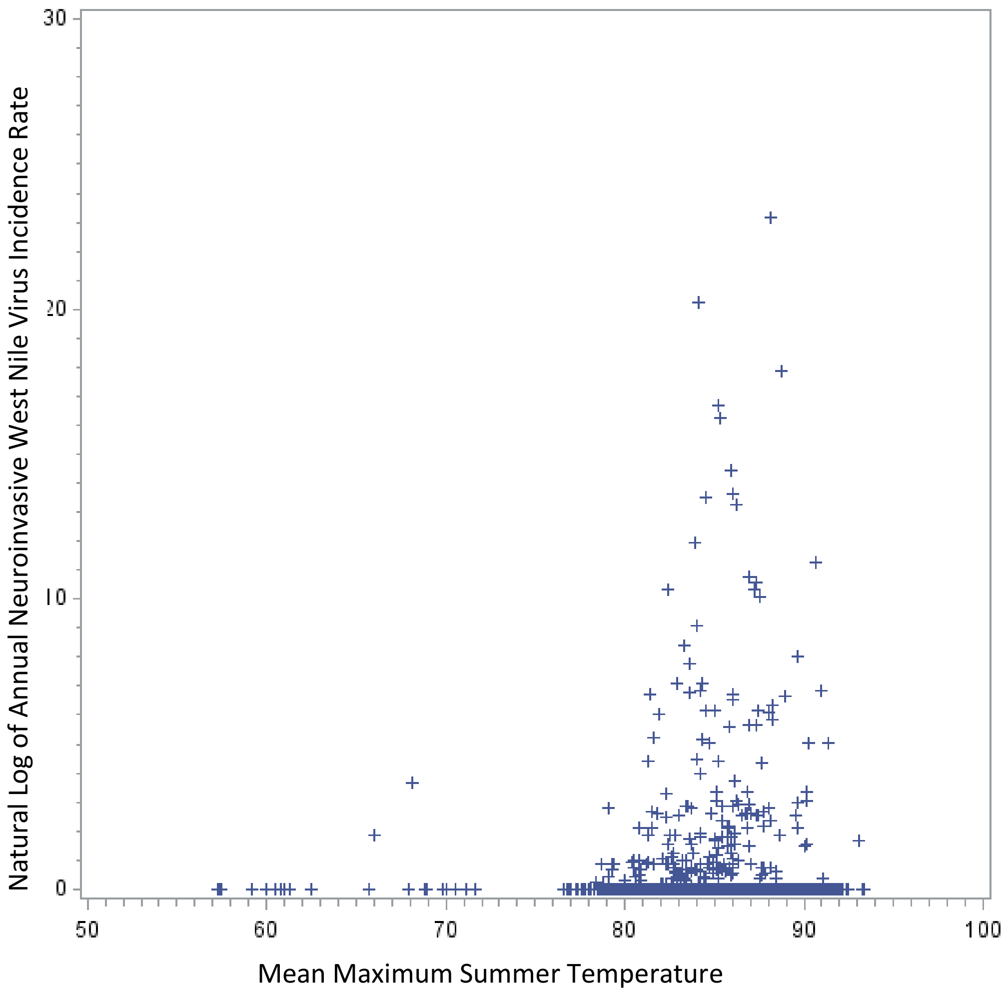
APPENDIX B (CONTINUED)

Figure 35. Scatterplot for the Natural Log of Annual Neuroinvasive West Nile Virus Incidence Rates by Spring Precipitation



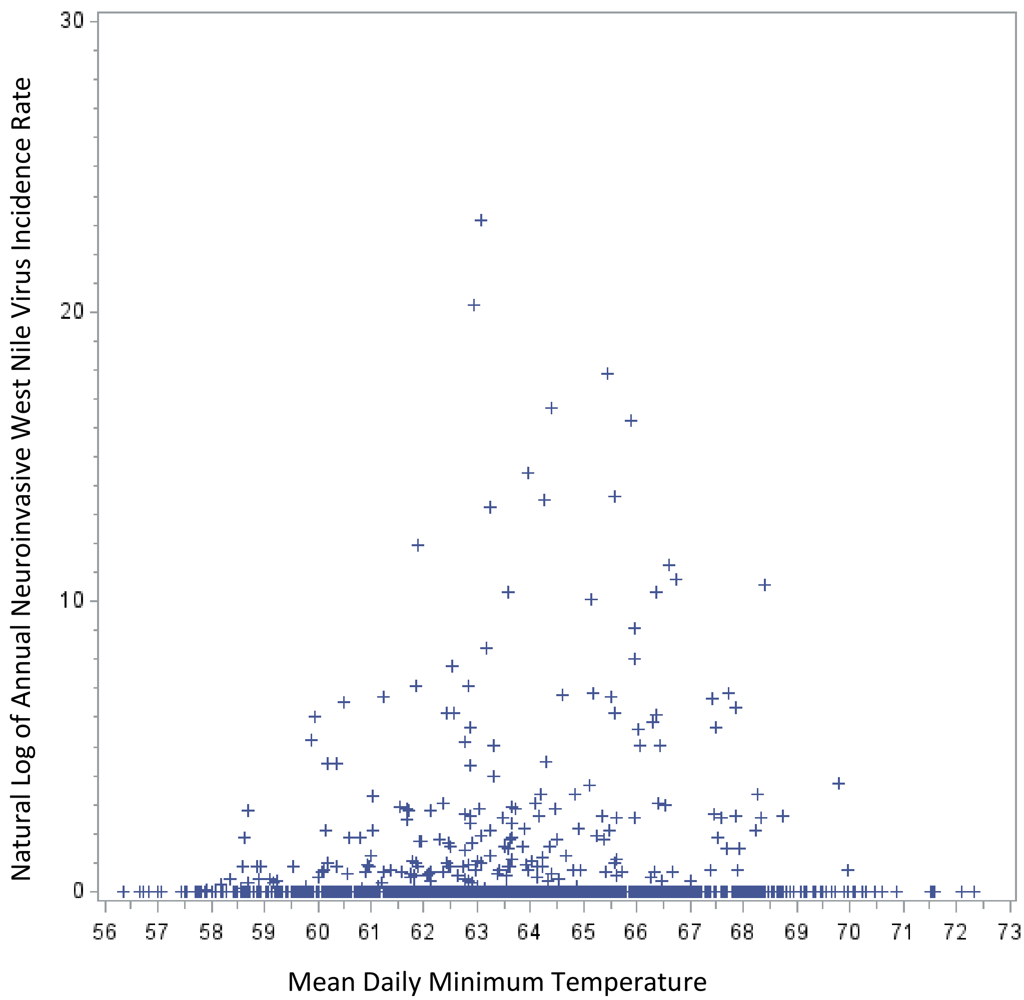
APPENDIX B (CONTINUED)

Figure 36. Scatterplot for the Natural Log of Annual Neuroinvasive West Nile Virus Incidence Rates by Mean Daily Maximum Summer Temperature



APPENDIX B (CONTINUED)

Figure 37. Scatterplot for the Natural Log of Annual Neuroinvasive West Nile Virus Incidence Rates by Mean Daily Minimum Summer Temperature



APPENDIX B (CONTINUED)

Figure 38. Scatterplot for the Natural Log of Annual Neuroinvasive West Nile Virus Incidence Rates by Mean Summer Temperature

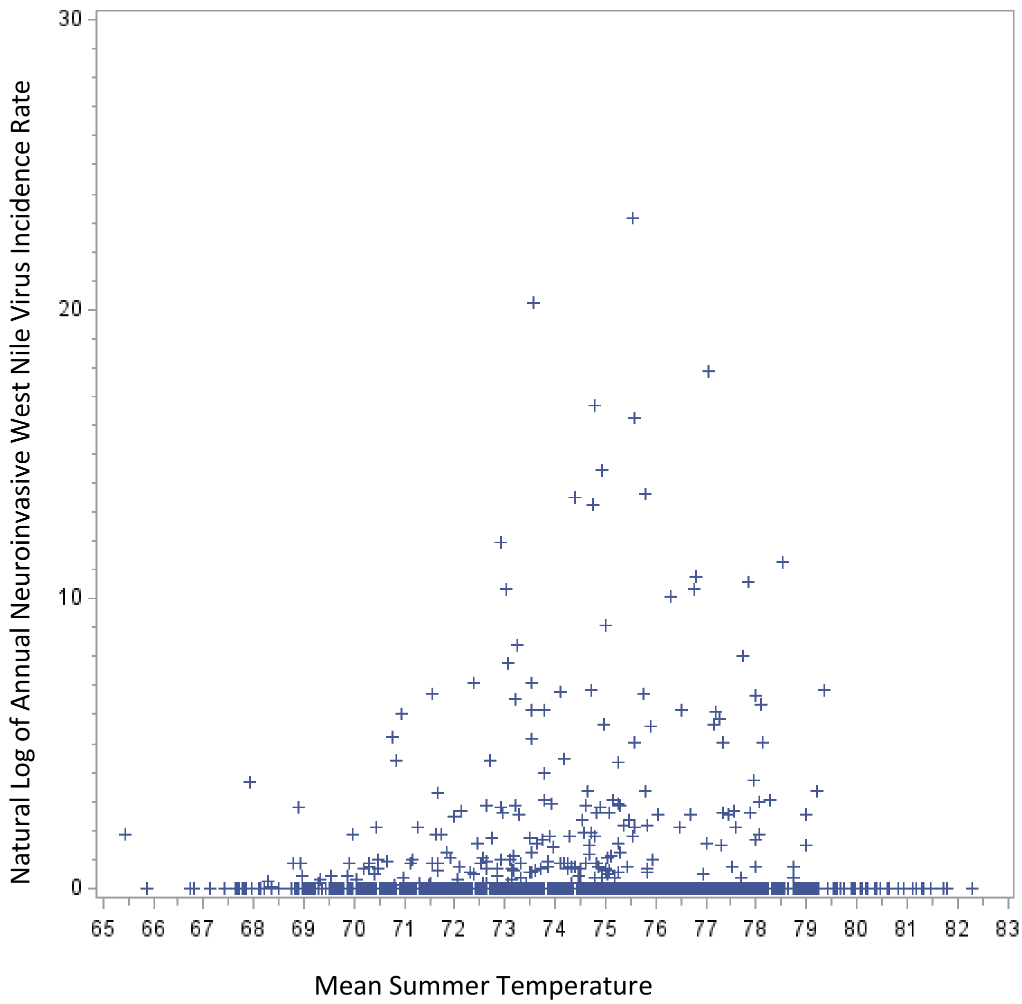
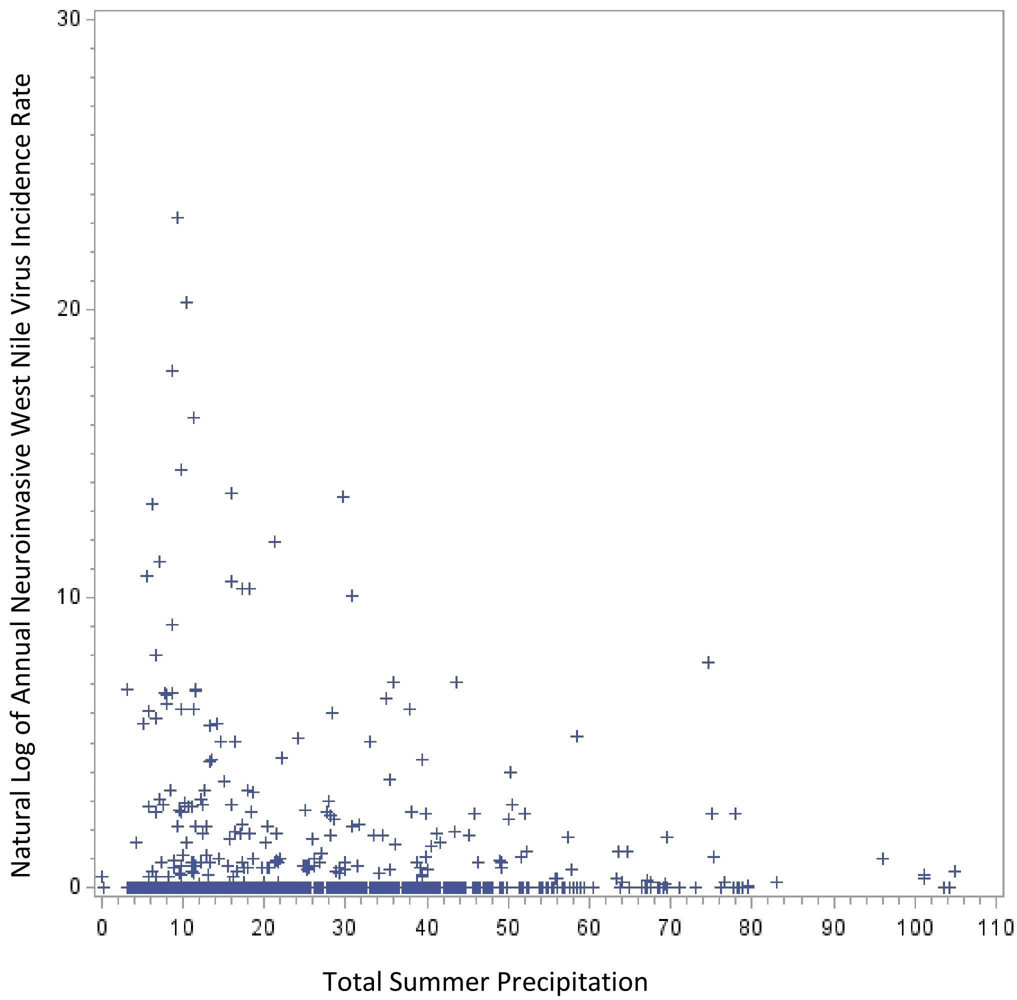
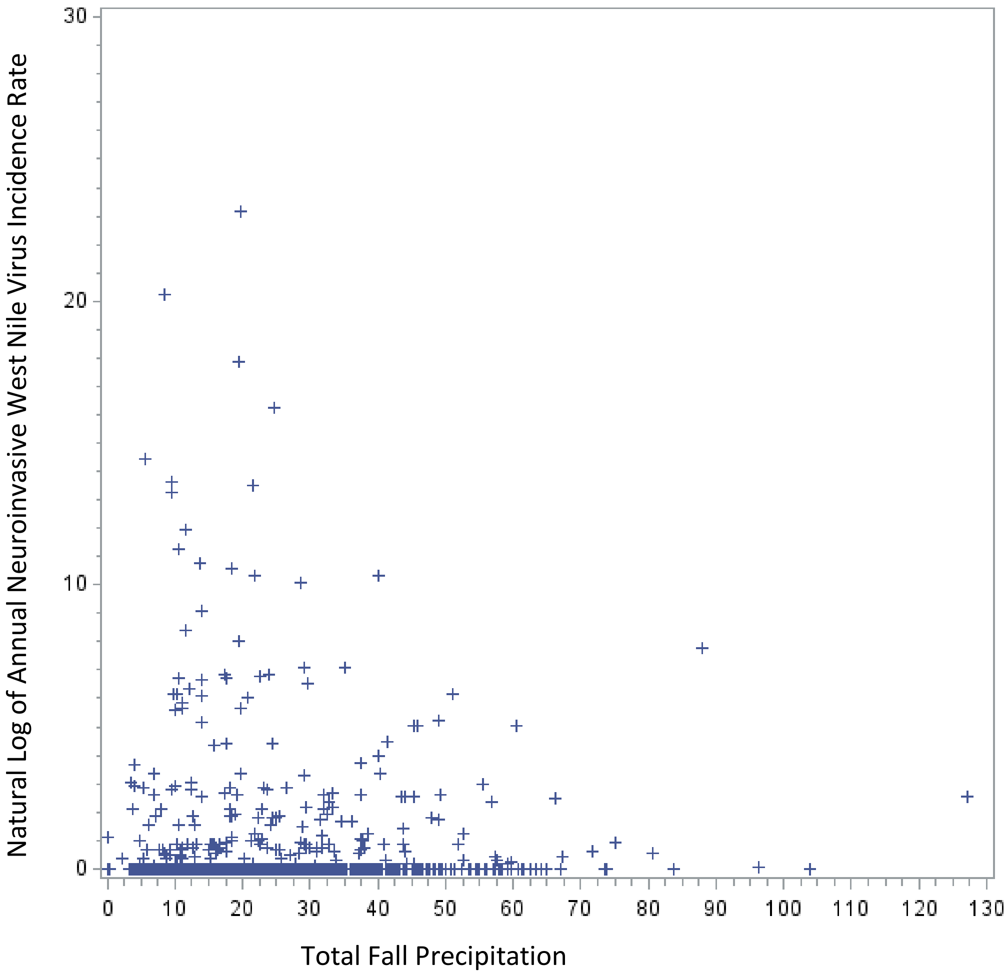


Figure 39. Scatterplot for the Natural Log of Annual Neuroinvasive West Nile Virus Incidence Rates by Total Summer Precipitation



APPENDIX B (CONTINUED)

Figure 40. Scatterplot for the Natural Log of Annual Neuroinvasive West Nile Virus Incidence Rates by Total Fall Precipitation



Appendix C

A. Land Cover Odds Ratio – Two Dichotomous Variables

TABLE XXII. ODDS RATIO FOR THE EFFECT OF LAND COVER ON 2002-2013 INCIDENCE RATES OF NEUROINVASIVE WEST NILE VIRUS IN ILLINOIS USING DICHOTOMOUS EXPOSURE AND OUTCOME VARIABLES

	OR	95% CI		p-value
In a Micropolitan or Metropolitan CBSA	0.9	0.4	2.1	0.894
Larger Elevation Range	0.6	0.3	1.5	0.316
High Peak Elevation	0.6	0.3	1.4	0.276
High Lowest Elevation	1.2	0.5	2.6	0.697
High Percent Evergreen Forest	0.9	0.3	2.9	0.875
High Percent Herbaceous Grassland	1.0	0.4	2.3	0.928
High Percent Herbaceous Wetland	0.3	0.1	0.7	0.006
High Percent Barren Land	1.4	0.6	3.7	0.458
High Percent Deciduous Forest	0.9	0.4	1.9	0.697
High Percent Non-Crop Agriculture	0.8	0.4	1.8	0.612
High Percent Open Water	0.7	0.3	1.6	0.469
High Percent Wooded Wetlands	0.4	0.2	0.8	0.015
High Percent All Crops	1.3	0.6	2.8	0.552
High Total Population Count Over 50	1.4	0.6	3.0	0.414
High Population Density	1.5	0.7	3.3	0.321
High Percent Open Space Developed	1.8	0.8	4.0	0.151
High Percent Low Intensity Developed Land	1.1	0.5	2.5	0.733
High Percent Medium Intensity Developed Land	0.8	0.4	1.7	0.552
High Percent High Intensity Developed Land	0.8	0.4	1.9	0.679

APPENDIX C (CONTINUED)

B. Weather Odds Ratio – Two Dichotomous Variables

TABLE XXIII. ODDS RATIO FOR THE EFFECT OF TEMPERATURE AND PRECIPITATION ON 2002-2013 INCIDENCE RATES OF NEUROINVASIVE WEST NILE VIRUS IN ILLINOIS USING DICHOTOMOUS EXPOSURE AND OUTCOME VARIABLES

Spring	Odds Ratio	95% Confidence		p-value
Cold spring average daily mean temperature	1.3	0.9	2.0	0.211
Warm spring average daily mean temperature	N/A			
Above average spring average daily mean temperature	0.7	0.5	0.9	0.014
Above the median total precipitation for spring	1.4	0.9	2.3	0.162
Extreme low total precipitation in spring	2.2	0.9	5.7	0.079
Extreme high precipitation in spring	1.0	0.7	1.4	0.904
Summer				
Cold summer average daily mean temperature	1.3	0.9	2.0	0.158
Warm summer average daily mean temperature	0.9	0.3	3.0	0.846
Above average summer average daily mean temperature	1.5	1.1	2.0	0.022
Above the median total precipitation for summer	0.8	0.6	1.1	0.151
Extreme low total precipitation in summer	0.7	0.5	1.0	0.068
Extreme high precipitation in summer	0.9	0.6	1.3	0.458
Fall				
Cold fall average daily mean temperature	1.3	0.8	2.1	0.242
Warm fall average daily mean temperature	3.6	0.3	40.0	0.265
Above average fall average daily mean temperature	0.9	0.6	1.3	
Above the median total precipitation for fall	1.1	0.7	1.8	0.733
Extreme low total precipitation in fall	1.9	0.8	4.9	0.156
Extreme high precipitation in fall				
Winter				
Cold winter average daily mean temperature	2.0	1.3	3.0	0.001
Warm winter average daily mean temperature	2.5	1.7	3.8	<.0001
Above average winter average daily mean temperature	3.3	2.3	4.7	<.0001

Appendix D

TABLE XXIV. DISTRIBUTION OF AGE OF THE POPULATION FOR COUNTIES IN ILLINOIS FROM 2010 UNITED STATES CENSUS, STRATIFIED BY URBAN AND RURAL COUNTIES

Urban and Suburban Counties							
	N	Mean	Lower 95% CL for Mean	Upper 95% CL for Mean	Median	Minimum	Maximum
Percent of Population Over 50	793	35.4	35.1	35.7	36.7	21.8	45.0
Percent of Population Over 60	793	21.1	20.8	21.4	22.1	11.2	28.2
Percent of Population Over 70	793	10.9	10.7	11.0	11.3	4.6	14.3
Percent of Population Over 80	793	4.6	4.5	4.6	4.7	1.7	6.7
Rural Counties							
	N	Mean	Lower 95% CL for Mean	Upper 95% CL for Mean	Median	Minimum	Maximum
Percent of Population Over 50	456	39.3	39.0	39.5	39.4	29.6	45.6
Percent of Population Over 60	456	24.4	24.2	24.7	24.4	16.6	29.6
Percent of Population Over 70	456	12.9	12.8	13.0	12.9	9.0	15.5
Percent of Population Over 80	456	5.3	5.2	5.4	5.4	3.5	6.7

APPENDIX D (CONTINUED)

TABLE XXV. SENSITIVITY ANALYSIS COMPARING THE EFFECT OF PERCENT OF POPULATION OVER 50 VERSUS PERCENT OF POPULATION OVER 60

	Percent over 50			**Percent over 60**		
	β Estimate	Standard Error	p-value	β Estimate	Standard Error	p-value
Intercept	-113.85	46.29	0.014	-114.76	46.18	0.013
RUCC 1 - 4	-1.02	0.21	<.0001	-1.02	0.21	<.0001
RUCC 5-9 – Reference	0.00	0.00	.	0.00	0.00	.
Percent of Population Over 50 or 60	0.09	0.02	<.0001	0.10	0.02	<.0001
Mean Minimum Winter Temperature	0.17	0.02	<.0001	0.17	0.02	<.0001
Mean Summer Temperature	2.97	1.25	0.018	3.02	1.25	0.016
Mean Summer Temperature Squared	-0.02	0.01	0.016	-0.02	0.01	0.014
First Year	0.31	0.96	0.744	0.34	0.97	0.722
Dispersion	0.61	0.17		0.62	0.17	
Zero Model	β Estimate	Standard Error	p-value	-16.92	2.71	<.0001
Intercept	-12.61	2.32	<.0001	-9.62	2.08	<.0001
RUCC 1 - 4	-1.43	0.27	<.0001	-1.40	0.28	<.0001
RUCC 5-9 – Reference	0.00	0.00	.	0.00	0.00	.
Percent of Population Over 50	0.25	0.04	<.0001	0.28	0.04	<.0001
First Year	-0.94	1.86	0.613	-0.83	1.82	0.648

APPENDIX D (CONTINUED)

TABLE XXVI. SENSITIVITY ANALYSIS COMPARING THE EFFECT OF PERCENT OF POPULATION OVER 70 VERSUS PERCENT OF POPULATION OVER 80

	Percent over 70			**Percent over 80**		
	β Estimate	Standard Error	p-value	β Estimate	Standard Error	p-value
Intercept	-112.03	46.32	0.016	-103.14	47.45	0.030
RUCC 1 - 4	-1.02	0.22	<.0001	-1.28	0.22	<.0001
RUCC 5-9 – Reference	0.00	0.00	.	0.00	0.00	.
Percent of Population Over 70 or 80	0.16	0.04	<.0001	0.24	0.09	0.007
Mean Minimum Winter Temperature	0.17	0.02	<.0001	0.17	0.03	<.0001
Mean Summer Temperature	2.96	1.25	0.018	2.73	1.28	0.034
Mean Summer Temperature Squared	-0.02	0.01	0.017	-0.02	0.01	0.032
First Year	0.34	0.96	0.720	0.41	1.07	0.702
Dispersion	0.61	0.17		0.83	0.24	
Zero Model	β Estimate	Standard Error	p-value	-16.92	2.71	<.0001
Intercept	-9.01	2.03	<.0001	-8.52	2.19	<.0001
RUCC 1 - 4	-1.28	0.29	<.0001	-1.87	0.34	<.0001
RUCC 5-9 – Reference	0.00	0.00	.	0.00	0.00	.
Percent of Population Over 50	0.48	0.07	<.0001	0.73	0.16	<.0001
First Year	-0.89	1.82	0.626	-0.78	1.91	0.682

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