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Introduction

West Nile Virus (Flavivirus) has become endemic in the United States and Canada. It was first identified in North America in 1999. This virus has become a global threat to both humans and animals due to its morbidity and mortality in humans, birds, and horses. This study aims to contribute to the understanding of the distribution and abundance of two mosquito species, *Culex restuans* and *Culex pipens*, which are both vectors for West Nile virus in regions of the US, where these two species overlap. Prior to the study, it is understood that these two species play an important role in spreading West Nile virus, however, these two species are often lumped together creating an issue in determining the precise epidemiology of West Nile Virus. The authors aim to address two central questions. First, how do ecological variables such as temperature, precipitation, and habitat type affect the abundance of *Culex restuans* and *Culex pipens*. And the second question is how predictive risk maps can help improve understanding of the ecological and spatial distribution in the regions where these two species exist together. These questions are important because to help predict the transmission cycle of West Nile virus it is imperative to understand the spatial and temporal distribution of these species of mosquitoes.

Methods

The Culex restuans and Culex pipens populations in Pennsylvania, USA are used in this study. Pennsylvania provides a diverse landscape because it has the Appalachian Mountains, urban areas, and agricultural regions. The climate is generally humid in the summers and cold in the winter. But these conditions vary depending on elevation and the proximity to bodies of water. The Pennsylvania Department of Environmental Protection conducted mosquito surveillance from 2002 to 2016 for West Nile virus by using gravid traps which work by capturing adult mosquitoes. These traps were set from April to September and the data collected included species abundance. Culex restuans and Culex pipens were identified using morphology characteristics. Ecological factors were then considered that could impact mosquito abundance. These variables include temperature, precipitation, snow water equivalent, elevation, land cover, normalized difference water index, Julian date, and year. These variables were selected due to previous research that linked them to the biology of Culex restuans and Culex pipens mosquitoes. These variables all affect the physiology, reproduction, and survival of these mosquito species. Each variable was obtained from raster data by year and the researchers then linked this data to the mosquito trap observations using R. By linking the two data this allowed the researchers to analyze how different environmental conditions affected the abundance of the two species. To analyze the Culex restuans and Culex pipens abundance data the researchers used hierarchical Bayesian spatial models in the program R. They specifically used stochastic partial differential equations (SPDEs) to model the spatial relationships between the two species. The researchers then used Poisson regression models to account for the number of Culex restuans and Culex pipens and adjusted the models using zero inflated Poisson negative binomial distributions if there were an excess in zeros. The spatial component was modeled by utilizing a random field, using the trap locations, and the spatial effect was spread across the study area. After the models were validated using leave-one-out-cross validation, they were used to predict the risk of transmission of West Nile Virus from the abundance of Culex restuans and Culex pipens.

Results

Based on the distributed traps from the study, the average amount of Culex restuans caught in each trap was 27 with a standard deviation of ± 66.8, ranging from 0-2246 caught in the traps (21% of traps had zero mosquitoes captured). The average amount of Culex pipiens caught in each trap was 12.7 with a standard deviation of ± 40.0, ranging from 0-1460 caught in the traps (37% of traps had 0 mosquitoes captured). Based on Bayesian terms defining statistically important relationships, there is a 95% interval to that of the coefficient (without including zero). From the R-INLA models representing the top different ecological factors, there were multiple positive and negative relationships seen between these factors and the C. restuans and C. pipiens, as seen in table 1. For C. restuans their abundance had a negative relationship with development, temperature and elevation; and positive relationship with precipitation. For C. pipiens, their abundance had a positive relationship with development but negative relationship with snow and normalized difference water index (NDWI). In a line plot of the posterior means for the effect of Julian date on abundance of both species, the relationship shifts with respect to each other can be seen visually. On Julian date 185 (July 4th), a switch is seen between the effect of abundance on Julian date for C. Pipiens and C. Culex (Figure 1). The effect of space on the abundance of these species can also be seen through predictive models (Figure 2), in which the negative values indicate special effect cause a decrease in abundance, zero values indicate no effect, and positive values indicate increase in abundance. Risk maps showed that C. restuans had low abundance within the largest city of Pennsylvania and high abundance in agricultural and rural areas of Pennsylvania, whereas C. pipiens had highest abundance within the two largest cities of Pennsylvania (Figure 3).

Species	Variable	Mean	SD	Median	$Q_{0.025quant}$	$Q_{0.975quant}$
Culex restuans	(Intercept)	2.965	0.086	2.965	2.797	3.133
	develop	−0.341	0.045	−0.341	−0.430	−0.253
	temp	−0.021	0.002	−0.021	−0.025	−0.018
	precip	0.061	0.002	0.061	0.056	0.066
	elev	−1.098	0.129	−1.098	−1.351	−0.844
Culex pipiens	(Intercept)	−0.538	0.128	−0.538	−0.788	−0.287
	develop	0.550	0.049	0.550	0.455	0.646
	snow	−0.004	0.000	−0.004	−0.005	−0.004
	NDWI	−1.002	0.058	−1.002	−1.115	−0.889

Table 1: Numerical summary showing the positive and negative relationships between the top R-INLA models of different ecological factors and the Culex restuans and Culex pipiens species.
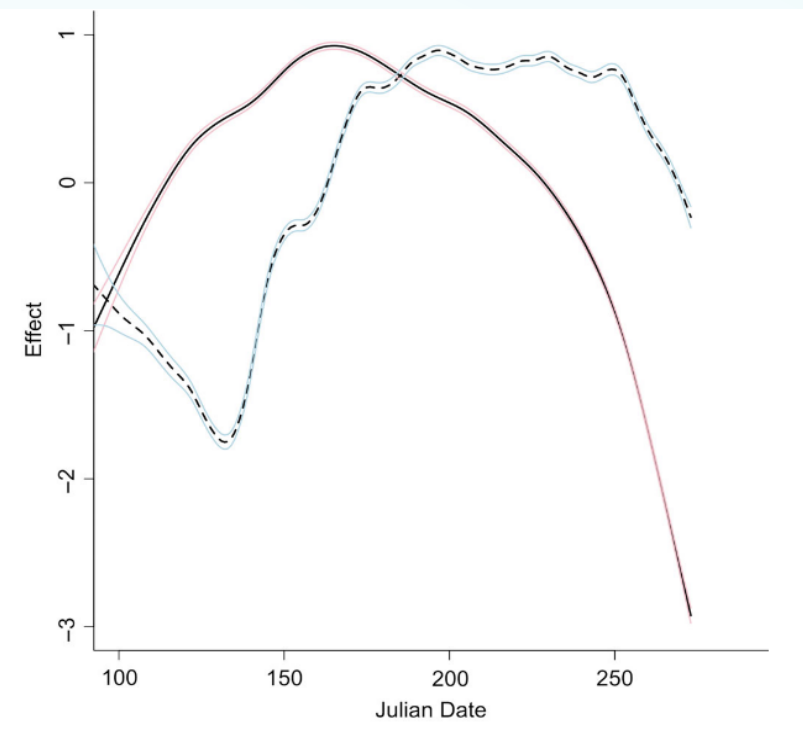

Figure 1: Effect of Julian date on abundance of each mosquito species. Culex restuans represented by solid black line and solid pink lines. Culex pipiens represented by dashed black line and solid blue lines. Colored lines representing 95% credibility intervals of the posterior means, represented by the black lines.
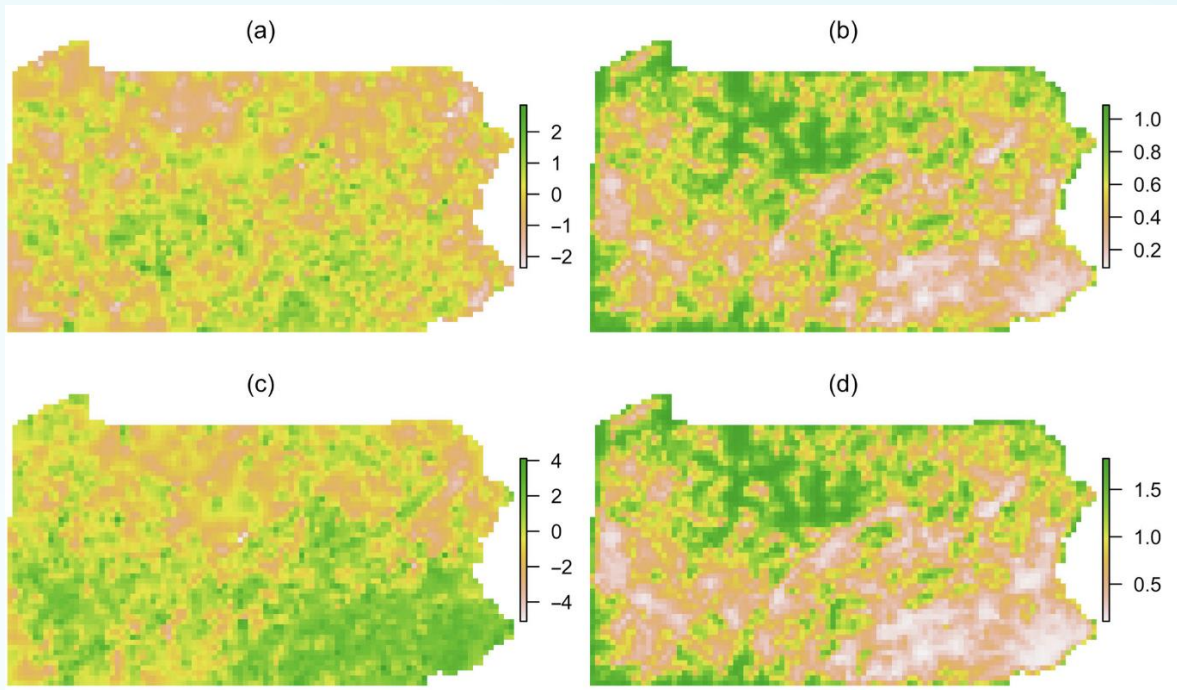
Figure 2: Posterior mean of spatial effect on abundance of Culex restuans on map of Pennsylvania, based on top predictive model (a), its standard deviation (b). Posterior mean of spatial effect on abundance of Culex pipiens on map of Pennsylvania, based on top predictive model (c), its standard deviation (d).

Discussion

The ecology and epidemiology of mosquito vectors, C. restuans and C. pipiens, of West Nile virus can be further assessed by the novel use of spatial statistical modeling, by assessing spatial distributions of abundance. The separate analysis of these two species provided a deeper understanding of the ecology and spatial distributions that are affected by the coexistence of these species in different regions. An effective analysis was able to be performed after data was collected from mosquito surveillance, consistent with other studies. Spatial autocorrelation was accounted for by incorporation of special random effect to the models. It was found that certain ecological factors including temperature, elevation, habitat development, and precipitation, have an affect on the abundance of C. restuans and C. pipiens. In addition to abundance, these ecological factors also have impacts on the population dynamics of these Mosquito species. Previous studies had yet to distinguish between the two species, therefore, the coexistence and the abundance with respect to time, ecological factors, and space, were further studied using predictive models. It was found that the abundance of C. restuans and C. pipiens were associated with different ecological factors, indicating that they may have different roles as vectors of West Nile virus. It was also found that C. restuans has a predominately higher abundance in regions that are less developed (rural and suburban areas), and C. pipiens has a predominately higher abundance in the largest cities of the state. Over the course of the year, the two species tend to overlap in the warmer months and experience a crossover in which they switch in abundance amounts. These factors all play important roles in assessing the different risks and overall epidemiology of WNV. Assessing this risk, especially for human populations, is dependent on mosquito surveillance data. This data allows for visualization and measurement through spatial scales, done via R-INLA with SPDE approach to compute large datasets and complying to Bayesian models without interfering with spatial models.
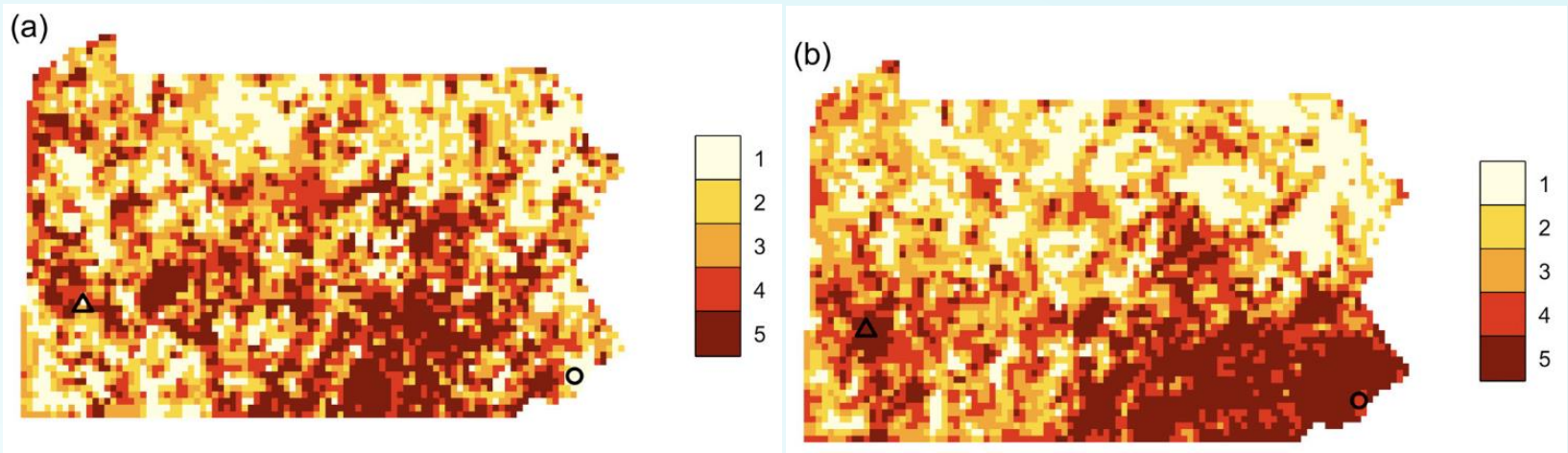
Figure 3: Risk map (1-low risk to 5-high risk) in Pennsylvania on June 15, 2011, of the predicted mean abundance of Culex restuans (a) and Culex pipiens (b). Two largest cities depicted, Philadelphia as a circle and Pittsburgh as a triangle.