WEST NILE VIRUS MONITORING IN NORTH CENTRAL TEXAS AND A PROPOSED SURVEILLANCE MODEL

by

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ABSTRACT

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In the past 4 years Tarrant County, Texas has monitored its mosquito population for the presence of West Nile virus. In cooperation with the cities that reside within its borders, the Tarrant County Public Health Department trapped and tested mosquitoes of the species *Culex quinquefasciatus*. The North Texas Regional Laboratory tested these mosquito pools with the TaqMan RT-PCR Protocol. The participants selected trap locations based on convenience, past experience and public interest and it should be beneficial to use geomorphological characteristics to dictate future trapping locations. A surveillance model is proposed here using existing land maps of Tarrant County, past trap locations and their test results. The model makes statistical comparisons among the trap sites, their results and the characteristics of the site. These include features such as

elevation, floodplain data, urbanization and population density. The statistical model then predicts grid locations throughout the county that have many of the same characteristics as the positive sample sites and would be more likely to test positive for West Nile virus.

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CHAPTER 1

INTRODUCTION

1.1 West Nile virus and Tarrant County

West Nile virus is a mosquito borne disease that has plagued North America since 1999 and the World for over 30 years (Allen, 2003; Meek, 2002). Mosquito surveillance is one of the primary and most effective tools used by city and local governments to ascertain the location of the virus in their area. Surveillance is commonly used as part of a larger Integrated Pest Management plan that government and pest control agencies institute to monitor and manage the spread of vector borne diseases such as West Nile virus. West Nile virus appears to have spread rapidly and is now endemic to the Texas region (Lillibridge, 2004; Nasci, 2001).

In the North Central Texas region, some cities that conduct mosquito WNv surveillance choose their trap locations haphazardly. A mixture of public interest, observed mosquito prevalence, random selection and ease of placement determine the locations that cities place traps. Useful information may not have been available to the city governments to help guide their location choices. While a good surveillance plan should not exclude low risk locations entirely, it should take into account factors that increase the likelihood of finding the virus to create an accurate representation of the presence of the virus in the area (Allen, 2003).

West Nile virus is most likely transmitted from the avian reservoir to humans via the mosquito vector *Culex quinquefasciatus* in Tarrant County. *Culex quinquefasciatus* may not be the most competent vector in the lab. In nature, however, it serves as the most common and most efficient vector of the virus in North Central Texas, most likely because its primary blood meal source is avian (Turrell, 2001).

Culex quinquefasciatus species are generally foul water breeders. They breed in catch basins, storm water outfalls and residential containers of all kinds. Past investigations have found *Culex quinquefasciatus* breeding the following locations: Septic tanks, sewage treatment plants, buckets, boats, ornamental ponds, tires, rubbish bins, dumpsters, plastic containers, vases flower pots, fountains, swimming pools, hot tubs and wheel barrows (Hribar, 2007). High organic loads and low rainfall periods produce ideal conditions for this mosquito to thrive (Subra, 1981). It is this aspect of its breeding habits that may indicate which data serve to best predict the presence of the virus (Subra, 1981).

Analysis of the geomorphological characteristics of Tarrant County coupled with WNv sampling history may be able to provide guidance as to future choices of trap locations. Past WNv positive mosquito pools may have some statistically significant geomorphological features at the trap site that increase the likelihood of detecting the virus in those mosquito populations. Past research in Malaria control and prediction indicates that use of remote sensing, high spatial resolution mapping and identification of areas that contain breeding conditions are vital in predicting other areas that contain the virus (De Castro, 2004). This thesis will attempt to ascertain if these factors can be deduced statistically from geomorphological data in maps of the North Central Texas Region.

Some research already suggests that *Culex quinquefasciatus* are negatively correlated with high surface wetness and that there are seasonal limitations to predicting its success (Shaman, 2002). For these reasons Land Use, Urbanized Area and Vegetative Cover were selected as alternate variables affecting WNv detection (O'Ruiz, 2002). Also, past studies have indicated a close association with *Culex quinquefasciatus* success and human urbanization (Subra, 1981). Population density should be calculated to test this association and will be used to calculate risk in this model. The model used here is an attempt to adapt other similar surveillance models to the North Central Texas Region (O Ruiz, 2002).

Assuming some or all of the factors affect the prevalence of the virus, a predictive aspect of the model will also be created. With the WNv positive location data and the total sample set of locations, a novel sampling grid will be generated that indicates locations that may have a higher risk for mosquitoes containing WNv. The local governments can use these location results and maps to evaluate their monitoring and control procedures. Using the information presented in the paper these agencies can refocus their efforts in these high risk areas. It will also be helpful to local agencies to have non-political information about their city upon which to base control decisions such as pesticide spraying, larvicide application and public education.

1.2 Past Mapping and Statistical Models

It has become clear to the participants of the WNv Monitoring Program that some form of site selection and priority criterion would be helpful when the participants place traps in their cities. This could best be achieved by creating a model or analyzing past testing results to predict areas that would have a higher risk for detecting the virus in the future. I used several previous computer and statistical models regarding WNv to generate this model.

First, many past studies have stressed the need to use mapping and geological data to predict future cases of human, bird and mosquito WNv outbreaks (Allen, 2003; Tachiiri, 2006). In Mississippi, Cooke (2006) created a model that served as a good example of the type of data to seek and how to structure the analysis. He stresses the need to use GIS, and includes variables such as slope, road density and stream density. He suggests that bird data alone may be too general and that landscape data adds to the strength of the model (Cooke, 2006).

Another study considered when creating the model is de Castro's (2004) surveillance model in Tanzania. She analyzed topographic maps, urban maps, drainage schematics and aerial photography. She generated precise maps that indicated high risk areas for mosquito breeding and incidence of Malaria (de Castro, 2004).

Yiannakoulias (2007) indicated that to predict future outbreaks of vector borne diseases like West Nile virus, geographic information is paramount. According to him, past WNv research generally ignores the geomorphology as a predictor to future outbreaks. The use of prior surveillance and its results, coupled with spatial statistics and GIS analysis would be the strongest method for making predictions (Yiannakoulias, 2007).

Finally, a fourth study was done by O Ruiz (2002). This research was a study of geographic and social determinants of West Nile virus Infections. The study used many factors to predict areas of high WNv risk such as vegetation, age, income, race, age of housing, mosquito abatement and geological factors (O Ruiz, 2002).

CHAPTER 2

MATERIALS AND METHODS

2.1 Trapping and Testing Procedures

In order to conduct mosquito surveillance for West Nile virus in Tarrant County, a cooperative program was created by Gene Rattan R.S., Vector Control Specialist for Tarrant County Public Health. The County cooperated with more than 30 cities within Tarrant County. These cities agreed to perform mosquito trapping in and around their cities. The vector of choice for this surveillance program was identified as the Southern House Mosquito, or *Culex quinquefasciatus.* This species of mosquito is more successful than other species at transmitting WNv from the bird reservoir to a human host (Turrell, 2001). 92% of mosquitoes carrying West Nile virus were found to be *Culex pipiens* in Connecticut by Anderson (2006), a close regional relative to the *Culex quinquefasciatus.* Also, this species was found to be the primary vector of WNv in the Midsouth U.S.A. region (Cupp, 2007). A risk based analysis performed by Kilpatrick (2005) rated the risk of transmitting WNv of 10 common mosquito species. The risk was based on abundance of the mosquito, mammalian blood meal fraction, WNv infection prevalence and vector competence. *Culex pipiens* and *Culex restuans* were a much higher risk of being a vector (80%) than the next closest species *Aedes vexans* (4.5%). After deciding which species would be most likely to carry the virus, the best trapping method was found.

Culex quinquefasciatus has been shown in most research to be a foul water breeder. The type of trap selected closely approximated its ideal breeding habitat. The mosquito's ability to detect foul water from a distance and the characteristics of its oviposition behavior were considered when choosing the trap. *Culex quinquefasciatus* can use olfactory cues to seek out oviposition sites. They also will determine the suitability of the water chemistry by actual contact with the water (Bentley, 1989).

The John W. Hock "Gravid Trap" made the most sense, given this species' propensity to land in stagnant water. Gravid traps select for mosquitoes that are pregnant or "gravid" and are ready to oviposit in a foul body of water. This trap has proven in past studies to be successful and specific for *Culex quinquefasciatus* (Kline, 2006). The trap consists of a small bucket or pan filled with water that has been infused with hay, manure, grass clippings or fish oil. Some combination of the above ingredients is often used and the concoction is allowed to become pungent. This selection of bait has been found to work quite well and most closely resemble the source water of choice for *Culex quinquefasciatus* (Rey, 2006). High organic content with detritus animal and plant matter are highly attractive to the *Culex quinquefasciatus* (Kesavaraju, 2007). Situated above the pan is a small electric fan and net that gently draws any mosquito into the net and traps them there. These traps are cheap, easy to use and generally very successful (Kline, 2006; Rey, 2006).

70 or more gravid traps were distributed to the participating cities within Tarrant County. They were trained on the setup, use and common problems with the traps. Small- and medium-sized cities were set up on a trapping schedule of about 2 trapping

events every 2 weeks. The larger cities were provided more traps and trapped much more frequently with about 5 or more trapping events per week occurring. Once the cities submitted their mosquito samples to Tarrant County, they were tested for the presence of the virus.

The North Texas Regional Laboratory (NTRL) at the Tarrant County Public Health Center was able to institute a testing procedure for these mosquito samples or “pools.” The NTRL conducted a “Real Time” or Reverse Transcriptase Polymerase Chain Reaction Test to determine the presence of the virus in the samples. The most sensitive, rapid and reliable testing procedure for this type of test was the TaqMan RTPCR Assay procedure (Lanciotti, 2000). This procedure has been utilized successfully by Tarrant County Public Health for the entire length of its WNv testing. It is rapid as it only takes a few days from the receipt of the sample to generate the results and confirmation of the test. It is reliable as the amplification method is not prone to false positives. The test is versatile as many different types of samples can be accepted, such as mosquitoes themselves, human or avian blood, and oral/cloacal swabs (van den Hurk, 2007). Finally, the test is very sensitive. The amplification method does not need large viral loads to be detected. The protocol’s speed and the reliability of the output are superior to most other RTPCR testing methods for WNv (Lanciotti, 2000; Anderson, 2004).

Upon receipt of the trap contents from the participating cities, the county froze the mosquitoes to prepare them for sorting. Once the mosquitoes were dead or were too cold to move, the county would sort them by species and sex. The county personnel randomly selected female *Culex quinquefasciatus* from the submitted trap contents and inserted the bodies into a small vial for lab processing. All of the female *Culex quinquefasciatus* were sorted into the vial until a maximum of 50 was reached. All of the other mosquitoes above the first 50 were destroyed.

The samples were then submitted to the laboratory for processing. The RTPCR test was run in accordance with the TaqMan Procedure. This sorting and testing process usually took a few days and the cities were immediately notified of the results. Finally, the county generated weekly maps in ArcGIS of sample locations (and their results) and distributed them to the cities.

2.2 The Mapping and Statistical Model

There were 1779 total past trapping locations that contained complete and usable data. These trap locations and dates spanned from the 2003 to the 2006 summer seasons (Figure 2.1). This map displays all of original samples and their WNv results, displayed above a map of the cities that are within Tarrant County.

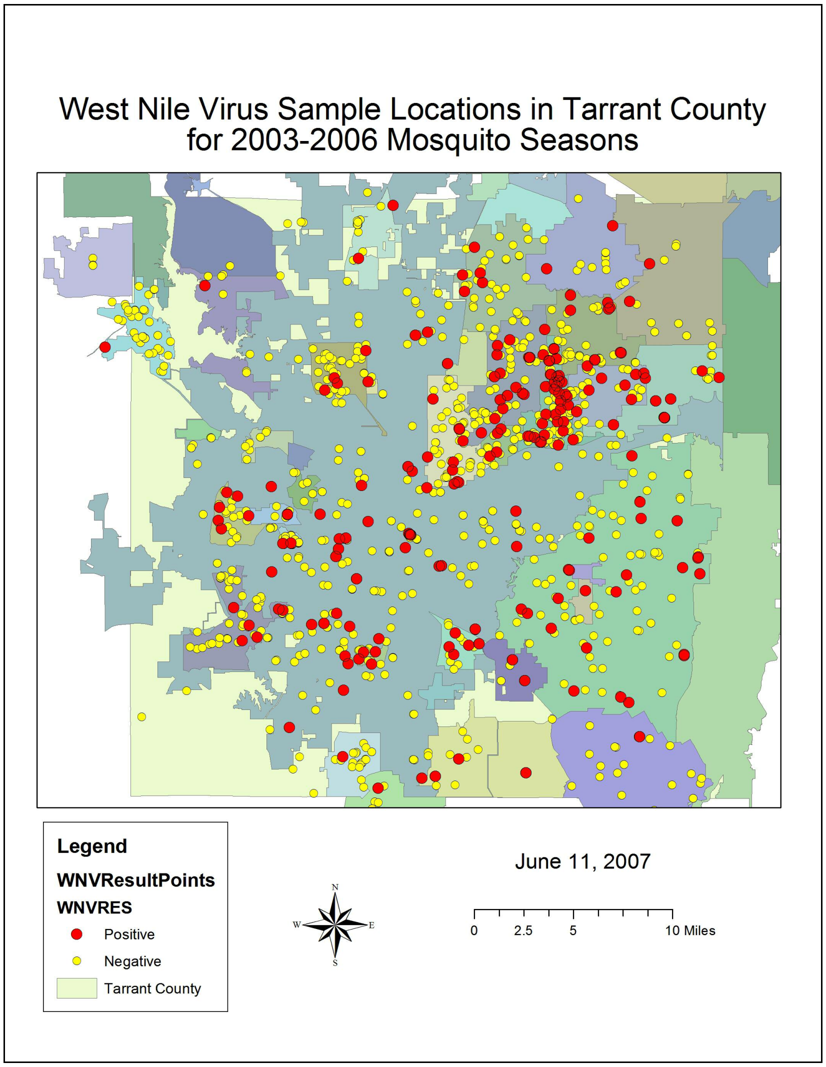

Figure 2.1 The 1779 Past Trap Locations

Each trapping season ran for 6 months, from May to October. I compiled all of these locations into a "trapping location database" containing the date of the trapping event, the latitude/longitude, the city that placed the trap, and the result of the RTPCR test for WNv (Table 2.1). The test results were displayed as binomial, 1 meaning a positive WNv test result and 0 being a negative WNv test result.

Table 2.1 Representative Data Sample for Statistical Analysis

MASTER_ID	YEAR	SAMPLEID	CITY	LATITUDE	LONGITUDE
0	2006	ARL001	Arlington	32.624240	-97.125740
1	2006	ARL002	Arlington	32.654440	-97.070120
2	2006	ARL003	Arlington	32.685970	-97.205570
3	2006	ARL004	Arlington	32.674830	-97.184980

Previous studies speak to the importance of trapping based on probabilities and risk based analysis (Ryan, 2004). Therefore, given the available data and the maps, I created an additional number of sample locations for future trap site predictions. I included these in the database to allow the model that was created to predict which of these grid locations would be more likely to produce a positive WNv result in the future. At current participation and effort levels, between 500 and 600 samples could be taken in a season by participating agencies. Thus I created a grid of 525 initial points throughout Tarrant County as proposed trap locations. The grid was created by locating the four corners of the county and generating points at decimal degree intervals within the boundaries to total 525 sample sites as presented in Figure 2.2. I used horizontal increments of 0.02125 and a vertical increment of 0.0215 in decimal degrees. There were thus a total of 2304 trap locations for analysis. 1779 of those were past trapping sites with a WNv result, and 525 future sites that have no WNv outcome yet known.

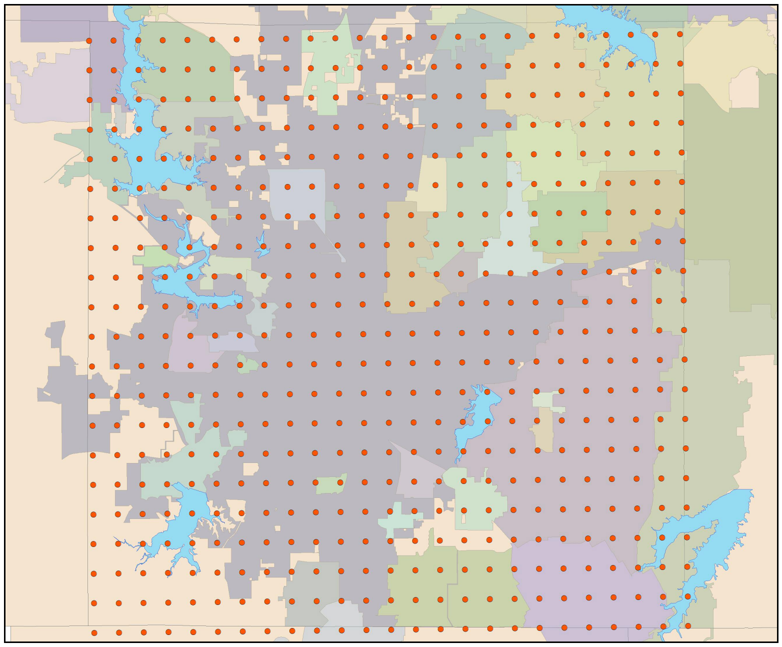

Figure 2.2 The 525 Future Trapping Locations

For the ArcGIS mapping analysis of the sample points, I sought out maps that most closely represent the information the four previous model studies (Cook, 2006; de Castro, 2004; Yiannakoulias, 2007; O Ruiz 2002). I acquired my maps for this study from the Tarrant County Public Health: GIS Division, Fort Worth GIS Department and the North Central Texas Council of Governments Data Clearing House. The maps I was able to acquire were: the 100 Year Floodplain Parcels of Tarrant County, the Census Bureau Urbanized Area of Tarrant County, Elevation of Tarrant County, the Census Bureau Population Density, NCTCOG Land Use, NCTCOG Vegetative Cover and City of Fort Worth Stormwater Outfall Locations.

I created fields in the sample location database and assigned values to each of the sample points based on data in the maps. The procedure for ascertaining geomorphologic characteristics for each trapping location was similar in most cases.

Each map with a variable of interest was loaded into ArcGIS as a layer beneath the 2304 trapping locations. The trapping locations were displayed on top of the map of interest to enable visual or computer aided analysis.

The first analysis was performed with the Tarrant County 100 Year Floodplain Parcel Map. The trapping locations were overlaid and displayed on top of this map. Then an 'Intersection Query' was run to define all trapping locations that fall within the floodplain parcels. The sample sites that "intersect," or fell within the floodplain parcels, were assigned a value of 1. Those that did not "intersect" and were located outside these parcels were assigned a value of 0. I included this map in an effort to reproduce the "stream density" value in Cooke's research (Cooke, 2006). Surface wetness is also used to predict the abundance of mosquito prevalence in past research, and this map may represent such information (Shaman, 2002).

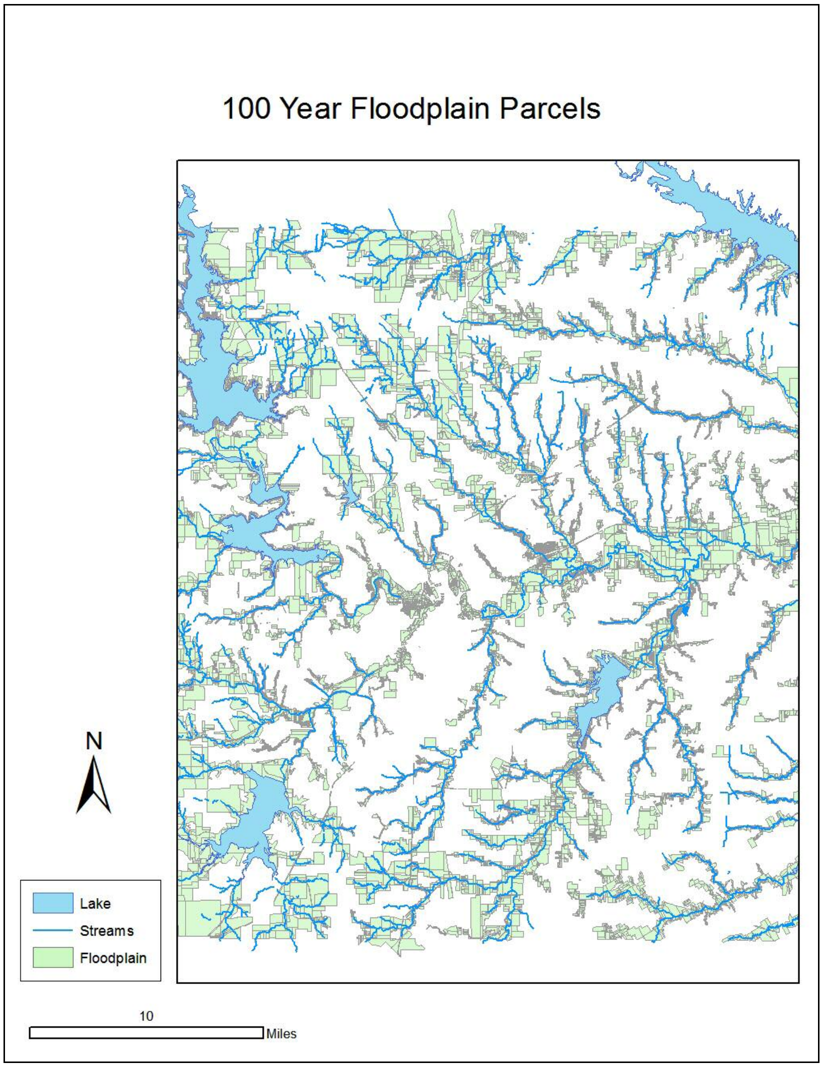

Figure 2.3 FEMA Designated 100 Year Floodplain Parcels

Next, a map of the Urbanized Areas of Tarrant County was loaded. This map contains areas within Tarrant County that have been designated by the Census Bureau as being "Urbanized." The sample locations were once again projected across the map as in Figure 2.4. An identical "Intersection Query" was performed, and when the sample locations fell within the Urbanized area, the samples were given a value of 1. Those locations that fell outside this region were assigned a value of 0. The use of the

Urbanized area map was intended to establish a similar relationship as observed in Subra's (1981) research and Cooke's (2006) model. These locations might be more likely to return positive results when trapped within an "Urbanized Area."

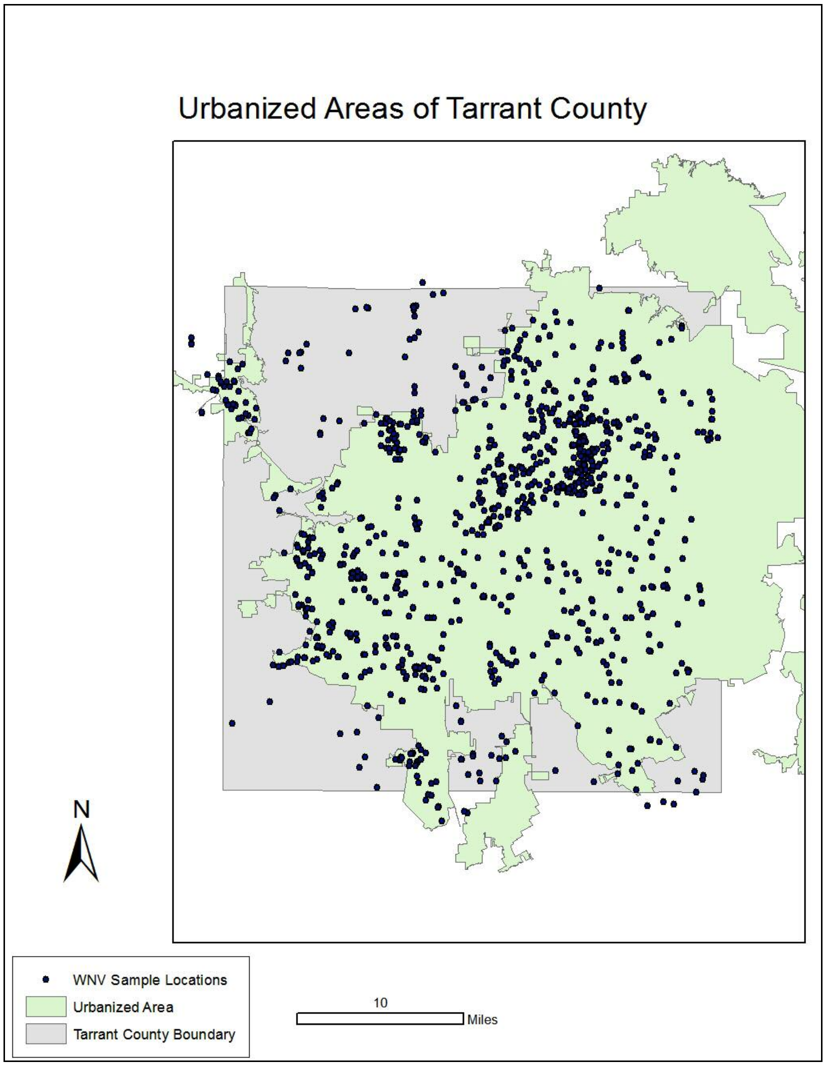

Figure 2.4 Census Bureau Classified Urbanized Area

To determine elevation, I used a graphic TIFF file that is a LIDAR image of the surface of the earth. This black and white image contains grey pixels of values from 70-450 shades of depth and is a Digital Elevation Map or DEM. These grey pixels represent elevation values from about 350 to 800 feet above sea level. This image is essentially a contour map of very high detail. For simplicity and ease of use, the elevations were coded into 5 categories (1-5): Lowest, Low, Medium, High and Highest. These categories were set to be an equal interval through each category level in ArcGIS. This helped ensure that in the statistical analysis, the levels could be treated as scale quantities. The trap locations were then imposed on the elevation image and the locations were manually assigned a value from 1 to 5. I included this elevation factor in the analysis to resemble the elevation and image analysis performed by Zou (2006) to predict ideal mosquito breeding habitats (Zou, 2006).

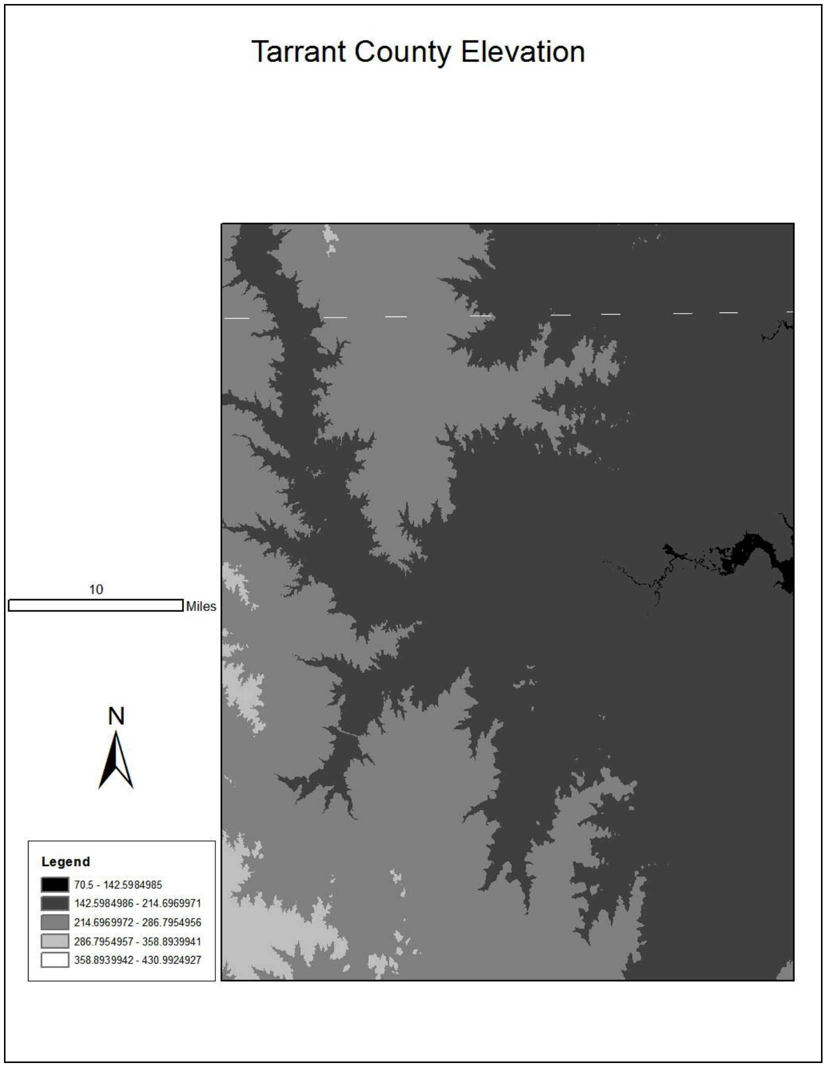

Figure 2.5 Digital Elevation Map of Tarrant County

To determine Population Density I used NCTCOG's Census Tract Population record map. I also used ArcGIS's ability to calculate area and determine the people per unit area. As this was a scalar value, the tracts were separated into 5 categories (1-5): Lowest, Low, Medium, High and Highest. As before, the categories were set to equal intervals to enable the data to be used as a ratio or scale quantity. I thus tested the association between higher population density and WNv positive mosquitoes that was demonstrated in O Ruiz's (2002) research. This use of population density is supported by additional papers (Subra 1981, Fyodorova, 2006). In areas that have higher population, there will likely be higher breeding locations and higher WNv mosquito incidence.

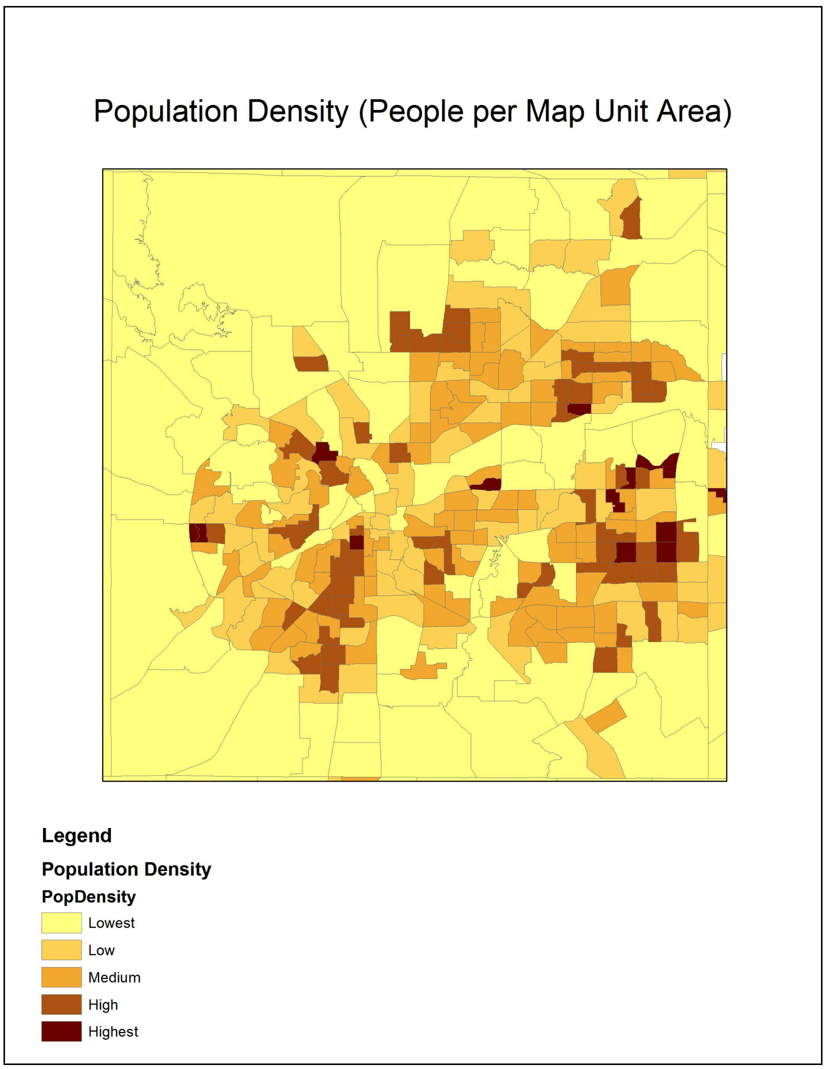

Figure 2.6 NCTCOG Census Tract Population Density

For a detailed description of land use, I obtained a map from NCTCOG depicting detailed descriptions of property tracts. The sampling points were intersected with this detailed land use map. Two of the most prevalent uses were “Single Family Residential” and “Industrial.” Along with those designations were the following: Residential Mobile Homes, Government/Education Group Quarters, Commercial Office, Commercial Retail, Government/Education Institutional, Commercial Hotel/Motel, Industrial, Infrastructure Transportation, Infrastructure Roadway, Infrastructure Utilities, Airports, Undeveloped Parking Garage, Airports Runway, Commercial Large Stadium, Dedicated Parks, Dedicated Landfill, Undeveloped Under Construction and Dedicated Flood Control. All of these uses I denoted as “Developed” and assigned the samples that occurred within these tracts a value of 1. Sample locations occurring within the “Undeveloped” land use were assigned a value of 0. These urban areas will likely demonstrate the *Culex quinquefasciatus* breeding success in places like dumpsters, tires and flower pots found in Hribar’s study in the Florida Keys (Hribar, 2007).

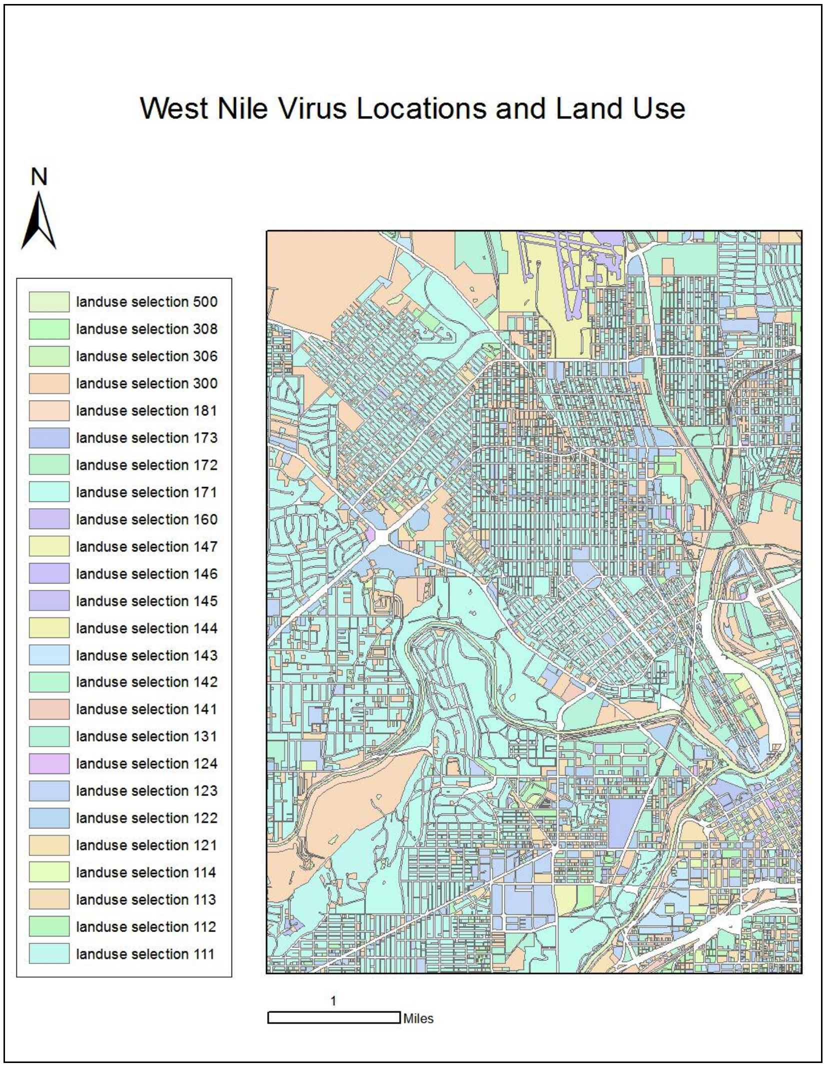

Figure 2.7 NCTCOG Land Use Parcels

Next, Vegetative Cover values were assigned based on *Culex quinquefasciatus* breeding habits. For a container and trash breeding mosquito which thrives in urban environments, sorting the samples into only 2 categories, Urban (1) or Vegetated (0) was appropriate. This map has many similarities to the Urbanized Area map, because it only distinguishes between Urban Vegetation and other types. Vegetation type, thickness and height above the ground can all determine trap success in capturing mosquitoes (Shone, 2006). This map represents similar data to what was used in the in the risk mapping performed in the O Ruiz (2002) research.

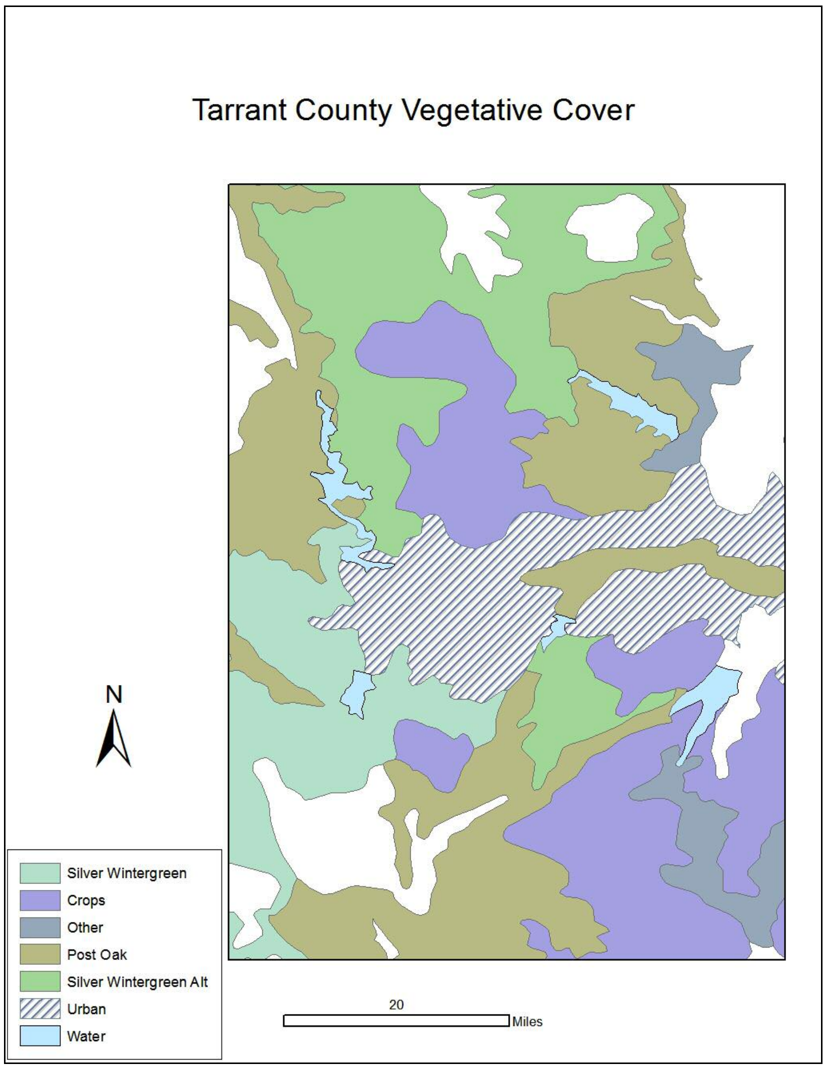

Figure 2.8 NCTCOG Vegetative Cover

Finally, given the likelihood of *Culex quinquefasciatus* to breed in and around Stormwater outfalls (Hribar 2007; Subra 1981), the City of Fort Worth provided a map of known outfalls within the city limits. A 500 foot buffer was generated around each location and then another intersection query was run to determine if any of the sample locations fall within these buffers. This distance was selected because the species of interest is not known to fly more than around 300 feet away from its hatch-out location (Subra, 1981). It would seem likely that proximity to the stormwater outfalls would increase the risk of returning a positive test (Allen, 2003).

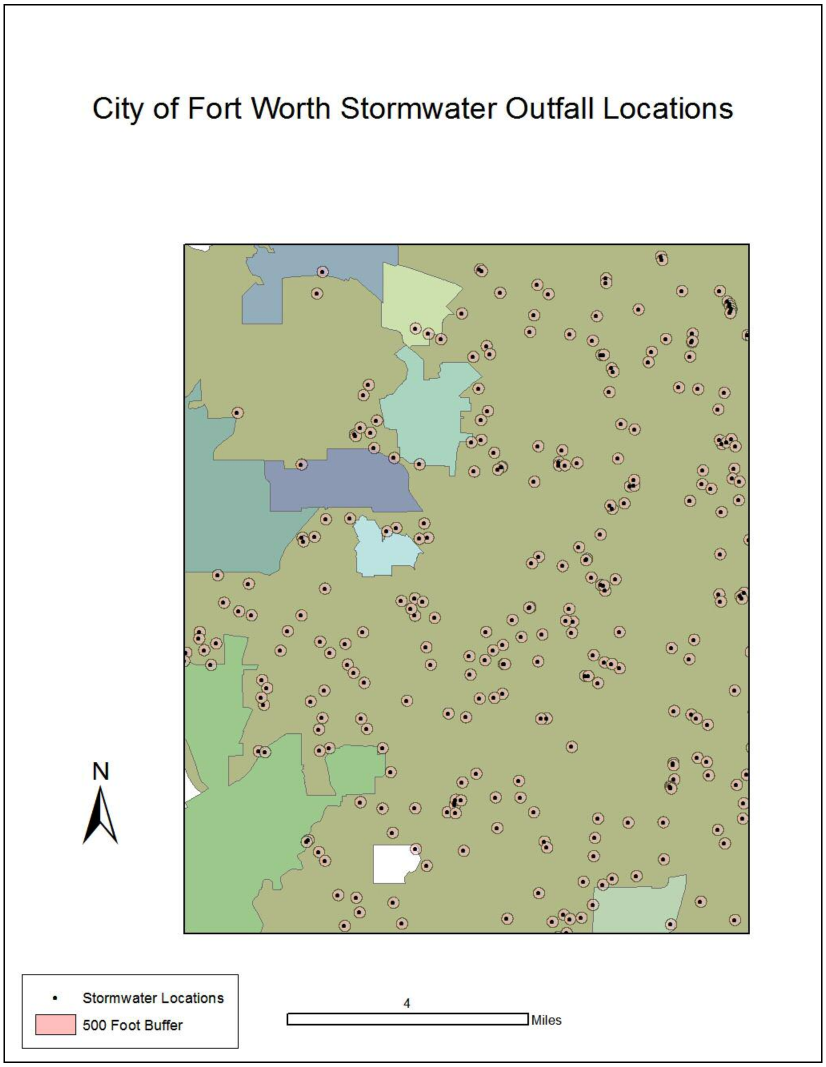

Figure 2.9 City of Fort Worth Stormwater Outfall Locations with a 500 ft Buffer.

2.3 Statistical Analysis

I generated the final data set containing values for all the mapped fields of the 2304 sample locations (an example is shown in Table 2.2). I applied two statistical analyses, the first to model the relationship between the independent variables derived from the maps and the WNv test outcome as the dependent variable. Second, I used the statistical model from the first analysis to predict future likelihood of the 525 added sample sites to detect WNv. These statistical analyses were conducted using the SPSS statistical software package.

Table 2.2 A Representative Sample of the Database of Trap Locations and their Map Variable Values

MasterID	LATITUDE	LONGITUDE	FLOODPLAIN	LandUse 1 = Developed	Urbanized	Vegetative Cover 1 = Urban
1	32.624240	-97.125740	0	1	1	0
2	32.654440	-97.070120	1	1	1	0
3	32.685970	-97.205570	0	1	1	0
4	32.674830	-97.184980	1	1	1	0

A preliminary Pearson Correlation analysis was performed to determine if any of the independent variables were correlated with each other. A correlation of greater than 0.7 between two of the independent variables was taken to indicate that they may be representing the same information in different ways. Ideally, none of the variables will be correlated with each other greater than 0.7, and they will be correlated with the dependent variable (WNv outcome) more than any other variable.

Binary (Binomial) logistic regression or Logit, was the primary analysis applied to this data. This analysis is a good tool to consider many independent variables and their influence on the binary outcome of a WNv test at a certain location. This technique is widely used in population predictions and disease risk modeling (e.g. O Ruiz, 2002; Yiannakoulias, 2007). The logistic regression quantified each independent variable's contribution to predicting the binary outcome of a positive or negative WNv result.

A significance level of 0.05, equivalent to a 95% confidence level was used to judge statistical significance. If the statistical significance value is greater than 0.05, the null hypothesis of no effect cannot be rejected, and the independent variable in question may not have any influence over the dependent variable and its effect cannot be distinguished from random "noise."

The test also generated an odds ratio indicating whether the independent variable is a risk factor which increases the odds, or a protective factor which decreases the odds of returning a positive WNv result. When 1 is subtracted from the odds ratio, a number representing risk is generated. If the value is positive, this indicates it is a risk factor, and sample locations that are positive for this variable are more likely to return a positive WNv sample result. If the value is negative, this indicates the variable is a protective factor and sample locations within it are less likely to return a positive WNv sample result.

The final output of this test was a predictive value for all 2034 sample locations, including the future 525 sample locations. This value reported the percentage likelihood of that particular sample location would return a positive WNv result. This value was calculated using all existing data about the first 1778 known locations to predict the unknown 525 sites.

CHAPTER 3

RESULTS

3.1 Statistical Results

None of the pairs of independent variables had Pearson Correlations exceeding about 0.40 (Table 3.1). This was below the criterion of 0.7 and indicates that none represent nearly the same information. Independent variables that may represent similar information are Vegetative Cover and Urbanized Area, since the Urban vegetation category overlaps with property in the Census Urbanized Area that is designated Urban. Nevertheless these designations are not excessively correlated (<0.7) among the sample locations that were present. However, the correlation coefficient between the two variables of 0.40 is greater than that of either of them with the dependent variable (WNv), 0.016 and 0.02, respectively. Although this result is not ideal, the low correlation between these two independent variables was taken to indicate that both are acceptable for use in the model.

Table 3.1 Pearson Correlation

		Floodplain	LandUse	Urbanized	VegCover	SW500ft	Elevation	PopDens	WNV
Floodplain	Pearson Correlation	1	-.300(**)	-.175(**)	-.072(**)	0.029	0.025	-.126(**)	-.048(*)
	Sig. (2-tailed)		0	0	0.001	0.163	0.225	0	0.044
	N	2304	2304	2304	2304	2304	2304	2304	1779
LandUse	Pearson Correlation	-.300(**)	1	.343(**)	.200(**)	-0.01	-.178(**)	.131(**)	.075(**)
	Sig. (2-tailed)	0		0	0	0.63	0	0	0.002
	N	2304	2304	2304	2304	2304	2304	2304	1779
Urbanized	Pearson Correlation	-.175(**)	.343(**)	1	.408(**)	.150(**)	-.368(**)	.280(**)	0.02
	Sig. (2-tailed)	0	0		0	0	0	0	0.401
	N	2304	2304	2304	2304	2304	2304	2304	1779
VegCover	Pearson Correlation	-.072(**)	.200(**)	.408(**)	1	.270(**)	-.271(**)	.394(**)	0.016
	Sig. (2-tailed)	0.001	0	0		0	0	0	0.508
	N	2304	2304	2304	2304	2304	2304	2304	1779
SW500ft	Pearson Correlation	0.029	-0.01	.150(**)	.270(**)	1	-.095(**)	.227(**)	-0.045
	Sig. (2-tailed)	0.163	0.63	0	0		0	0	0.058
	N	2304	2304	2304	2304	2304	2304	2304	1779
Elevation	Pearson Correlation	0.025	-.178(**)	-.368(**)	-.271(**)	-.095(**)	1	-.156(**)	-.070(**)
	Sig. (2-tailed)	0.225	0	0	0	0		0	0.003
	N	2304	2304	2304	2304	2304	2304	2304	1779
PopDens	Pearson Correlation	-.126(**)	.131(**)	.280(**)	.394(**)	.227(**)	-.156(**)	1	.073(**)
	Sig. (2-tailed)	0	0	0	0	0	0		0.002
	N	2304	2304	2304	2304	2304	2304	2304	1779
WNV	Pearson Correlation	-.048(*)	.075(**)	0.02	0.016	-0.045	-.070(**)	.073(**)	1
	Sig. (2-tailed)	0.044	0.002	0.401	0.508	0.058	0.003	0.002	
	N	1779	1779	1779	1779	1779	1779	1779	1779

** Correlation is significant at the 0.01 level (2-tailed).
* Correlation is significant at the 0.05 level (2-tailed).

Table 3.2 Binomial Logistic Regression Analysis

		B	S.E.	Wald	Df	P-value	Odds Ratio	95.0% C.I.for Odds Ratio	
								Lower	Upper
	Floodplain	-.237	.208	1.299	1	.254	.789	.525	1.186
	LandUse	.543	.220	6.067	1	.014	1.720	1.117	2.649
	Urbanized	-.214	.323	.439	1	.507	.807	.429	1.520
	VegCover	-.094	.165	.327	1	.567	.910	.659	1.257
	Elevation	-.676	.252	7.211	1	.007	.509	.311	.833
	PopDens	.374	.117	10.290	1	.001	1.454	1.157	1.827
	SW500ft	-.481	.238	4.092	1	.043	.618	.388	.985
	Constant	-1.158	.687	2.839	1	.092	.314		

Table 3.3 Summary of Results

Variable	Significant	Protective/ Risk Factor
LandUse	Yes	Developed = Risk
PopDens	Yes	High Density = Risk
Elevation	Yes	High = Protective
SW500ft	Yes	Near Outfall = Protective
Floodplain	No	On Floodplain = Protective
Urbanized	No	Urbanized = Protective
VegCover	No	Urban Vegetation = Protective

The results of the binomial logistic regression are summarized above (Table 3.2). The two primary fields of interest are the P-value and the Odds Ratio. A summary is presented in Table 3.3.

The population density variable was statistically significant (P = 0.001), and the Odds Ratio indicated a 45.4% increase in the likelihood of finding the virus with every categorical increase in population density. This is logical as, once again, where there are people there are breeding containers in and around homes. Areas of high population density seemed to have a significantly higher amount of positive sample results. This result is consistent with past research and the conclusion is statistically strong (Fyodorova, 2006; O Ruiz, 2002).

The elevation variable was categorized from 1 through 5 representing equal interval elevation levels Lowest, Low, Medium, High and Highest. The elevation variable shows high statistical significance (P = 0.007) and an Odds Ratio of 0.509. This indicates that for every categorical increase in elevation, it is 49.1% less likely to obtain a positive for WNv. It is unclear how a relatively small difference in elevation of 400 vertical feet would affect the success of the mosquito to breed or to transmit the virus. With large elevation changes, temperature can be very different and alter the behaviors of the mosquito (Zou, 2006) but that seems unlikely here. Although the model indicated that elevation is protective and the cause is unknown.

The land use variable was statistically significant (P = 0.014). Testing in a land parcel designated as Developed would be 72% more likely to return a positive result than other parcels. This would seem reasonable as the mosquito of interest is a container breeder. Areas that have this classification would most likely be in proximity to both underground storm drain systems and all of the breeding containers that residential homes usually provide like rain gutters, flower pots and swimming pools in disrepair (Hribar, 2007). Similarly industrial areas that contain waste and breeding containers such as used car tires would contribute to this problem and increase the chances of getting a positive sample result at these sites (Fyodorova, 2006).

The Stormwater Outfall variable was statistically significant (P = 0.043). Being within 500 feet of a stormwater outfall was protective, reducing the probability of a positive test result by 38.2%. These results were unexpected. Given this mosquito species' propensity to breed in and around storm drains and stormwater outfalls, it would seem logical that samples taken nearby would have an increased chance of testing positive (Allen, 2003). One possible explanation may be that all of the samples that were not near the mapped storm drains are not necessarily far from a storm drain. The data presented here indicated only outfalls located in the city limits of Fort Worth. Other cities certainly have outfalls and storm drains within their borders that were not present in this analysis.

The floodplain was indicated to be mildly protective, with the sample less likely to be positive in a floodplain parcel. Subtracting 1 from the odds ratio value yielded a value of -0.211, indicating that a sample taken from within a floodplain is 21.1% less likely to be positive for WNv. This may seem counterintuitive, but the mosquitoes trapped are trash breeders and not generally more successful around lakes and rivers (Shaman, 2002). This also confirms Cooke's (2006) findings as he concluded that areas with higher stream densities had lower risks of finding WNv (Cooke, 2006). However, the floodplain variable was not statistically significant (P = 0.254).

The Urbanized area variable was not statistically significant (P = 0.507). The majority of samples were taken in the area designated as Urbanized, so this did not provide much of a distinction upon which to separate negative from positive WNv results. The results indicated a 19.3% decreased likelihood of finding WNv inside an Urbanized area, but this protective characteristic of Urbanized area was not statistically significant.

The Vegetative Cover variable was also not statistically significant (P = 0.567). The odds ratio indicated it is weakly protective (9.0% reduction in probability of a positive result). This was not expected as most research indicated the *Culex quinquefasciatus* are more successful in urbanized settings (Subra, 1981), and thus positive results were expected to be more likely in the Urban vegetation category.

3.2 Surveillance Model Predictive Results

In the 525 future sample sites, the values returned for each location ranged from 0% to as high as 30%, representing the chance of returning a positive WNv result at that location. These values seemed appropriate as other average values in the past in Tarrant County have ranged year to year from as low as 4% overall to as high as 20% overall in 2003. Similarly, Dimenna (2006) found infection rates on average around 18% in his samples (Dimenna, 2006). The positive event is still unlikely but the results are encouraging as they indicate that the probability of obtaining positive results can be increased above the overall historical value of around 15%, by considering the risk and protective factors analyzed. Utilizing these, the model can give a useful description of each location and its chances of returning a positive WNv test result.

I generated a map to display these new risk values in a meaningful way. All of the 525 future sample locations were placed on a map that indicated what level of positive WNv likelihood these locations were calculated to have. Larger size dots on the map were used to indicate a higher chance of returning a positive result. These locations would be considered more likely to contain mosquitoes that are positive for WNv, and as such, the cities they fall into should consider trapping there consistently.

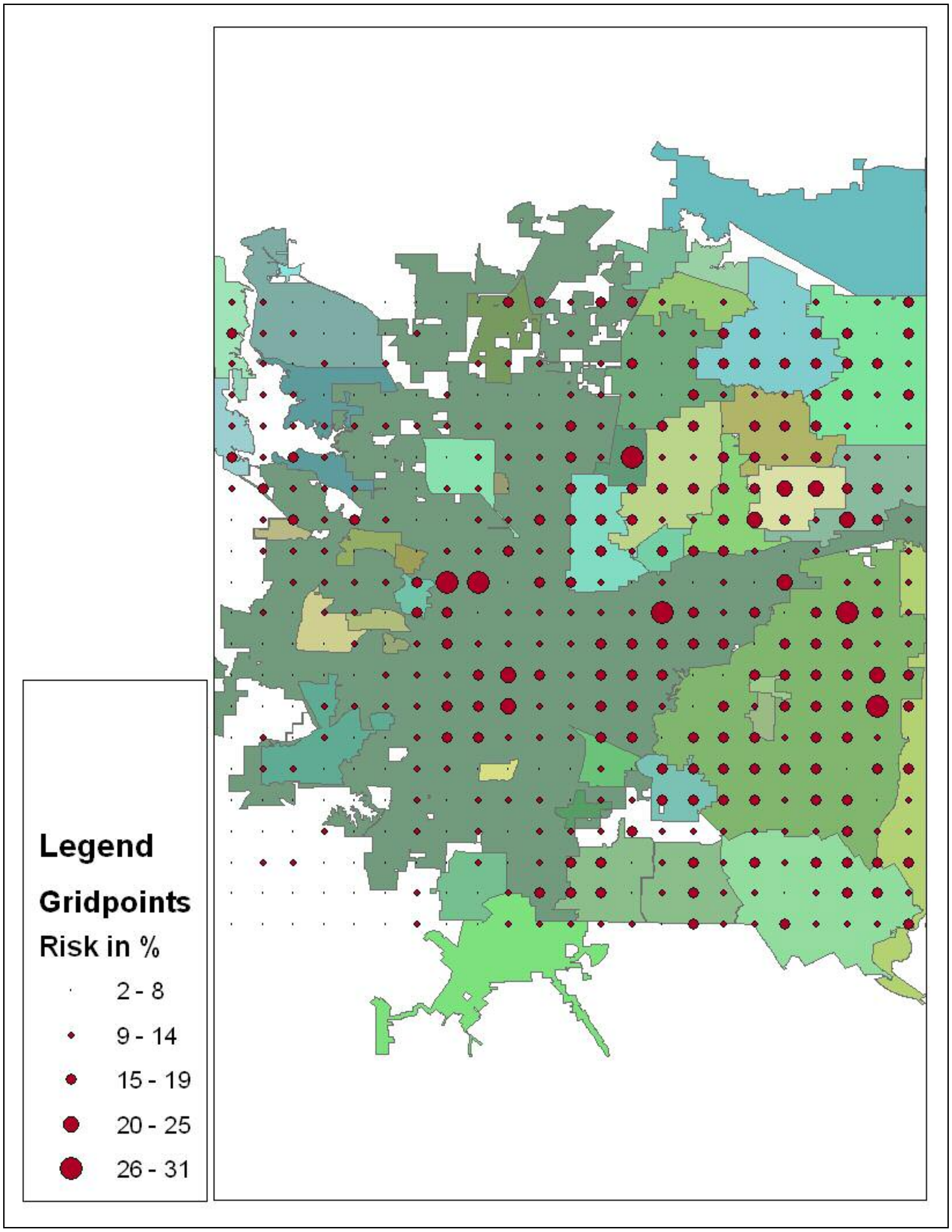

Figure 3.1 Proposed Sampling Grid with Priority for Higher Predicted WNv Outcomes

CHAPTER 4

DISCUSSION

4.1 Evaluation and Limitations of the Model

The binary logistic regression model yielded many useful results. The choice of using logistic regression to model these variables' influence on the outcome of the WNv test seemed appropriate given the type of data present and was supported by other research (Shaman, 2002; Yiannakoulias, 2007). Elevation, population density and land use are important factors that appeared to increase the likelihood of trapping positive mosquito pools. Land use alone was very predictive and may represent a coarse predictor of increased probability with no other considerations.

The calculated probabilities of positive results at the 525 future sample locations seemed to confirm that low-lying areas in the East and very densely populated areas in the West were more likely to return a positive sample test. The increase in likelihood from around 12% (original average incidence) to up to 30% in some sites indicates that while these variables affect the outcome, finding the virus is still unlikely even at the sites with highest probability of positive results. Once the grid sampling method is attempted, it may be possible to compare the predicted positive locations with the actual positive locations and predicted numbers over a season. A regression analysis of the new grid could be highly valuable.

There are many limitations to this analysis. The study assumes very little error in trap location placement and reported GPS Coordinates. Inherent in a consumer handheld GPS device is an induced error called Selective Availability that can exceed 300 feet. In actual field studies the errors seem to be around +- 10 feet.

Many of the sites were also located using Google Earth's Geocoding ability. Geocoding is the calculation of a latitude and longitude when only an address is known. This can also lead to errors of an unknown quantity as the geocode may not place the address in the correct location on the Earth.

There may be slight projection errors in ArcGIS's "On the Fly" Projection. The scale of these errors, in comparison with the size of the land in most of the maps could be considered negligible. I assumed ArcGIS mapped the Latitude and Longitude correctly and this model assumes a high level of accuracy.

In the statistical analysis, there were a couple areas for potential errors. The regression assumes all of the values assigned in each field were correct. Also the stormwater outfall locations were only represented within the city limits of Fort Worth, so any other stormwater outfall locations were not accounted for in the model.

Another weakness of this analysis is site selection bias. The method of site selection of the first 1779 samples is unknown. It is possible that areas that had returned positives in the past were trapped consistently and given closer scrutiny. This would inflate the likelihood of finding the virus as trapping was dictated by past experience. Similarly, areas within the cities that presented easier access, familiarity of a trapper with a specific location and general convenience may have overrepresented many trap locations.

The analysis of the data and predictions of which locations test positive may be in effect be an analysis of the presence of mosquito numbers in general. It is likely that this analysis reflects not just the likelihood of finding WNv, but of catching large numbers of mosquitoes. No analysis was conducted to determine the influence of numbers of mosquitoes trapped to WNv outcome. Though pooling large numbers of mosquitoes for testing is a common procedure, I am unaware of any research that compares trap counts with WNv positive rates. It seems likely that there would be a positive correlation between the number of mosquitoes trapped and the probability of testing positive. This unknown relationship is present in other research and is not unique to this study (Cooke, 2006).

Other factors a surveillance model might include are bird and human WNv positive locations (Rappole, 2000). This data was not in a complete and useful form at this time for the North Central Texas region. Certainly past research indicates that guiding trap choice and efforts with human WNv and bird deaths is preferable (McLean, 2006; Nasci, 2002).

4.2 Model Implementation into the Existing Surveillance Program

For the next trapping season, 2007 or 2008, Tarrant County could disseminate the future trapping location map (Figure 3.1) to their cities along with the actual latitudes and longitudes in which to trap. This would offer the cities much needed guidance for trap placement and provide justification for trapping and treating certain areas in Tarrant County. Having a highly detailed map, an ArcGIS layer of WNv locations and surveillance test results will benefit both smaller and large cities. This method of surveillance using both a grid coupled with geomorphological characteristics and prior surveillance results will give a much better picture of the presence of the virus and where to expect its occurrence in the future.

This research may be used as justification for attempts to eradicate adult mosquitoes. Many cities are active in such efforts and are looking for methods to guide their spraying activities. New research suggests that the risk of humans contracting WNv is now greater than the risk of humans being harmed by most methods of controlling adults (Peterson, 2006). This research could be used in the absence of any other guiding information. More reliable would be a citywide analysis of the source maps which were used in this paper which will be made available.

4.3 Future Research

Future research could occur in several ways. The first possibility would be to verify the proposed grid. After a season of sampling, a statistical comparison of the predicted WNv positive locations and actual positive locations would check the validity of the predictive model. A grid sampling system would enhance any surveillance program. Once the cities place traps in the majority of the grids locations, a study could be done to see how prevalent the virus was in grid locations with a higher predicted positive outcome. Regression analyses could be performed to related variables of interest to the presence of West Nile virus, and construct improved predictive models.

APPENDIX A

METADATA FOR SOURCE MAPS

Appendix A: Metadata for Source Maps

1.1 FEMA 100 Year Floodplain Parcels: Tarrant County Public Health, GIS Division 2006.

1.2 NCTCOG Census Bureau Designated Urbanized Area: http://www.dfwmaps.com/clearinghouse/metadata/urbanized_area.html

1.3 Digital Elevation Map Tarrant County: Tarrant County Public Health, GIS Division 2006.

1.4 NCTCOG Census Tract Population Density: http://www.dfwmaps.com/clearinghouse/metadata/tract.html

1.5 NCTCOG Land Use Parcels http://www.dfwmaps.com/clearinghouse/metadata/landuse.html

1.6 NCTCOG Vegetative Cover: No Citable reference provided by NCTCOG.

1.7 City of Fort Worth Stormwater Outfall Locations. Detailed Metadata Unavailable, but information acquired from Fort Worth GIS Department. Released 2006.

APPENDIX B

SPSS STATISTICAL ANALYSIS RAW OUTPUT

Appendix B: SPSS Statistical Analysis Raw Output

Logistic Regression

Notes

Output Created		21-Jun-2007 17:22:45
Comments		
Input	Data	F:\Thesis\Statgood621a.sav
	Active Dataset	DataSet8
	Filter	<none>
	Weight	<none>
	Split File	<none>
	N of Rows in Working Data File	2,304
Missing Value Handling	Definition of Missing	User-defined missing values are treated as missing
Syntax		LOGISTIC REGRESSION WNV /METHOD = ENTER Floodplain LandUse Urbanized VegCover Elevation PopDens SW500ft /PRINT = CI(95) /CRITERIA = PIN(.05) POUT(.10) ITERATE(20) CUT(.5) .
Resources	Elapsed Time	00:00:00

Case Processing Summary

Unweighted Cases(a)		N	Percent
Selected Cases	Included in Analysis	1,779	77.2
	Missing Cases	525	22.8
	Total	2,304	100.0
Unselected Cases		0	0.0
Total		2,304	100.0

a. If weight is in effect, see classification table for the total number of cases.

Dependent Variable Encoding

Original Value	Internal Value
0.00	0
1.00	1

Block 0: Beginning

Block

Classification Table(a,b)

	Observed		Predicted		
			WNV		Percentage Correct
			0.00	1.00	
Step 0	WNV	0.00	1,550	0	100.0
		1.00	229	0	0.0
	Overall Percentage				87.1

Variables in the Equation

		B	S.E.	Wald	df	Sig.	Exp(B)
Step 0	Constant	-1.912	0.071	729.622	1	0.000	0.148

Variables not in the Equation

			Score	df	Sig.
Step 0	Variables	Floodplain	4.057	1	0.044
		LandUse	10.063	1	0.002
		Urbanized	0.705	1	0.401
		VegCover	0.440	1	0.507
		Elevation	8.599	1	0.003
		PopDens	9.397	1	0.002
		SW500ft	3.601	1	0.058
	Overall Statistics		33.157	7	0.000

Omnibus Tests of Model Coefficients

		Chi-square	df	Sig.
Step 1	Step	34.993	7	0.000
	Block	34.993	7	0.000
	Model	34.993	7	0.000

Model Summary

Step	-2 Log likelihood	Cox & Snell R Square	Nagelkerke R Square
1	1331.114(a)	0.019	0.036

a. Estimation terminated at iteration number 5 because parameter estimates changed by less than .001.

Classification Table(a)

	Observed		Predicted		
			WNV		Percentage Correct
			0.00	1.00	
Step 1	WNV	0.00	1,550	0	100.0
		1.00	229	0	0.0
	Overall Percentage				87.1

a. The cut value is .500

Variables in the Equation

		B	S.E.	Wald	df
Step 1(a)	Floodplain	-0.237	0.208	1.299	1
	LandUse	0.543	0.220	6.067	1
	Urbanized	-0.214	0.323	0.439	1
	VegCover	-0.094	0.165	0.327	1
	Elevation	-0.676	0.252	7.211	1
	PopDens	0.374	0.117	10.290	1
	SW500ft	-0.481	0.238	4.092	1

Constant	-1.158	0.687	2.839	1

Variables in the Equation

		Sig.	Exp(B)	95.0% C.I.for EXP(B)	
				Lower	Upper
Step 1(a)	Floodplain	0.254	0.789	0.525	1.186
	LandUse	0.014	1.720	1.117	2.649
	Urbanized	0.507	0.807	0.429	1.520
	VegCover	0.567	0.910	0.659	1.257
	Elevation	0.007	0.509	0.311	0.833
	PopDens	0.001	1.454	1.157	1.827
	SW500ft	0.043	0.618	0.388	0.985
	Constant	0.092	0.314		

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BIOGRAPHICAL INFORMATION

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