Title

Environmental and Socioeconomic Factors Associated with West Nile Virus in the Northern San Joaquin Valley

Author

Hernandez, Eunis

Publication Date

2018-04-01

Eunis Hernandez

Master's Article

Advisor: Dr. Andrea Joyce, Committee: Dr. Nancy Burke and Dr. Ricardo Cisneros

9/28/2017

Environmental and Socioeconomic Factors Associated with

West Nile Virus in the Northern San Joaquin Valley

Introduction

Mosquito-borne illnesses are a threat to global health and are known to affect nearly 700 million people annually (Caraballo and King 2014). One of these diseases is West Nile virus (WNV), an arthropod-borne virus of the family Flaviviridae that is commonly spread by mosquitoes (Kleinschmidt-DeMasters and Beckham 2015). This virus emerged from Uganda in 1937 and since this initial discovery, has spread geographically to places including Europe, Australia, Asia, the Caribbean, and South America (Petersen and Roehrig 2001). In 1999, WNV reached the United States when its first reported case emerged in New York City with a group of patients diagnosed with encephalitis (Nash et al. 2001). By July 2003, WNV arrived in California and shortly afterwards its activity was detected across all 58 counties (Reisen et al. 2004).

West Nile virus is a member of the Japanese encephalitis virus (JEV) serogroup that is composed of viruses associated with human encephalitis such as Japanese encephalitis, St. Louis encephalitis, Kunjin virus, Usutu virus, and Murray Valley encephalitis (Murray, Walker, and Gould 2011). WNV is phylogenetically divided into 5 lineages, but only lineages 1 and 2 have been linked to outbreaks in humans (Petersen, Brault, and Nasci 2013); a majority of animal and human disease cases that have been reported are attributed specifically to lineage 1 including

New York City's 1999 outbreak (Petersen and Marfin 2002; Hoover and Barker 2016).

Mosquitoes belonging to the genus *Culex* are considered the primary carriers of WNV. In California, the mosquitoes which commonly carry WNV are *Culex pipiens, Culex quinquefasciatus,* and *Culex tarsalis* (Lindsey et al. 2010). The northern house mosquito, *Cx. pipiens*, is found primarily in the northern part of California, *Cx. quinquefasciatus* (the southern house mosquito) is found in the southern portion of the state, and *Cx. tarsalis* exists in many areas of the state (Petersen, Brault, and Nasci 2013). The Central Valley of California is an area that additionally has hybrids of *Cx. pipiens* and *Cx. quinquefasciatus* (Ciota, Chin, and Kramer 2013). The larvae of *Cx. pipiens* and *Cx. quinquefasciatus* develop in water sources rich in organic matter such as sewers, abandoned swimming pools, and dairy lagoons, and are commonly found in urban habitats, while *Cx. tarsalis* is more common in rural areas including artificial and natural wetlands. These mosquitoes are competent vectors of WNV as they are the most prevalent species found in urban areas, they create disease outbreaks during their period of greatest abundance (Fonseca 2004).

WNV Transmission Cycle

The majority of human WNV cases are as a result of mosquito bites (Shapshak et al. 2015). WNV is not spread through casual contact such as touching or breathing in the virus. The virus is maintained primarily through a cycle between birds and mosquitoes. In this transmission cycle, female mosquitoes are able to transmit WNV after they feed on infected birds; more than 300 species of birds are considered reservoir hosts, some of which include crows, magpies, jays, finches, blackbirds, warblers, sparrows, and raptors (Shapshak et al. 2015). In a WNV transmission cycle, the female mosquito will feed on an infected bird, the virus will then infiltrate the gut of the female mosquito, replicate, and journey back to the mosquito's salivary glands in a period of about one week (Shapshak et al. 2015). In the next feeding, the mosquito may inject their infected saliva into their blood meal (hosts such as birds, humans, horses or other mammals) where the host may or may not exhibit symptoms. A small proportion of human WNV cases have developed as a result of blood transfusions, organ transplants, and transmittance from mother to child during pregnancy, delivery, or through breastfeeding (Kramer, Li, and Shi 2007).

Symptoms

Throughout the world, WNV is considered the arthropod-borne disease responsible for the greatest number of neuroinvasive disease outbreaks that have ever been reported (Ciota and Kramer 2013). Neuroinvasive diseases include meningitis, encephalitis, or acute flaccid paralysis/poliomyelitis (Davis et al. 2006). People who are 50 years of age or older are at the greatest risk of developing a severe illness (Petersen and Marfin 2002). WNV encephalitis can range in the severity of its symptoms which may include mild disorientation, neck stiffness, severe tremors, Parkinsonism, coma, and death. In addition, people that have certain medical conditions such as hypertension, cancer, diabetes, and kidney disease are at higher risk for developing severe illnesses. Approximately 20% of people who are infected with WNV will display milder symptoms such as headaches, body aches, rashes, weakness, joint pains, vomiting, and diarrhea, while about 80% of people will be asymptomatic. The time of recovery from this virus will vary on the severity of symptoms. Those who display milder symptoms can expect that the virus's effects will last from several days to weeks or months. The duration of neurologic illnesses as a result of WNV may take months or years to recover from. In New York City, a study was conducted to describe the process of recovery from WNV encephalitis of residents that were infected during the 1999 WNV encephalitis outbreak (Klee et al. 2004). This cohort of New York City residents was monitored for 18 months where health status was assessed at 6, 12, and 18 months after illness onset (Klee et al. 2004). Only 37% of patients achieved a full recovery. In comparison, 45% of patients that were 65 or older experienced some recovery either physically, cognitively, or functionally. A positive predictor of recovery in this specific study was age. Young individuals that were less than 65 years old were more likely to reach a full recovery.

Treatment and Prevention

At this time, there is no vaccine available for humans against WNV. There are however, vaccines available for horses to minimize their risk of infection: a vaccine composed of formalin-inactivated whole virus with adjuvant, a DNA plasmid vaccine, a recombinant vaccine that consists of a canarypox virus vector, and also an attenuated WNV, live flavirvirus chimera vaccine (Seino et al. 2007). Recommended treatment for mild cases in humans has been taking over-the-counter pain relievers to reduce joint pains or fevers. With severe WNV cases, hospitalization may be necessary along with fluids that are given intravenously. People infected with WNV may experience difficulty breathing and will need respiratory support through a ventilator. Prevention treatment of secondary infections such as urinary tract infections or pneumonia may be required as well as nursing care. WNV is an important issue for the population of California, and the risk of developing severe illnesses may vary among these residents. For medical costs in the United States, each case of WNV neuroinvasive disease (WNND) was $46,530 and $302 for each case of West Nile fever (WNF) (Petersen, Brault, and Nasci 2013).

California Mosquito Surveillance and West Nile Virus

The state of California has a WNV disease surveillance program, which includes monitoring human cases, determining WNV infection in two mosquito species (*Cx. pipiens* and *Cx. tarsalis*), testing sentinel bird flocks for presence of WNV, and testing dead birds for the virus as well. The purpose of monitoring mosquitoes and birds for WNV is to detect the presence of WNV *before* it occurs in humans. This process is called 'active surveillance', and is conducted statewide in most counties in California by county Mosquito Abatement Districts. Since 1969, California has monitored abundance and activity of mosquitoes through their mosquito surveillance program (California Department of Health Services, Mosquito and Vector Control Association of California, University of California 2015). Mosquito abatement agencies are associated with the county Department of Environmental Health. California is divided into five regions for mosquito control. The Northern San Joaquin region consists of Merced, Stanislaus, and San Joaquin Counties.

Each Mosquito Abatement district is funded by local property taxes (Merced, $8/year/home), and the agency takes the responsibility of conducting surveillance for WNV, and conducting educational activities to educate the community about disease prevention. Merced's educational outreach program for WNV prevention consists of a team which visits every 3rd grade classroom each year to discuss mosquito biology, and prevention of WNV. Mosquito surveillance includes determining adult mosquito abundance which provides information on the vector population size. Four trap collection methods are used to detect *Culex* mosquito species in California; New Jersey light traps, carbon dioxide-baited traps, gravid female traps, and resting adult mosquito collections (California Department of Health Services, Mosquito and Vector

Control Association of California, University of California 2015; Goddard 2013). Adult mosquitoes are then identified and tested for WNV. When WNV is detected, a county mosquito abatement agency works to reduce mosquito populations in the area where WNV was detected. This reduces the probability of humans contracting WNV. Mosquito surveillance which detects WNV is considered a fundamental predictor for human WNV cases (Brownstein, Holford, and Fish 2004). Surveillance and trapping focuses on areas and environmental factors most likely to produce large numbers of *Culex* mosquitoes. We are interested in investigating, in our region of the Central Valley, the role of both environmental and socioeconomic factors and their association with where human WNV cases occur.

Environmental Factors

The importance of environmental factors and their influence of where human WNV cases occur has been investigated since WNV arrived to the United States (Gibbs et al. 2006). In southern California, a study included ten counties and examined factors associated with where WNV cases developed; the counties included were San Luis Obispo, Kern, San Bernardino, Santa Barbara, Ventura, Los Angeles, Orange, Riverside, San Diego, and Imperial County (Liu and Weng 2012). The purpose of this study was to provide a more precise surveillance method by incorporating additional environmental factors not typically included in mosquito surveillance. The study found that climatic factors that contributed significantly to the transmission of WNV were summer mean temperature, annual mean deviation from the mean temperature, and land surface temperature. Elevation and slope of different regions were also significantly related to the spread of WNV. Landscape patterns and vegetation water content also had a significant influence in the distribution of the virus.

High temperature has been consistently associated with WNV outbreaks (Hoover and Barker 2016). The rise in temperature can cause an increase in growth rates of vector populations, reduce the time between blood meals, increase transmission to birds, and reduce incubation time from infection to infectiousness in mosquitoes (Paz 2015). Seasonal variation that results in mild winters has been associated with increased WNV activity and if followed by hot, dry summers can favor the transmission of infections (Epstein 2001; Wimberly et al. 2014). In the United States from 2001 to 2005, a 5°C increase in mean maximum weekly temperature was associated with a 32-50% higher incidence of WNV infection (Soverow et al. 2009).

Mosquito species thrive in specific habitats. In Shelby County, Tennessee, *Cx. pipiens* and *Cx. quinquefasciatus* were positively associated with urban habitats (Savage et al. 2008). In contrast, the abundance of *Cx. pipiens* complex mosquitoes was lower in rural sites. A common principle in epidemiology states that above-average precipitation may lead to greater mosquito abundance and an increase for WNV outbreaks in humans (Landesman et al. 2007; Soverow et al. 2009). However, in urban areas a prolonged time of drought can result in increased abundance of mosquitoes that can intensify transmission of WNV (Hoover and Barker 2016). The reason for this is the result of urban storm water management systems are not being regularly flushed by rainwater during droughts. Thus, they are supplied with water from landscape irrigation systems that is mixed with organic materials. This quickly becomes a favorable habitat for mosquitoes, especially *Cx. pipiens*. In particular, *Cx. pipiens* (one of the principal WNV vectors) prefers water sources with high organic material such as dairy ponds, catch basins, and septic tanks (Meyer and Durso 1998). *Cx. tarsalis* instead favors open habitats such as swamps, marshes, rice fields, and open pastures (Horsfall 1955).

Socioeconomic Factors

Some studies have found that economic variables and anthropogenic characteristics of the environment best explained and predicted WNV prevalence. Orange County, an urban region in southern California, has been considered a WNV hotspot since the year 2004. A study which examined human WNV cases from 2004-2008 revealed that lower-income areas (per capita income) had higher prevalence levels of WNV in vectors (Harrigan et al. 2010). An important variable that provides an explanation for years of high WNV prevalence was the density of neglected swimming pools (Reisen et al. 2008). During the period that this study was performed, Orange County experienced a large increase in home foreclosures and neglected swimming pools. As a result, these findings suggested that abandoned pools stimulated WNV amplification and provided a link between WNV propagation and lower income, populated areas (Harrigan et al. 2010). In addition, Bakersfield, Kern County experienced a 300% increase in notices of delinquency that produced large numbers of neglected swimming pools; this was associated with a 276% increase in human WNV cases (Reisen et al. 2008). There are three possible explanations as to why a link may exist between low income areas and WNV propagation. The first explanation states that areas with dense populations predominately occur in flatland areas of low elevation. These areas are characterized with old infrastructure and outdated water runoff systems that can serve as breeding habitats for mosquitoes (Su et al. 2003).The second explanation is that lower-income communities are less likely to spend for upkeep of their property. Findings for this conclusion were drawn from the rise in number of neglected swimming pools. The last reason for the link between WNV and lower income areas would be education (Harrigan et al. 2010; Milligan, Moretti, and Oreopoulos 2004). People with higher education show more involvement with mosquito control. For example, they request pest control services such as spraying. This study also suggests prevalence of WNV is related to the bird diversity in an area. Avian species are considered the primary hosts, and if there is a loss of diversity, an increase of WNV infections is seen among humans the alternative hosts.

An aerial survey conducted in the Bakersfield area of Kern County (Reisen et al. 2008) similarly found a substantial amount of swimming pools that had been neglected. Neglected pools contain large amounts of organic matter that result in green-colored water. Neglected swimming pools were frequently found among new housing areas and not just restricted to just old neighborhoods.

Considered as a hotspot of WNV activity, Orange County from 2004-2012 had a record total of 240 human WNV infections and 9 deaths (Liao et al. 2013). WNV is now considered endemic in this area; its particularly important to identify which predictors are significant. The strongest predictor in the Orange County study were street gutters followed by housing unit density, neglected swimming pools, mean per capita income, number of mosquito breeding sites and ditches, and housing average age. Variables that were found to be statistically insignificant were catch basins and flood control waterways, population density, the proportion of people who were over the age of 65, and the location of WNV positive mosquitoes as well as the area where infected dead birds were found.

A study in Chicago and Detroit assessed which natural and socioeconomic factors greatly influenced the transmission of WNV (Ruiz et al. 2007). A strong association was seen between the risk of WNV transmission and the age of housing in both locations. A possible explanation for this relationship would again be the storm water sewer system. The combination of standing water and organic matter can affect breeding of *Culex* mosquitoes. Vegetation found in the inner suburb areas of Chicago and Detroit was found to be significant for WNV transmission. Distinct features of these areas consisted of small yards with cemeteries, shrubs, hardwood trees, and grassy alleys. Results of this study provided insight to factors that are strong predictors for urban areas. However, additional socioeconomic factors in this study would have been helpful to understand WNV amplification.

In Suffolk County, New York, socioeconomic conditions were the key predictors of human WNV infections (Rochlin et al. 2011). The effects of urbanization and increased WNV activity were strongly associated. Such effects included fragmented natural areas, increased road density, and urban areas where there were high numbers of people with a college education. This finding suggested that middle class suburban neighborhoods or "inner suburbs" had the greatest activity and risk for WNV. The inner suburbs appear to provide favorable conditions for WNV transmission in contrast to wealthier neighborhoods. In this study, high income neighborhoods have more vegetation, less habitat fragmentation, and higher biological diversity. This high biological diversity resulted in less activity of human WNV infections as there was higher availability of preferred avian hosts. Natural wetland areas that have fragmented areas provide favorable habitat to mosquito larvae in contrast to manmade wetlands.

West Nile Virus in the Northern San Joaquin Valley of California

The purpose of this study is to investigate the environmental and socioeconomic factors associated with human WNV cases in the Northern San Joaquin Valley, in San Joaquin, Stanislaus, and Merced counties. Environmental variables herein consisted of mosquitoes which tested positive for WNV and habitat, and socioeconomic variables included age, education, income, housing age, population density. In addition to investigating variables previously found to be associated with WNV, two new variables were considered which were not examined in previous studies; language spoken at home and ethnicity. These two variables were also considered to investigate whether non-English speakers or non-white ethnic groups may be more likely to contract WNV due to language barriers or difficulty in accessing media about WNV prevention. We considered whether the number of human WNV cases in each census track in the three counties was related to these nine variables, and attempted to determine which variables were most strongly related to where the human WNV cases occurred.

Methods

Study Area:

The areas of study consist of 3 counties in the Northern San Joaquin Valley of California, from north to south, the counties are San Joaquin, Stanislaus, and Merced County (Fig. 1). The Northern San Joaquin Region is one of 5 regions in the Mosquito and Vector Control Association of California. In this region, there are two mosquito species, *Cx. pipiens* and *Cx. tarsalis*, which are the primary vectors of WNV. Mosquito management is relatively similar throughout the region as it is largely rural, with several large cities in each county.

San Joaquin County is located approximately 91 miles east of San Francisco and just south of the Sacramento area. According to the 2010 United States Census, the population size of this county is 685,306 and has a total area of 1,426 square miles. Large cities that are located within this county include Manteca, Ripon, Stockton, Lodi, and Tracy. The county is a rural area and its ethnic composition is 51.0% White, 25.9% Hispanic, 14.4% Asian, 7.6% African American, and 1.1% American Indian. In San Joaquin County, 76.7% of people have a high school diploma or higher while 17.5% have a bachelor's degree or higher. The median household income from the 2010 census is $54,341.

Stanislaus County is located south of San Joaquin County and north of Merced County. The population size is 514,453, and the county has a total area of 1,514 square miles. Stanislaus County contains cities including Ceres, Modesto, Oakdale, and Turlock. The ethnic makeup of this county consists of 65.6% White, 25.3% Hispanic, 5.1% Asian, 2.9% African American, and 1.1% American Indian. In this county, 75.3% of the population has a high school diploma or higher while 16.3% have a bachelor's degree or higher. The county's median household income from the 2010 Census is $51,094.

Merced County is at the southern end of the three counties. As of the 2010 United States Census, the population size was 255,793. The total area of the county is approximately 1,978 square miles and includes the cities of Atwater, Livingston, Merced and Los Banos. The ethnic composition of Merced County is 58.0% White, 29.3% Hispanic, 7.4% Asian, 3.9% African American, and 1.4% American Indian. Approximately 67.0% of people living in Merced County have a high school diploma or higher while 12.5% have a bachelor's degree or higher. The median household income from the 2010 Census is $43,844.

Data Collection:

Data used in this study include environmental and socioeconomic variables, and the number of human WNV cases from 2011-2015 in each census track of San Joaquin, Stanislaus, and Merced Counties. The environmental variables include the number of mosquito's positive for WNV and the habitat for each census track. Socioeconomic variables were obtained from the 2010 United States Census Bureau Census. The socioeconomic variables are defined in Appendix 1.

Statistical Analysis

Descriptive Overview

The number of human WNV cases were found for each of the three counties (Table 1). Subsequently, the number of census tracks with and without human WNV cases were determined for each county (Table 1). For Stanislaus County, one census tract was dropped from analyses due to incomplete data. For each year from 2011-2015, the frequency of human WNV cases in each county were plotted for comparison (Figure 2). Finally, for each county a map was made to show census tracks with and without human WNV cases (Figs. 3-5).

Chi-square Analyses

For each county, chi-square independence tests were run to determine whether a significant association exists between human WNV cases and each variable of interest. For each variable (for example age), we used the 2010 Census to first determine the mean for the variable for each county. For example, for age, the mean percent of the population over age 65 was used to then categorize the census tracks in the county into those with a 'lower' or 'higher' than the average percent of the population over age 65. Subsequently, a chi-square independence test was used to determine if WNV human cases were associated with age (average percent of the population over age 65). A similar analysis was run for the other eight variables. Variables tested were age, education, income, housing age, ethnicity, language, population density, habitat, and WNV positive mosquitoes. STATA 14.2 was used to run these analyses.

An overall chi-square independence test for all data from the three counties was similarly conducted (Table 5). For most of the nine variables, the mean for the category was found by averaging the mean for that variable from the three counties; census tracks were then classified into ‘low’ or ‘high’ for that variable. All chi-square tests were run using STATA 14.2. The result of an analysis was considered significant if $p<0.05$.

Logistic Regressions

Logistic regressions were conducted to examine the relationship between the variables and the dependent dichotomous variable, which was the presence or absence of human WNV case in each census track. Analyses were conducted for each county, with an additional analysis for all three counties combined. A human case of WNV (yes or no) was selected as the dependent variable in each census track, and analyses were conducted using STATA 14.2.

Socioeconomic and environmental variables used in analyses included language spoken at home, median year structure built, ethnicity, age, population density, median household income, education, mosquitoes positive for WNV, and habitat. Most of the independent variables were continuous (numeric) except for two variables, habitat and WNV positive mosquitoes, which were categorical. Habitat was treated as a dummy variable, with rural habitat represented by a ‘1’ and non-rural by a 0; WNV mosquitoes were similarly coded with mosquitoes testing negative for WNV with a ‘0’ and those testing positive with a ‘1’. All data were entered into the model for each county, and then a model was run for the 3 counties combined. The output was used to examine the overall Chi-square test, and whether it was significant at $p<0.05$. The Z-score and its related p-value was determined for each variable in the model. Variables with Z-scores that had p-values <0.05 were considered significant. Finally, the odds ratios were determined for each variable in the model.

Results

Descriptive Overview

The number of human cases of West Nile Virus (WNV) for 2011-2015 totaled 169 cases for the three counties (Table 1). The largest number of cases was in Stanislaus County (114), followed by San Joaquin County (39), and the lowest number of cases occurred in Merced County (16) (Table 1).

The WNV cases were plotted for each county for 2011-2015 (Fig. 2). Stanislaus had the highest number of cases overall for all five years (Fig. 2), followed by San Joaquin County, and Merced County had the lowest number of cases. A map with census tracks in each county was used to highlight the census tracks for presence or absence of human WNV cases (Fig.3-5). In San Joaquin County, there were 27 tracks with 1 human WNV case, and 5 tracks with 2 human cases, and 0 tracks with 3 or more cases (Fig. 3). In Stanislaus County, there were 22 tracks with 1 case, 19 tracks with 2 cases, and 13 tracks with 3 or more cases (Fig. 4). In Merced County, there were 7 tracks with 1 case, 1 track with 2 cases, and 2 tracks with 3 or more cases (Fig. 5).

Chi-square Analyses

San Joaquin County

In San Joaquin County, 139 census tracts were used for chi-square tests, which included 39 human WNV cases (Table 1). A chi-square test of independence was performed for San Joaquin County to examine the relation between WNV human cases and the nine variables previously mentioned. A significant relationship was found between human WNV cases and the variables: rural habitat, X^2 (1, N=139) =4.993, $p < .05$, with more cases in tracks with lower population density; population density, X^2 (1, N=139) =4.100, $p < .05$ with higher density tracks having more human WNV cases (8 vs 25), and finally tracks with WNV positive mosquitoes, X^2 (1, N=139) =11.29, $p < .05$. The relation between language spoken at home and WNV cases was not significant, X^2 (1, N=139)=0.247, $p > .05$, nor was the relationship between median housing year and human WNV cases, X^2 (1, N=139) =0.530, $p > .05$, percent Caucasian, X^2 (1, N=139) =2.99e-04, $p > .05$, age, X^2 (1, N=139) =0.001, $p > .05$, median household income, X^2 (1, N=139) =0.052, $p > .05$, and education (high school graduates or higher), X^2 (1, N=139) =3.302, $p > .05$.

Stanislaus County

Chi-square independence tests were calculated to determine if a statistical association exists between human WNV cases and the nine variables of interest for Stanislaus County. In Stanislaus County, 93 census tracts were used for chi-square tests, and 114 human WNV cases. Rural habitat was positively associated with cases of WNV, X^2 (1, N=93) =32.09, $p < .05$. Census tracks with higher than the median household income were more likely to have WNV cases, X^2 (1, N=93) =5.646, $p < .05$, as were tracks with a higher number of high school graduates or higher, X^2 (1, N=93) =9.603, $p < .05$. Finally, tracks where WNV positive mosquitoes were found were also significantly associated with WNV cases, X^2 (1, N=93) =4.857, $p < .05$. In contrast, results found the relationship was not significant between WNV human cases and language, X^2 (1, N=93) =1.447, $p > .05$, median housing year, X^2 (1, N=93) =0.022, $p > .05$, age, X^2 (1, N=93) =2.688, $p > .05$, and population density, X^2 (1, N=93) =1.734, $p >.05$.

Merced County

In Merced County, 49 census tracts were included for analyses, and 16 human WNV cases. Chi-square independence tests were again used to examine whether a relationship exists between WNV human cases and the nine variables previously described. WNV human cases were significantly related to WNV positive mosquitoes, X^2 (1, N=49) =: 9.70e-05, $p < .05$, but all other variables were not significantly associated with WNV (Table 2). Language, X^2 (1, N=49) =0.278, $p > .05$, median housing year, X^2 (1, N=49) =0.291, $p > .05$, Caucasian vs. non-Caucasian, X^2 (1, N=49) =0.720, $p > .05$, rural habitat, X^2 (1, N=49) =0.141, $p > .05$, median household income, X^2 (1, N=49) =0.090, $p > .05$, high school graduates or higher, X^2 (1, N=49) =0.291, $p < .05$, ages 65 and over, X^2 (1, N=49) =0.047, $p >.05$, and population density, X^2 (1, N=49) =0.201, $p <.05$ were insignificant (Table 2).

All 3 Counties Combined Analysis

Data from San Joaquin, Stanislaus, and Merced County were pooled in order to conduct a combined chi-square test of independence for WNV cases and each of the nine variables for the three counties. Population density was significantly related to WNV cases, with higher density tracks having less cases of WNV, X^2 (1, N=281) =7.382, $p < .05$, and tracks where WNV positive mosquitoes were found were more likely to have WNV cases, X^2 (1, N=281) =25.70, $p < .05$. Results suggest that human cases of WNV were not associated with language spoken at home, X^2 (1, N=281) =2.260, $p > .05$, median housing year, X^2 (1, N=281) =0.793, $p > .05$, age 65 and over, X^2 (1, N=281) =0.929, $p > .05$, and median household income, X^2 (1, N=281) =2.402, $p > .05$.

Logistic Regression

In the logistic regression model for San Joaquin County, the only statistically significant factor was WNV positive mosquitoes [Odds Ratio (OR)= 3.836; 95% CI, 1.377-10.680; p= 0.010]. None of the nine variables in a logistic regression model for Stanislaus County were significant. In Merced County, WNV positive mosquitoes again were the only significant factor (OR=41.72; 95% CI, 2.586-673.0; p=0.009).

Discussion

In this study, we examined the influence of factors on a total of 169 WNV human cases reported by three counties: San Joaquin, Stanislaus, and Merced County. The chi-square analysis revealed that WNV incidence in humans is influenced by many factors. Key factors that contributed to human cases were identified for these three rural counties; education and West Nile positive mosquitoes were significant and consistent with findings of previous studies that were done in urban areas. The majority of studies examining environmental and socioeconomic factors have been conducted in urbanized areas (Ruiz et al. 2004; Gibbs et al. 2006; Harrigan et al. 2010). Rural areas have been neglected in terms of research related to WNV.

Socioeconomic conditions have been found to significantly relate to WNV activity. Investigators in Chicago and Detroit found that middle class neighborhoods were associated with high WNV activity and human infection (Ruiz et al. 2007). The risk of WNV human infection was also associated with low income areas in Orange County, CA, Harris County, TX, and Shelby County, TN (Harrigan et al. 2010; Rios et al. 2006; Savage et al. 2008). In Stanislaus County, high income was statistically significant in the chi-squared analyses while for the other counties and the overall analysis of three counties, income was not significant. Increasing population density was also significantly related to higher numbers of WNV cases for San Joaquin County and for the three counties combined.

When all factors were combined in the logistic regression of nine variables, two factors were highly significant for the three-county analysis. This study along with those previously mentioned suggest that socioeconomic factors including ethnicity, income, and education are closely intertwined with human risk of WNV. Wealthier neighborhoods may provide a favorable habitat for mosquitoes consisting of more open space than in lower income rural counties and a certain combination of landscape vegetation that is preferable for mosquito breeding. Overall, these results show that WNV human cases were significantly associated with a high percentage of people who are Caucasian, high school graduates or of higher education, and have high income.

It has been previously reported that with advancing age the incidence of neuroinvasive disease and death due to WNV increases. However, results in this study showed that census tracks with more people ages 65 or over, a population at high risk for severe infection, were not significantly associated with WNV cases for either San Joaquin, Stanislaus, Merced, and the three counties combined. Although the factor age (65 and over) was not significant with human cases, people of all ages have an equal susceptibility to infection and therefore surveillance should continue to target all age groups. Although other studies found housing age strongly associated with human cases of WNV, it was not strongly associated with WNV cases in the analysis of the three counties individually or combined. Housing age has been a significant factor in prior studies as a result of old storm water sewer systems that can serve as a breeding source for *Culex* mosquitoes (Ruiz et al. 2007; Liao et al. 2013). This is a variable that could be further investigated for our region. Overall, WNV positive mosquitoes in a census track was a key determinant of WNV human cases for all counties individually and combined.

This analysis of environmental and socioeconomic factors that influence human cases of WNV can contribute to mosquito abatement districts' efforts towards reducing mosquito populations and containing these risk factors associated with WNV. Education and communication are key elements in public health prevention programs. Both are of key importance during a disease outbreak in order to adequately inform citizens of the risk of WNV and how to protect themselves. It is critically important that this information is available to all populations that are representative of each county which include the Hispanic, Asian, and Indian communities. Public health prevention messages could be created in other languages to inform the public especially those at greater risk of the dangers of WNV. Mosquito abatement districts can expand their form of communication to the general public through pamphlets, newspaper ads, TV advertisements, billboards, and town hall meetings. Greater involvement with social media, a platform that mosquito abatement districts utilize, is needed to inform the greater public and could be used in multiple languages to reach all ethnic groups living in this region of the Central Valley.

Literature Cited

Brown, Heidi E., James E. Childs, Maria A. Diuk-Wasser, and Durland Fish. 2008. "Ecological Factors Associated with West Nile Virus Transmission, Northeastern United States." *Emerging Infectious Diseases* 14 (10): 1539–45. doi:10.3201/eid1410.071396.

Brownstein, John S., Theodore R. Holford, and Durland Fish. 2004. "Enhancing West Nile Virus Surveillance, United States." *Emerging Infectious Diseases* 10 (6): 1129–33. doi:10.3201/eid1006.030457.

California Department of Health Services, Mosquito and Vector Control Association of California, University of California. 2015. "California State Mosquito-Borne Virus Surveillance and Response Plan." https://www.cdph.ca.gov/programs/vbds/Documents/2015CAResponsePlan.pdf.

Caraballo, Hector, and Kevin King. 2014. "Emergency Department Management of Mosquito-Borne Illness: Malaria, Dengue, and West Nile Virus." *Emergency Medicine Practice* 16 (5): 1–23; quiz 23–24.

Ciota, Alexander, and Laura Kramer. 2013. "Vector-Virus Interactions and Transmission Dynamics of West Nile Virus." *Viruses* 5 (12): 3021–47. doi:10.3390/v5123021.

Ciota, Alexander T, Pamela A Chin, and Laura D Kramer. 2013. "The Effect of Hybridization of Culex Pipiens Complex Mosquitoes on Transmission of West Nile Virus." *Parasites & Vectors* 6 (1): 305. doi:10.1186/1756-3305-6-305.

Davis, Larry E., Roberta DeBiasi, Diane E. Goade, Kathleen Y. Haaland, Jennifer A. Harrington, JoAnn B. Harnar, Steven A. Pergam, Molly K. King, B. K. DeMasters, and Kenneth L.

Tyler. 2006. “West Nile Virus Neuroinvasive Disease.” *Annals of Neurology* 60 (3): 286–300. doi:10.1002/ana.20959.

Epstein, P. R. 2001. “West Nile Virus and the Climate.” *Journal of Urban Health: Bulletin of the New York Academy of Medicine* 78 (2): 367–71. doi:10.1093/jurban/78.2.367.

Fonseca, D. M. 2004. “Emerging Vectors in the Culex Pipiens Complex.” *Science* 303 (5663): 1535–38. doi:10.1126/science.1094247.

Gibbs, Samantha E. J., Michael C. Wimberly, Marguerite Madden, Janna Masour, Michael J. Yabsley, and David E. Stallknecht. 2006. “Factors Affecting the Geographic Distribution of West Nile Virus in Georgia, USA: 2002-2004.” *Vector Borne and Zoonotic Diseases (Larchmont, N.Y.)* 6 (1): 73–82. doi:10.1089/vbz.2006.6.73.

Goddard, Jerome. 2013. *Public Health Entomology*. Boca Raton: CRC Press.

Harrigan, Ryan J., Henri A. Thomassen, Wolfgang Buermann, Robert F. Cummings, Matthew E. Kahn, and Thomas B. Smith. 2010. “Economic Conditions Predict Prevalence of West Nile Virus.” Edited by Wayne M. Getz. *PLoS ONE* 5 (11): e15437. doi:10.1371/journal.pone.0015437.

Hoover, Kara C., and Christopher M. Barker. 2016. “West Nile Virus, Climate Change, and Circumpolar Vulnerability: West Nile Virus, Climate Change, and Circumpolar Vulnerability.” *Wiley Interdisciplinary Reviews: Climate Change* 7 (2): 283–300. doi:10.1002/wcc.382.

Horsfall, W.R. 1955. “Mosquitoes Their Bionomics and Relation to Disease.” New York: The Ronald Press Company.

Klee, Anne Labowitz, Beth Maldin, Barbara Edwin, Iqbal Poshni, Farzad Mostashari, Annie Fine, Marcelle Layton, and Denis Nash. 2004. “Long-Term Prognosis for Clinical West

Nile Virus Infection.” *Emerging Infectious Diseases* 10 (8): 1405–11. doi:10.3201/eid1008.030879.

Kleinschmidt-DeMasters, Bette K., and J. David Beckham. 2015. “West Nile Virus Encephalitis 16 Years Later: West Nile Virus Encephalitis 16 Years Later.” *Brain Pathology* 25 (5): 625–33. doi:10.1111/bpa.12280.

Kramer, Laura D, Jun Li, and Pei-Yong Shi. 2007. “West Nile Virus.” *The Lancet Neurology* 6 (2): 171–81. doi:10.1016/S1474-4422(07)70030-3.

Landesman, William J., Brian F. Allan, R. Brian Langerhans, Tiffany M. Knight, and Jonathan M. Chase. 2007. “Inter-Annual Associations Between Precipitation and Human Incidence of West Nile Virus in the United States.” *Vector-Borne and Zoonotic Diseases* 7 (3): 337–43. doi:10.1089/vbz.2006.0590.

Liao, Zhen, Kiet Nguyen, John Newton, and Robert Cummings. 2013. “Developing a Predictive Risk Map for West Nile Virus Activity Based on Mosquito Breeding Sources and Socioeconomic Factors in Orange County, California.” Mosquito and Vector Control Association of California.

Lindsey, Nicole P., J. Erin Staples, Jennifer A. Lehman, Marc Fischer, and Centers for Disease Control and Prevention (CDC). 2010. “Surveillance for Human West Nile Virus Disease - United States, 1999-2008.” *Morbidity and Mortality Weekly Report. Surveillance Summaries (Washington, D.C.: 2002)* 59 (2): 1–17.

Liu, Hua, and Qihao Weng. 2012. “Environmental Factors and Risk Areas of West Nile Virus in Southern California, 2007–2009.” *Environmental Modeling & Assessment* 17 (4): 441–52. doi:10.1007/s10666-011-9304-0.

Meyer, R.P., and S.L. Durso. 1998. “Identification of the Mosquitoes of California. Mosquito and Vector Control Association of California, Sacramento.”

Milligan, Kevin, Enrico Moretti, and Philip Oreopoulos. 2004. “Does Education Improve Citizenship? Evidence from the United States and the United Kingdom.” *Journal of Public Economics* 88 (9-10): 1667–95. doi:10.1016/j.jpubeco.2003.10.005.

Murray, K. O., C. Walker, and E. Gould. 2011. “The Virology, Epidemiology, and Clinical Impact of West Nile Virus: A Decade of Advancements in Research since Its Introduction into the Western Hemisphere.” *Epidemiology and Infection* 139 (06): 807–17. doi:10.1017/S0950268811000185.

Nash, Denis, Farzad Mostashari, Annie Fine, James Miller, Daniel O’Leary, Kristy Murray, Ada Huang, et al. 2001. “The Outbreak of West Nile Virus Infection in the New York City Area in 1999.” *New England Journal of Medicine* 344 (24): 1807–14. doi:10.1056/NEJM200106143442401.

Paz, S. 2015. “Climate Change Impacts on West Nile Virus Transmission in a Global Context.” *Philosophical Transactions of the Royal Society B: Biological Sciences* 370 (1665): 20130561–20130561. doi:10.1098/rstb.2013.0561.

Petersen, L. R., and J. T. Roehrig. 2001. “West Nile Virus: A Reemerging Global Pathogen.” *Emerging Infectious Diseases* 7 (4): 611–14. doi:10.3201/eid0704.010401.

Petersen, Lyle R., Aaron C. Brault, and Roger S. Nasci. 2013. “West Nile Virus: Review of the Literature.” *JAMA* 310 (3): 308. doi:10.1001/jama.2013.8042.

Petersen, Lyle R., and Anthony A. Marfin. 2002. “West Nile Virus: A Primer for the Clinician.” *Annals of Internal Medicine* 137 (3): 173–79.

Reisen, William K., Richard M. Takahashi, Brian D. Carroll, and Rob Quiring. 2008. “Delinquent Mortgages, Neglected Swimming Pools, and West Nile Virus, California.” *Emerging Infectious Diseases* 14 (11): 1747–49. doi:10.3201/eid1411.080719.

Reisen, William, Hugh Lothrop, Robert Chiles, Minoo Madon, Cynthia Cossen, Leslie Woods, Stan Husted, Vicki Kramer, and John Edman. 2004. “West Nile Virus in California.” *Emerging Infectious Diseases* 10 (8): 1369–78. doi:10.3201/eid1008.040077.

Rios, Janelle, Carl S. Hacker, Christina A. Hailey, and Ray E. Parsons. 2006. “Demographic and Spatial Analysis of West Nile Virus and St. Louis Encephalitis in Houston, Texas.” *Journal of the American Mosquito Control Association* 22 (2): 254–63. doi:10.2987/8756-971X(2006)22[254:DASAOW]2.0.CO;2.

Rochlin, Ilia, David Turbow, Frank Gomez, Dominick V. Ninivaggi, and Scott R. Campbell. 2011. “Predictive Mapping of Human Risk for West Nile Virus (WNV) Based on Environmental and Socioeconomic Factors.” Edited by Wayne M. Getz. *PLoS ONE* 6 (8): e23280. doi:10.1371/journal.pone.0023280.

Ruiz, Marilyn O., Carmen Tedesco, Thomas J. McTighe, Connie Austin, and Uriel Kitron. 2004. “Environmental and Social Determinants of Human Risk during a West Nile Virus Outbreak in the Greater Chicago Area, 2002.” *International Journal of Health Geographics* 3 (1): 8. doi:10.1186/1476-072X-3-8.

Ruiz, Marilyn O, Edward D Walker, Erik S Foster, Linn D Haramis, and Uriel D Kitron. 2007. “Association of West Nile Virus Illness and Urban Landscapes in Chicago and Detroit.” *International Journal of Health Geographics* 6 (1): 10. doi:10.1186/1476-072X-6-10.

Savage, Harry M., Michael Anderson, Emily Gordon, Larry McMillen, Leah Colton, Mark Delorey, Genevieve Sutherland, et al. 2008. “Host-Seeking Heights, Host-Seeking

Activity Patterns, and West Nile Virus Infection Rates for Members of the Culex Pipiens Complex at Different Habitat Types within the Hybrid Zone, Shelby County, TN, 2002 (Diptera: Culicidae).” *Journal of Medical Entomology* 45 (2): 276–88.

Seino, K. K., M. T. Long, E. P. J. Gibbs, R. A. Bowen, S. E. Beachboard, P. P. Humphrey, M. A. Dixon, and M. A. Bourgeois. 2007. “Comparative Efficacies of Three Commercially Available Vaccines against West Nile Virus (WNV) in a Short-Duration Challenge Trial Involving an Equine WNV Encephalitis Model.” *Clinical and Vaccine Immunology* 14 (11): 1465–71. doi:10.1128/CVI.00249-07.

Shapshak, Paul, John T. Sinnott, Charurut Somboonwit, and Jens H. Kuhn, eds. 2015. *Global Virology I - Identifying and Investigating Viral Diseases*. New York, NY: Springer New York. http://link.springer.com/10.1007/978-1-4939-2410-3.

Soverow, Jonathan E., Gregory A. Wellenius, David N. Fisman, and Murray A. Mittleman. 2009. “Infectious Disease in a Warming World: How Weather Influenced West Nile Virus in the United States (2001–2005).” *Environmental Health Perspectives* 117 (7): 1049–52. doi:10.1289/ehp.0800487.

Su, Tianyun, James P. Webb, Richard P. Meyer, and Mir S. Mulla. 2003. “Spatial and Temporal Distribution of Mosquitoes in Underground Storm Drain Systems in Orange County, California.” *Journal of Vector Ecology: Journal of the Society for Vector Ecology* 28 (1): 79–89.

Wimberly, M. C., A. Lamsal, P. Giacomo, and T.-W. Chuang. 2014. “Regional Variation of Climatic Influences on West Nile Virus Outbreaks in the United States.” *American Journal of Tropical Medicine and Hygiene* 91 (4): 677–84. doi:10.4269/ajtmh.14-0239.

Figure 1. Map of California counties. Three counties in the Northern San Joaquin valley were included in the study. From the north to south, the three counties are San Joaquin, Stanislaus, and Merced (shown in yellow).

County	Total Human Cases 2011-2015	Number of Census Tracts in County	Census Tracts with WNV	Census Tracts without WNV
Merced	16	49	10	39
Stanislaus	114	94	54	40
San Joaquin	39	139	33	106
Total	169	282		

Table 1. The number of human West Nile virus cases in Merced, Stanislaus, and San Joaquin counties

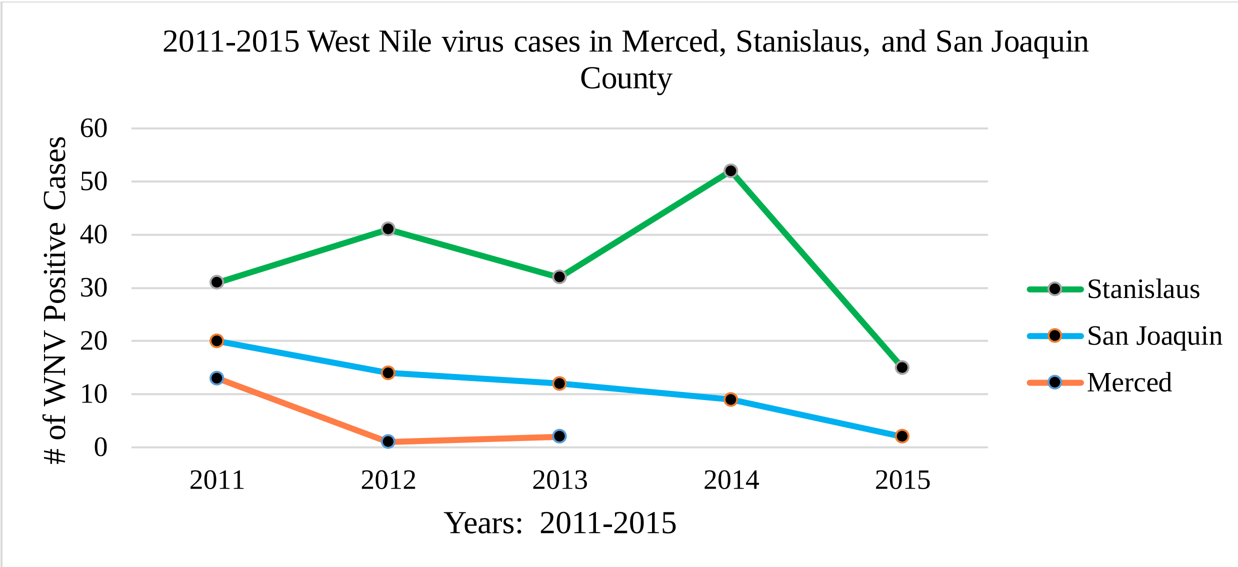

Figure 2. Human West Nile virus cases for the three counties from 2011-2015.

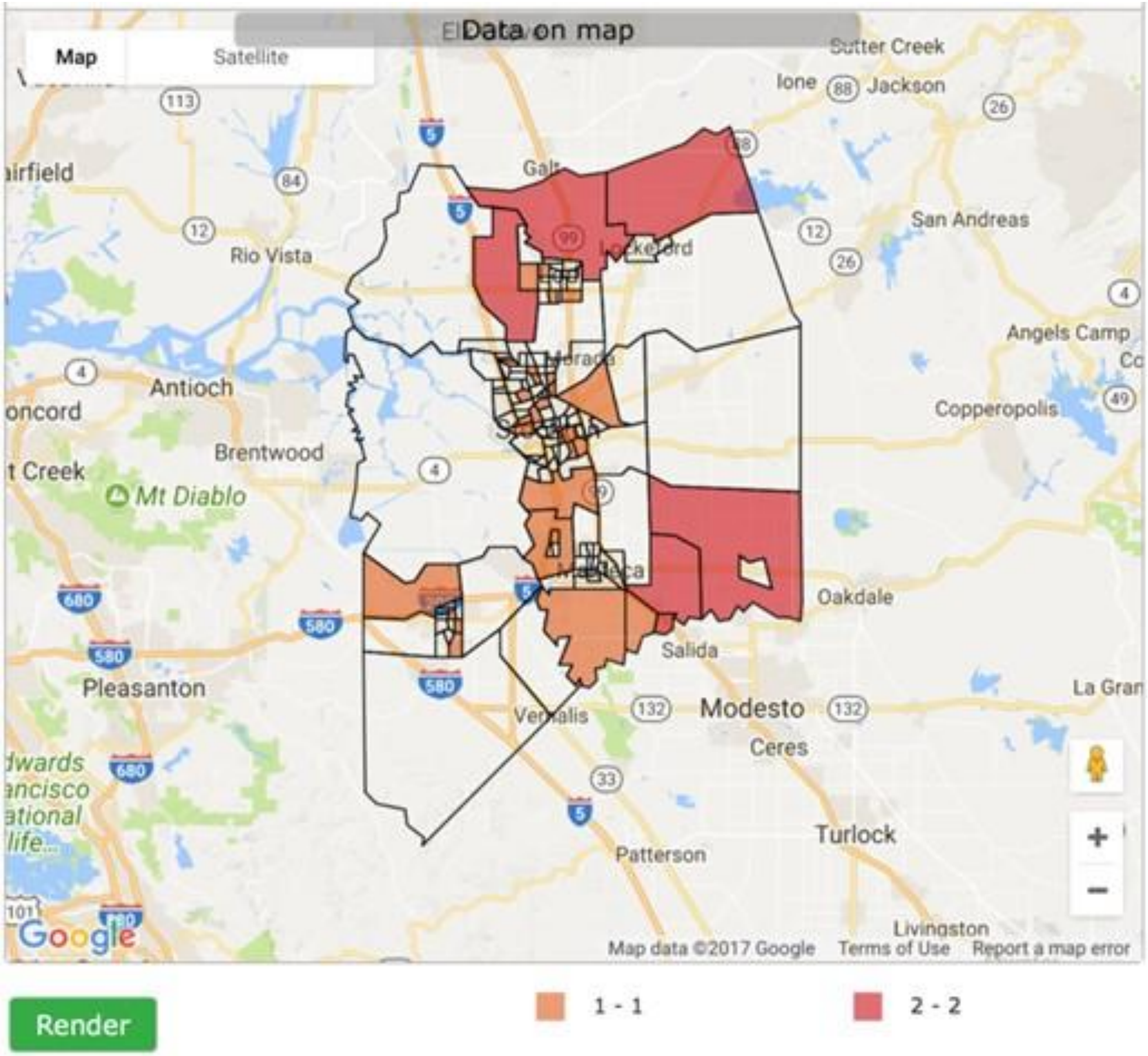

Figure 3. Map of San Joaquin County, showing census tracks and number of West Nile virus cases. Tracks shaded in orange have one human WNV case, while those in red have 2 cases.

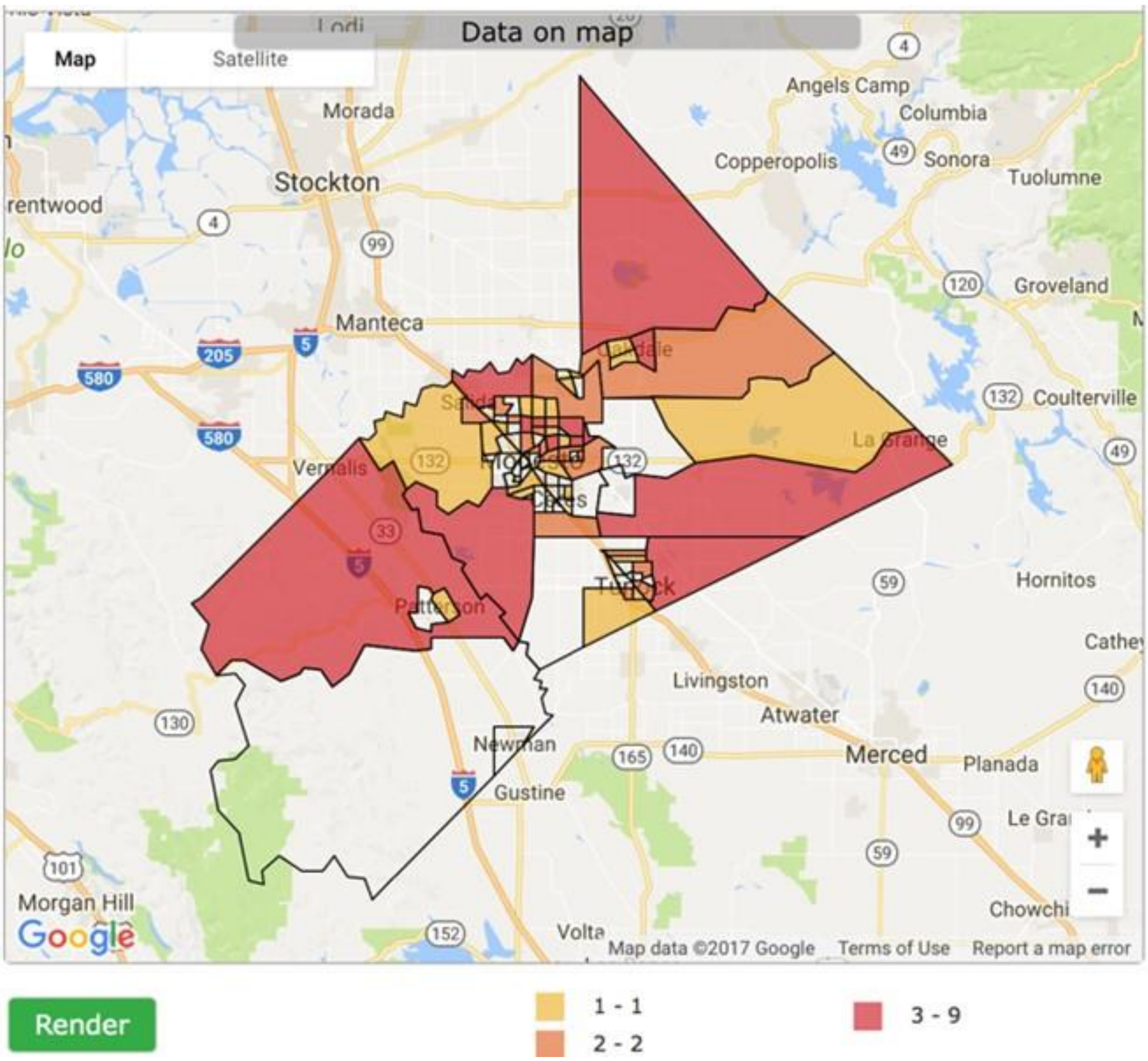

Figure 4. Map of Stanislaus County, showing census tracks and number of West Nile virus cases. Tracks in yellow have one human WNV case, while those in orange have 2 cases, and tracks in red have 3-9 cases.

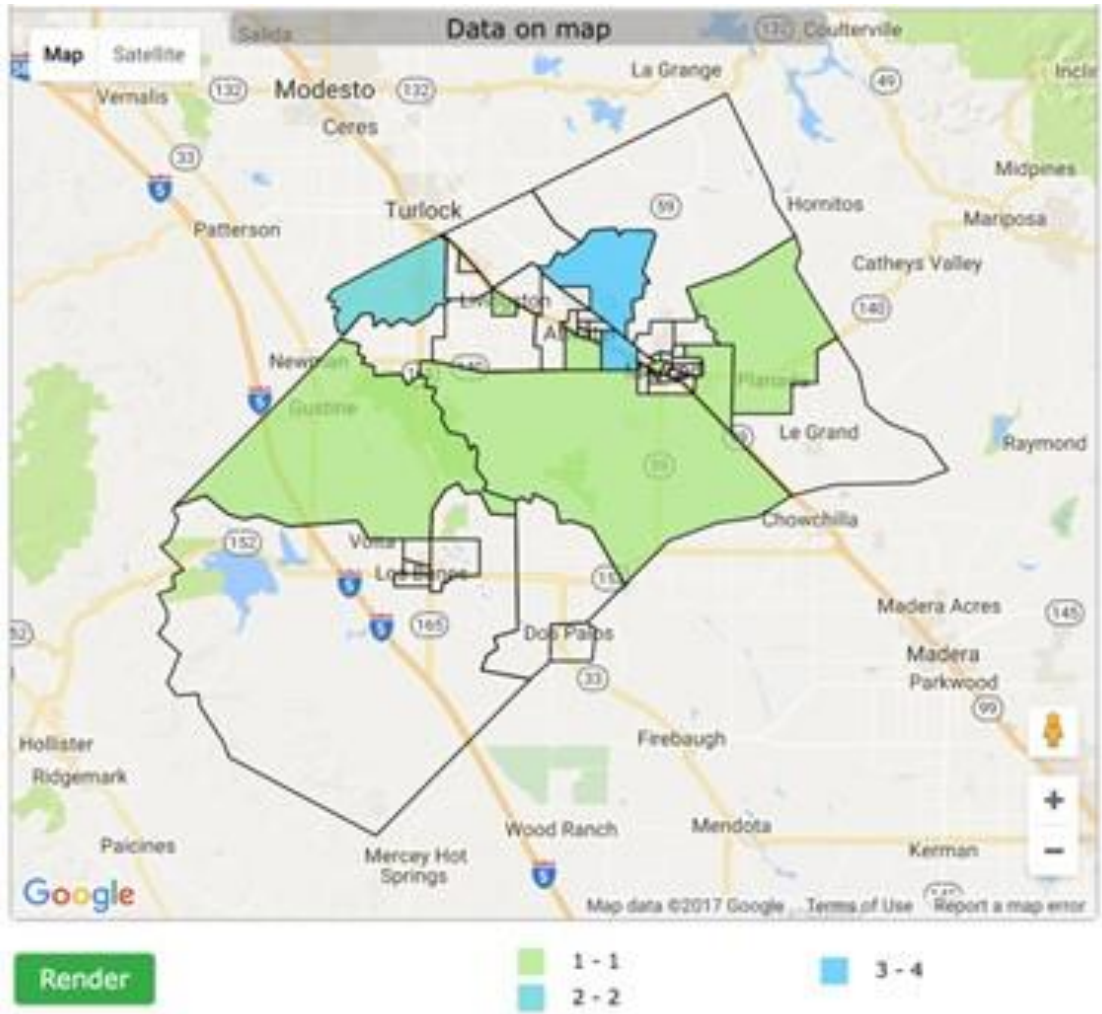

Figure 5. Map of Merced County, showing census tracks and number of West Nile virus cases. Census tracks shaded in green have 1 case, tracks filled with an aqua color have 2 cases, and census tracks with light blue had 3-4 reported human cases of WNV.

County	Variable	Mean	Low	High	Chi-square value/p-value
Merced	Language: English vs. non-English	English: 48.5%	< 48.5% 8 cases	≥ 48.5% 2 cases	X^2: 0.278 *P*>0.05
	Median Housing Year	1980	< 1980 4 cases	≥ 1980 6 cases	X^2: 0.291 *P*>0.05
	Ethnicity: Caucasian or non-Caucasian	Caucasian: 58.0%	< 58% 3 cases	≥ 58% 7 cases	X^2: 0.720 *P*>0.05
	Habitat: Rural or non-Rural	Rural: At least 1,000 people per square mile	Not rural 4 cases	Rural 6 cases	X^2: 0.141 *P*>0.05
	Age: 65 and over	65 and over: 9.4%	< 9.4% 5 cases	≥ 9.4% 5 cases	X^2: 0.047 *P*>0.05
	Median Household Income	$43,844	< $43,844 3 cases	≥ $43,844 7 cases	X^2: 0.090 *P*>0.05
	Education: High School Grads or Higher	67%	< 67% 4 cases	≥ 67% 6 cases	X^2: 0.291 *P*<0.05
	Population Density (per sq. mile of land area)	132.2	< 132.2 4 cases	≥ 132.2 6 cases	X^2: 0.201 *P*<0.05
	Positive WNV Mosquito in track or not	Positive: Previous history of mosquitoes carrying WNV in tract	No positive 39 tracks	Positive 10 tracks	X^2: 9.70e-05 *P*<0.05

Table 4. Chi-square variables for Merced County. The mean value for each variable is shown, and the number of census tracks which were classified as ‘low’ or ‘high’ for each variable. Results from the chi-square analysis and p-value are indicated as well.

Variables		
Variable	Label in Stata	Definition
Median Household Income	hhincome chh_income	Median income divides the income distribution into two equal groups, one having incomes above the median, and other having incomes below the median. The Census Bureau collected this data from surveys such as the American Community Survey (ACS).
Language	cnoneng_lang ceng_lang cspani_lang cindoe_lang casian_lang cother_lang	It is the language currently used by respondents at home. The percent of language spoken at home was retrieved from the U.S. Census Bureau. Non-English language was calculated by subtracting 100 from the "English language spoken at home" category.
Population Density	pop_density ctotal_pop	Population Density is measured per square mile. This information was retrieved from the OSHPD Healthcare Atlas. The total population number was retrieved from the U.S. Census Bureau.
Habitat	habitat rural_dummy sub_dummy urb_dummy	Using coordinates of census tracts and google earth, the tract's habitat was categorized as either rural, suburban, or urban.
Land area	cland_area	Land area is measured per square mile. This information was retrieved from the OSHPD Healthcare Atlas.
Housing Year	chousing_yr	Year structure built refers to when the building was first

		constructed. This information was retrieved from the U.S. Census Bureau.
Age	cage_under18 cage18_64 cage_65over	Age categories: under 18, 18-64, and 65 and over were retrieved from the U.S. Census Bureau.

Appendix 1. Variables used in Stata Analyses.