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Estimation of Dispersal Distances of *Culex erraticus* in a Focus of Eastern Equine Encephalitis Virus in the Southeastern United States

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ABSTRACT Patterns of mosquito dispersal are important for predicting the risk of transmission of mosquito-borne pathogens to vertebrate hosts. We studied dispersal behavior of *Culex erraticus* (Dyar & Knab), a potentially significant vector of eastern equine encephalitis virus (EEEV) that is often associated with foci of this pathogen in the southeastern United States. Using data on the relative density of resting adult female *Cx. erraticus* around known emergence sites in Tuskegee National Forest, Alabama, we developed a model for the exponential decay of the relative density of adult mosquitoes with distance from larval habitats through parameterization of dispersal kernels. The mean and 99th percentile of dispersal distance for *Cx. erraticus* estimated from this model were 0.97 and 3.21 km per gonotrophic cycle, respectively. Parameterized dispersal kernels and estimates of the upper percentiles of dispersal distance of this species can potentially be used to predict EEEV infection risk in areas surrounding the Tuskegee National Forest focus in the event of an EEEV outbreak. The model that we develop for estimating the dispersal distance of *Cx. erraticus* from collections of adult mosquitoes could be applicable to other mosquito species that emerge from discrete larval sites.

KEY WORDS eastern equine encephalitis, insect dispersal, GIS-based modeling, *Culex erraticus*, larval habitat

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Urbanization and the accompanying modification of natural landscapes are increasing human exposure to mosquito-borne pathogens (Norris 2004, Pimentel et al. 2007, Patz et al. 2008). Predictions of such increases follow directly from Pavlovsky's theory of the natural nidality of transmissible diseases, which states that transmission of a vector-borne pathogen to humans occurs via association with the natural focus of the pathogen, with the focus defined as the specific conditions of habitat and geography that allow for maintenance of the natural transmission cycle of the pathogen (Pavlovsky 1966). Development adjacent to natural habitats such as wetlands increases the frequency of association between humans and isolated pathogen foci, either through the encroachment of human populations on foci or through areal expansion of foci themselves via habitat alteration that creates novel breeding sites for mosquito vectors (Norris 2004). Delineation of the geographic boundaries of mosquito-borne disease foci is thus necessary for accurate quantification of the degree of spatial association between humans and disease-causing pathogens.

Such delineation is particularly relevant in the case of eastern equine encephalitis virus (EEEV), a mosquito-borne pathogen that exhibits relatively stable foci and that is the most severe of the arboviral encephalitides in the United States. The human mortality rate of persons infected with EEEV is 30–40% (Whitley and Gnann 2002). Survivors of infection suffer mild to severe neurological damage and commonly require expensive, long-term medical care (Villari et al. 1995). Horses and gamebirds are also commonly infected with the virus and experience mortality rates >80% (Scott and Weaver 1989).

Although it is an extremely pathogenic disease of humans and horses, EEEV is one of the rarer viral encephalitides causing clinical infections in the United States. The rarity of EEE cases may be due, in part, to the geographic isolation of the virus from areas of human habitation. EEEV is endemic to freshwater swamp habitats, where its primary enzootic vector, *Culiseta melanura* (Coquillett), and avian reservoir hosts are sympatric (Scott and Weaver 1989). As such, delineation of the boundary zones surrounding these foci is critical to identifying high-risk areas as population growth and expansion in the Southeast potentially lead to human encroachment on EEEV foci (Wear and Greis 2001, Alig et al. 2004).

In the southeastern United States, the mosquito species *Culex erraticus* (Dyar & Knab) has recently

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been identified as a bridge vector that may play a key role in the transmission of EEEV to humans and horses (Cupp et al. 2003, 2004; Cohen et al. 2009). This mosquito is a moderately competent vector of EEEV and feeds on both birds and mammals (Chamberlain et al. 1954, Hassan et al. 2003). Its typical larval habitat in the southeastern United States is permanent bodies of fresh water overgrown with surface plants (Horsfall 1955). These larval habitats can overlap areas where EEEV is endemic, and in such areas, relative densities of adult *Cx. erraticus* are high compared with other potential bridge vector species of the virus (Cupp et al. 2003, 2004).

A previous study of the dispersal behavior of *Cx. erraticus* indicated that it is a long-distance flier with a maximum flight range of 1.4–2.2 km and a mean dispersal distance of 0.73 (±0.61) km (Morris et al. 1991). One approach to delineating the boundaries of areas where humans would be at risk of EEEV infection is to buffer all larval sites in an EEEV focus with a distance equivalent to the upper limit of the maximum flight range of *Cx. erraticus.* Flight distance estimates for *Cx. erraticus* reported by Morris et al. (1991) were based upon a mark-release-recapture approach, but these results must be viewed with caution for several reasons. First, the mosquitoes used in the study were not dispersing from their natural emergence or oviposition site, but from an arbitrary location in the middle of a wastewater treatment facility. Because the flight range of mosquitoes is known to vary with habitat (Silver 2008), mosquitoes released in this environment may display different patterns of dispersal relative to mosquitoes dispersing from a natural emergence site. Second, mosquitoes were released in the morning in an open, sunny area. *Cx. erraticus* seeks resting sites during the morning hours to avoid desiccation (Gray et al. 2010) and would likely undertake an initial movement in search of such a suitable resting site. Finally, marking, trapping, and handling mosquitoes may alter mosquito dispersal behavior (Silver 2008). These potential complications with the previous mark-release-recapture study highlight the need for the development of alternative approaches to estimating dispersal distances of *Cx. erraticus.*

In the current study, we developed new estimates of the dispersal distance of *Cx. erraticus* that do not rely on mark-release-recapture methods. Because *Cx. erraticus* typically oviposits in rather large, easily located bodies of water, we were able to identify the most likely sites of emergence in a study area in central Alabama. We then parameterized a dispersal kernel for this species using the straight-line distances between the sampling sites where adult females were collected and their putative sources of emergence. Dispersal kernel parameterization is an approach to the study of dispersal in other organisms, most notably angiosperms. We develop a novel application of such seed dispersal models to the study of dispersal of *Cx. erraticus.* Our goal was to both estimate the dispersal distances of *Cx. erraticus* and assess the general utility of seed dispersal models for studying mosquito dispersal and predicting relative mosquito densities on a local scale.

Materials and Methods

Field Methods. We estimated the dispersal distances of female *Cx. erraticus* using data on the relative density of adult and larval mosquitoes collected between 2006 and 2009 in Tuskegee National Forest (TNF) in Macon County, Alabama. TNF has served as the site of an ongoing study of the vector and vertebrate host dynamics of EEEV since 2001 and is described more fully in Cupp et al. (2003). Briefly, the study site encompassed a 28-km^2 circular area centered on a core wetland complex located ≈3.0 km from the town square of the city of Tuskegee (32°38′40″N, 85°25′59″W).

In 2006–2008, we sampled the adult mosquito population within the study site by aspirating resting mosquitoes from artificial shelters. In 2006, we used a variety of shelter types, including fiber pots, resting boxes, and 50-gallon plastic cans, but in 2007 and 2008 we exclusively used 50-gallon black plastic cans, the most attractive type of shelter for resting mosquitoes in TNF (Burkett-Cadena et al. 2008). Because the shelters used in 2006 varied in attractiveness to female mosquitoes, the data we used for the 2006 analysis were derived only from fiber pots and resting boxes, which showed no difference in attractiveness (Burkett-Cadena et al. 2008). Restricting mosquito samples to the same type of collecting container within any year assured that attractiveness of shelters ultimately used in our analyses varied between, but not within, years.

The number and locations of the adult sampling sites also varied between years. In 2006, six sampling sites were spaced ≈0.43 km along each of five transects radiating out 3.0 km from a point at the center of the study site (Fig. 1). In 2007 and 2008, seven sampling sites were located every 0.19 km along six transects radiating 1.5 km (Fig. 2). We excluded six sampling sites, which were either moved between 2007 and 2008 or were adjacent to private lands on which we were unable to sample for larval mosquitoes. As such, the data used in our analyses originated from 26 of the 31 sampling sites where fiber pots and resting boxes were used for collection in 2006, and 41 of the 43 sampling sites from 2007 and 2008.

Adult mosquitoes were collected between March and October, with regular sampling occurring between June and September. In 2006, regular sampling consisted of seven sampling intervals, each of which was 2 wk in length and during which one mosquito collection was made at all 26 sampling sites. In 2007 and 2008, we collected mosquitoes once at all 41 sampling sites during each of 15 sampling intervals, with each sampling interval lasting 1 wk. Results of any sampling outside of these regular sampling intervals were excluded from our analyses, so that the number of mosquito samples collected is constant across sites sampled in the same year. After collection in the field, mosquitoes were transported to the laboratory, anesthetized with CO_2, and sorted on a chill table by species and gender.

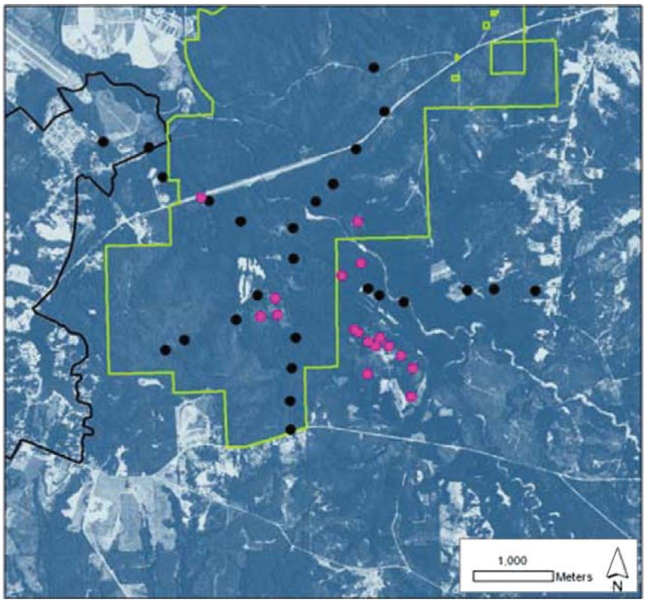

Fig. 1. Map of the EEEV study area in the TNF study area during 2006. Forest boundaries are shown in green, and the political boundary of the city of Tuskegee is shown in black. Black circles represent the locations of the adult sampling sites in 2006, and pink circles mark the centroids of all larval ponds of *Cx. erraticus.* (Online figure in color.)

We also sampled all permanent ponds within the study site for mosquito larvae to identify *Cx. erraticus* larval habitats, and hence the sites of emergence of the population under study. We censused the ponds from mid-July to mid-September in 2009 by repeatedly dip sampling along the perimeter of each pond. Thirty

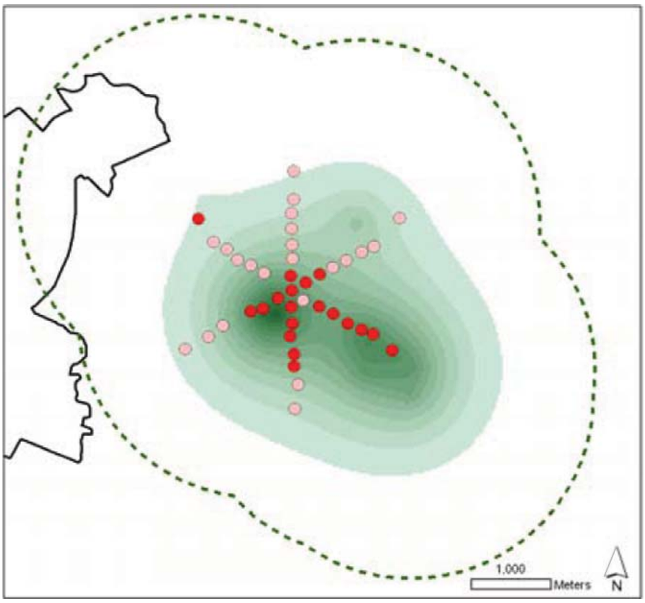

Fig. 2. Map of the EEEV study area in the TNF study area during 2007 and 2008. The dotted green line delineates the area beyond which a *Cx. erraticus* female that emerges from any of the outer larval sites in the TNF has <0.05 probability of dispersing. Circles represent the locations of adult sampling sites in 2007 and 2008, with sites with a low rate of capture of *Cx. erraticus* coded as light pink, and sites with high rates of capture coded as red. Classification of sampling sites is based on whether the total number of *Cx. erraticus* adult females was below or above the median count among all sampling sites during 2007–2008. The total height of dispersal kernels as parameterized in the area model centered on each larval pond, weighted by pond area, and overlapping in each pixel is shown with equal interval symbology ranging from lowest (white) to highest (dark green). (Online figure in color.)

dips were taken at 100-m intervals along the perimeter of each pond using a larval dipper. All larvae collected from each sampling point on a pond perimeter were transported back to the laboratory, heat killed, and sorted by species.

Dispersal Distance Estimation. Our approach to estimating the dispersal distances of *Cx. erraticus* was based on fitting a model of exponential decay with distance to *Cx. erraticus* relative density data from adult sampling sites. The model we used was a special case of the general set of models originally developed to estimate the number of seeds arriving at seed traps from multiple source trees distributed throughout a landscape (Clark 1998; Clark et al. 1998, 1999). Such seed dispersal models specify the number of seeds at trap *i* as originating from a Poisson distribution with the mean and variance parameter λ_i equal to the product of trap area A_i and the sum, over all source trees $j = 1 \ldots n_i$ that are located within the maximum seed dispersal distance of the trap, of the product of b_{ij}, the estimated fecundity of each source tree *j*, a parameter γ, and $g(d_{ij})$, a probability density function for seed arrival from each source tree to an infinitely small area centered on the location of the trap $g(d_{ij})$ is understood to be a dispersal kernel (Nathan and Muller-Landau 2000), with a functional form that varies with the species for which dispersal is being modeled. Formally,

$$Y_i \sim \text{Pois}\ (\lambda_i)$$

$$\lambda_i = A_i \sum_{j=1}^{n_i} \gamma b_{ij} g(d_{ij})$$

There is a clear correspondence between the problem of estimating the number of seeds collected at a trap after dispersal from their parent trees and that of estimating the relative density of adult mosquitoes collected at a sampling site after dispersal from their larval habitats. As such, we adapted the above-formulated model to achieve the latter goal. In the model that we developed for *Cx. erraticus* dispersal, Y_i is a random variable of the total number of female *Cx. erraticus* collected over the course of the 2006 sampling season at an adult sampling site. Whereas males were occasionally collected at the adult sampling sites, we restricted our analysis to dispersal of female adults. Ponds throughout the TNF that had at least one larva collected during the first 60 dips of sampling, the number of dip samples taken at the smallest pond, were designated as suitable larval habitats. These ponds were classified as the sources of dispersing female adults, and hereafter they will be referred to as larval ponds. We estimated the distance between all adult sampling sites and all larval ponds by delineating the perimeter of each pond using the global positioning system coordinates of larval sampling points to create a polygon shapefile for each pond in ArcGIS v.9.2. We then calculated the Euclidean distances between all adult sampling sites and larval pond centroids using universal transverse mercator coordinates.

A rigorous approach for estimating larval pond fecundity, or rate of productivity of adult *Cx. erraticus*, was not available. Therefore, we developed a series of models with productivity parameterized in various manners, as follows: assumed constant across ponds (constant), proportional to pond area (area), proportional to pond perimeter (perim), proportional to the number of larvae collected in the first 60 dips (larv), proportional to pond area × the number of larvae in the first 60 dips (area × larv), or proportional to pond perimeter × the number of larvae in the first 60 dips (perim × larv).

The functional form of the dispersal kernel we used in the model was that of the exponential described in Clark et al. (1999), as follows:

$$g(d_{ij}) = \frac{1}{2\pi\beta^2} \exp\left(-\left(\frac{d_{ij}}{\beta}\right)\right).$$

This dispersal kernel models the rate of decay of the density with distance from the source as exponential. Parameterization of this part of the model effectively allows for estimation of the mean and percentiles of the dispersal distance of *Cx. erraticus.* Specifically, 2β is an estimate of mean dispersal distance (Clark et al. 1998, Cousens et al. 2008). Percentiles of dispersal distance were estimated by first converting the dispersal kernel $g(d)$ to the distance probability density function $f(d)$ (Cousens et al. 2008), as follows:

$$f(d) = \frac{d}{\beta^2} \exp\left(-\left(\frac{d}{\beta}\right)\right).$$

We then calculated the median, 95th, and 99th percentiles of $f(d)$ through integration using the rectangle method with subintervals that were 1 m in length. We used an exponential form for the dispersal kernel because it has received the most support from empirical studies of insect dispersal (Gratton and Vander Zanden 2009).

Finally, counts of *Cx. erraticus* at adult sampling sites were often zero, so that the negative binomial was a more appropriate distributional assumption for these data than the Poisson (χ^2 test of goodness-of-fit (GOF) – Poisson: $\chi^2 = 304.36$, df = 12, $P < 0.001$; χ^2 test of GOF – negative binomial: $\chi^2 = 5.90$, df = 11, $P = 0.88$). As such, we modeled the number of females collected at each adult sampling site as following a negative binomial (p,k) distribution, whereby k is a dispersion parameter and p is a function of the mean. Formally, the models we used were of the following form:

$$Y_i\text{: NB}\ (p_i,\ k)$$

$$p_i = \frac{k}{k + \mu_i}$$

$$\ln(\mu_i) = \ln(A_i) + \ln \sum_{j=1}^{m} \gamma b_j \frac{1}{2\pi\beta^2} \exp\left(-\left(\frac{d_{ij}}{\beta}\right)\right)$$

$$= \alpha + \ln \sum_{j=1}^{m} b_j \exp\left(-\left(\frac{d_{ij}}{\beta}\right)\right)$$

where

$$\alpha = \ln(A) + \ln\left(\gamma \frac{1}{2\pi\beta^2}\right),\ A_i$$

$$= A \text{ for all sampling sites } i = 1 \ldots n$$

In these models, α and β are the two parameters to be estimated, d_{ij} is the distance in meters between adult sampling site i and larval pond j, b_j is the variable serving as the proxy measure of larval pond productivity, the index i runs over all sampling sites $1 \ldots n = 26$, and the index j runs over all larval ponds $1 \ldots m = 15$.

We conducted modeling in a Bayesian framework, whereby inference about parameters is based upon examination of their posterior distributions. We used Markov Chain Monte Carlo algorithms to sample from the posterior distribution of the unknown parameters, circumventing the need for their explicit derivation (Gilks et al. 1995). We ran these algorithms using R2WinBUGS, the implementation of WinBUGS in R. We set three chains to run for 2,000 iterations with a burn-in period of 1,000 iterations. The prior distributions were uniform ($-1{,}000$, 1,000) for α, uniform(0, 5,000) for β, and gamma (0.001, 0.001) for k. Initial values for the parameters were drawn from uniform (-10, 10) and uniform (0, 500) distributions for α and β, respectively, whereas the initial value for k was set to 1, 2, or 3. Convergence diagnostics were assessed using the Coda package. We based model selection on minimization of the deviance information criterion (DIC).

After selecting a final model for the 2006 data, we validated the model by applying it to 2007 and 2008 adult sampling sites. We used the 2007 and 2008 adult sampling sites for validation because they differed in location from the ones used in model development. Because the counts of *Cx. erraticus* at the adult sampling sites were summed over a different number of visits to each sampling site in 2007 and 2008 compared to 2006, we used Spearman's rank correlation coefficient to assess model performance (Guisan and Zimmermann 2000).

Additionally, we were interested in the performance of the predicted relative density of mosquitoes from the model compared to another possible correlate that could be used to rank areas in terms of their relative densities. Specifically, we considered the number of overlapping larval buffers at a sampling site, using the mean dispersal distance derived from the model as the buffer radius-length, as this other possible correlate. We estimated the Spearman's rank correlation coefficient between the relative density of *Cx. erraticus* and this variable, and then compared both correlation coefficients using the test of two correlated correlation coefficient (Meng et al. 1992).

Finally, we were interested in the relative performance of variables representing the total number of overlapping buffers of any radius-length in predicting the ranking of areas in terms of relative density of *Cx. erraticus.* As such, we created a set of variables similar to the last one described above, with buffer radius-lengths differing between variables by increments of 100 m. We then estimated Spearman's rank correlation coefficient between each of these variables and relative density of *Cx. erraticus* in 2007 and 2008.

Results

We collected a total of 2,900 resting female *Cx. erraticus* from the adult sampling sites throughout TNF between 2006 and 2008. A total of 707 (24. 4%) of these individuals was excluded from our analyses because they were collected during sampling intervals when not all traps were visited, yielding a total of 205 individuals used in dispersal distance models developed using data from 2006 and 1988 individuals used in model validation and correlation analyses using the 2007–2008 data.

In 2006, when the adult sampling sites were distributed throughout an area radiating out 3.0 km from the core site (Fig. 1), we collected an average 7.88 (SD = 11.73, $n = 26$) females/sampling site over the seven 2-wk intervals used in the analysis (Fig. 3). The distances between adult sampling sites where resting females were collected and the nearest larval pond ranged from 0.107 to 1.946 km.

In 2007 and 2008, when the adult mosquito collections were focused within the circular area radiating out only 1.5 km from the core site (Fig. 2), we collected an average of 48.49 females/sampling site (SD = 56.08, $n = 41$) over the 15 wk used in our analyses (Fig. 3). All adult sampling sites in the study area had individuals present during at least one visit, and were distributed at distances ranging from 0.05 to 1.04 km from the nearest larval pond.

In total, *j* were 787 *Cx. erraticus* larvae was collected, with an average of 3.47 larvae/30-dip sample (SD = 6.64, $n = 227$). Of the 21 ponds we sampled, 15 were found to have *Cx. erraticus* larvae present during the first 60 dips, and were thus classified as larval ponds (Fig. 1).

Chains used in the model development all indicated a satisfactory degree of convergence after 2,000 iterations, as the Gelman-Rubin statistics for parameter values in each model were all <1.10. Average values of samples from the posterior distributions of α ranged from -8.61 to 2.16, from 483.46 to 1,682.38 for β, and from 0.44 to 0.82 for k. Overall, model deviances ranged from 136.65 to 148.59 (Table 1). DIC of the models ranged from 137.33 to 145.03, with the area model, the one that used the area of the larval pond of origin as a measure of productivity, selected as the best-fitting model based on its lower DIC compared with the other five models (Table 1).

The estimate of α based on the mean of samples from its posterior distribution in the area model was -6.37 with a 95% credible interval of (-8.08, -4.80). The dispersal parameter, β, was estimated as 483.46 with a 95% credible interval of (258.53, 1,058.95). The

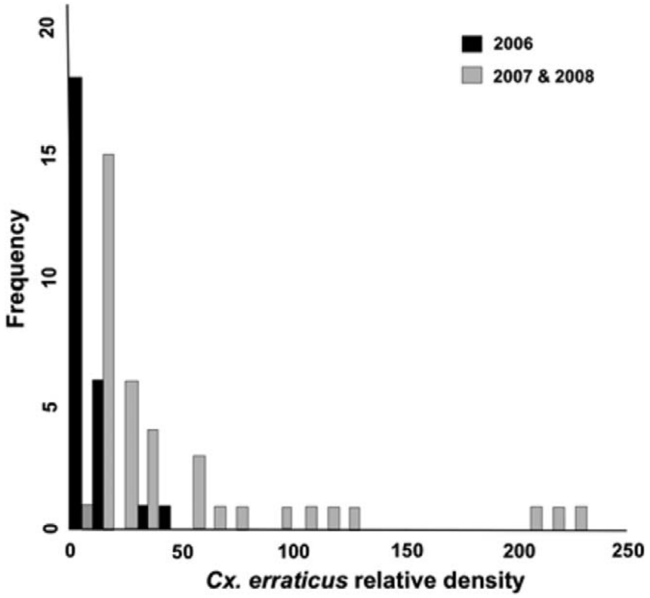

Fig. 3. Histograms of the total number of female *Cx. erraticus* collected at adult sampling sites in the TNF study area in either 2006 or 2007 and 2008 cumulatively.

mean value of the *k* for the area model was 0.82, with a 95% credible interval of (0.34, 1.70) (Table 1; Fig. 4). The estimated mean dispersal distance of *Cx. erraticus* (2β) was 0.966 km. The median, 95th, and 99th percentiles for dispersal distance were estimated as 0.811, 2.291, and 3.206 km, respectively.

Application of the model to the test validation dataset validation gave strong support to the model, as *Cx. erraticus* relative density at 2007 and 2008 adult sampling sites was significantly associated with the total height at the sampling site of the overlapping dispersal kernels centered on each larval pond and weighted by pond area ($r_S = 0.689$; 95% confident interval (CI) = 0.477, 0.82; $n = 41$; $P < 0.0001$; Fig. 5). The rank correlation between *Cx. erraticus* relative density and the number of overlapping larval buffers of radius-length equal to the mean dispersal distance was also significantly associated ($r_S = 0.597$; 95% CI = 0.347, 0.761; $P < 0.0001$; $n = 41$; Fig. 6). Overall, there was no evidence to suggest that the strengths of association between these two variables— overlapping dispersal kernel height weighted by pond area and the number of overlapping buffers—and *Cx. erraticus* relative density were significantly different from one another ($Z = 1.04$, $P > 0.10$, $n = 41$). Associations between ranks of *Cx. erraticus* relative density and the number of overlapping larval buffers at a site were positive for all other radius-lengths considered other than mean dispersal distance. However, these associations were only significant ($P < 0.05$) for buffers of radius-lengths <2,500 m. All associations were weaker than that between the rank of *Cx. erraticus* relative density and predicted relative density based on the best-fitting model parameterized by 2006 data (overlapping dispersal kernel height weighted by pond area) (Fig. 6).

Discussion

The distance that a mosquito can disperse from its site of emergence is of critical importance in studies of arboviruses. The conventional mark-release-capture approach to the study of mosquito dispersal has the potential to bias estimates of dispersal distance because animals are often captive reared and/or released in an unfamiliar area that is not necessarily suitable habitat (Silver 2008). In this study, we used a modeling

Table 1. Mean (95% credible intervals) for parameters and deviance of the models developed for *Cx. erraticus* abundance at 2006 adult sampling sites in Tuskegee National Forest, Alabama, with DIC of the models also presented

Model	α	β	k	Deviance	DIC
Constant	2.16 (0.09, 3.66)	600.81 (288.23, 2,198.90)	0.69 (0.28, 1.41)	140.56 (136.70, 150.10)	138.34
Larv	−1.45 (−3.03, 1.09)	1,682.38 (323.92, 4,640.25)	0.44 (0.21, 0.81)	148.59 (144.40, 155.30)	145.03
Area	−6.37 (−8.08, −4.80)	483.46 (258.53, 1,058.95)	0.82 (0.34, 1.70)	136.65 (133.30, 144.20)	137.33
Perim	−3.89 (−5.81, −2.42)	523.57 (278.45, 1,478.63)	0.76 (0.31, 1.54)	138.94 (135.3, 147.80)	138.86
Area × larv	−8.61 (−10.83, −6.94)	628.58 (273.42, 2,678.75)	0.62 (0.26, 1.23)	142.05 (138.20, 150.20)	138.66
Perim × larv	−6.94 (−8.86, −4.82)	1,224.30 (302.30, 4,536.65)	0.48 (0.22, 1.02)	147.20 (142.70, 154.00)	141.56

Model names correspond to the variable used as a metric for larval pond productivity in each model, as explained in *Materials and Methods.*

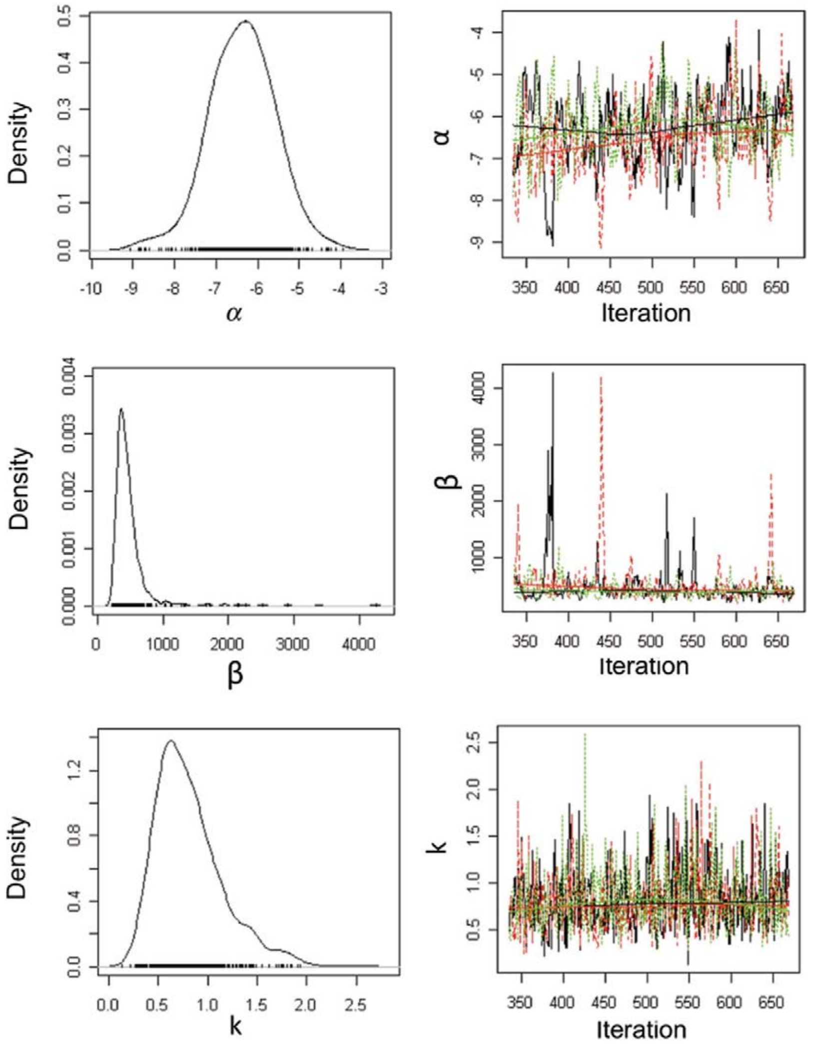

Fig. 4. Density and trace plots of Markov Chain Monte Carlo samples from the posterior densities of parameters in the best-fitting model for *Cx. erraticus* abundance at 2006 adult sampling sites, model area. Three chains were run for 2,000 iterations with a burn-in period of 1,000. A thinning interval of 3 was used, so that the number of samples shown is 334. (Online figure in color.)

approach based on sampling of wild *Cx. erraticus* to estimate the dispersal distances of mosquitoes emerging from natural wetlands during a gonotrophic cycle. Despite concerns about overestimation of dispersal distances based on mark-release-recapture of laboratory-bred mosquitoes, our estimate of the average dispersal distance of 0.967 km is close to the mean dispersal distance of 0.73 (±0.61) km for *Cx. erraticus* based on mark-recapture (Morris et al. 1991). Moreover, our results confirm that *Cx. erraticus* is a strong flier, given that most mosquito species studied typically disperse no more than a few hundred meters during appetential flight (Service 1997).

Knowledge of dispersal patterns can be used to predict the relative abundance of *Cx. erraticus* anywhere in the area of the study, as demonstrated by the strong association between the two correlates that we derived based on modeling results, total height of overlapping dispersal kernels weighted by pond area, and number of overlapping buffers with radius length equal to the mean dispersal distance. For vector control, accurate estimates of mosquito abundance have obvious utility. Knowledge of mosquito abundance across a landscape could help formulate a strategy for adulticide application, and it could also be used to predict changes in relative density of *Cx. erraticus* under various scenarios of larvicide applications to larval ponds.

Estimates of dispersal distance resulting from the current study could also potentially be used to delineate boundary zones around the TNF focus outside of which a potentially infected bridge vector—i.e., a female ovipositing in a pond that overlaps an area of enzootic transmission between *Cs. melanura* and avian

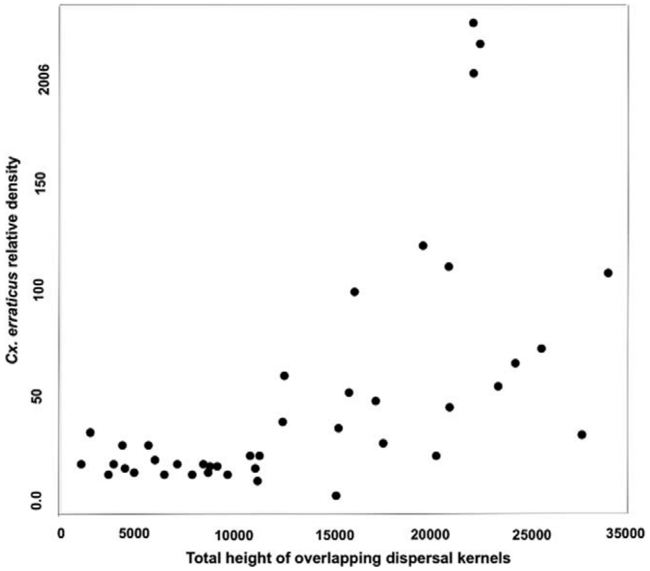

Fig. 5. Scatterplot of the total number of *Cx. erraticus* females collected at 41 adult sampling sites between 2007 and 2008 in the TNF study area versus the total height of dispersal kernels as parameterized in the area model centered on each larval pond, weighted by pond area, and overlapping at a site.

hosts—has a low probability of dispersing. For example, based on our results of the current study, the probability that a *Cx. erraticus* female will disperse further than 3.21 km from an oviposition site is 0.01. However, exposure to the bite of a bridge vector such as *Cx. erraticus* will be a function of not only the probability of dispersal to a given distance of a single vector, but also the total number of vectors dispersing such that a proposed radius ≈3 km for the TNF EEEV boundary zones could still involve an unacceptable exposure risk. Risk assessment from a human health perspective is beyond the scope of this work, but we have provided important information for the development of such assessments.

Whether or not *Cx. erraticus* proves to be an important component in EEEV transmission in the

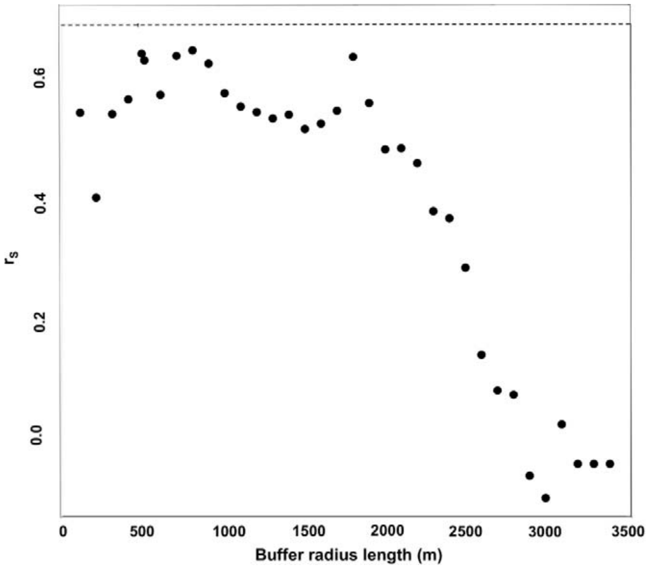

Fig. 6. Scatterplot of Spearman's rank correlation coefficient (r_S) for associations between the relative density of *Cx. erraticus* at adult sampling sites in the TNF study is in 2007 and 2008 and the total number of overlapping larval buffers at the site. The dotted line across the top of the figure indicates the r_S (0.69) for the association between the relative density of *Cx. erraticus* at an adult sampling site in 2007 and 2008, and the total height of dispersal kernels as parameterized in the area model centered on each larval pond, weighted by pond area, and overlapping at a site.

southeastern United States, in this study we have demonstrated that models originally developed to model seed dispersal are adaptable to the problem of estimating mosquito dispersal. Mark-release-recapture studies are subject to biases resulting from the use of captive bred animals, and studies of wild mosquitoes emerging from natural wetlands should be preferable for estimating the movement patterns of mosquitoes. Whereas we applied the seed dispersal modeling approach to a species of mosquito that breeds in relatively discrete areas that are easily identifiable, it could potentially be applied to mosquito species that have a more continuous distribution of breeding habitat, by representing sites of emergence as the centroids of pixels classified as suitable breeding habitats in raster images and using associated attribute data on productivity of the habitats represented by those pixels (Brown et al. 2008, Jacob et al. 2009).

The accuracy of our estimates of the mean and upper percentiles of the dispersal distance of *Cx. erraticus* is contingent upon three assumptions. First, a critical assumption is that all potential larval ponds of *Cx. erraticus* that could contribute adults to the population were identified. Whereas we feel confident in our inventory of source ponds, some of the adults that we captured could have emerged from ponds outside of our sampling area or from small aquatic habitats. Minimally, we identified all of the larval ponds near each sampling site. Thus, we are confident that our model included the primary sources of, and all of the sources that make a significant contribution to, adults for each sampling site.

The second assumption is that the measure of pond productivity used in the final model (larval pond area) is proportional to the number of adults that originate from that pond. Factors such as availability of aquatic vegetation and predator densities could affect productivity. However, as ponds in TNF are all relatively homogenous, being relatively shallow with vegetation scattered across their surface, the use of pond area as a proxy for mosquito production seems reasonable. Without more information on habitat needs of larval *Cx. erraticus* with which to assess pond quality and more data on the characteristics of each pond in the study area, there is no simple means to improve upon the use of pond area as a proxy for mosquito production.

Our final assumption stems from the reduction of the areal extent of each larval pond down to representation by its centroid alone, so the dispersal distance may have been overestimated given dispersal of adults from any point between a pond centroid and its edge. However, the average distance between each larval pond's centroid and larval sampling points along its edge was only 0.063 km, a negligible distance relative to the estimated mean dispersal distance.

In conclusion, use of models originally developed to estimate seed dispersal distances appears to be a powerful approach to characterizing the dispersal of *Cx. erraticus* given knowledge solely of the location of larval habitats and adult sampling sites. Evidence of the utility of this approach is the strong association between overlapping dispersal kernel heights predicted from the model and relative density when the model was applied to new sampling site locations. Our approach could be used in characterizing dispersal for other mosquito vector species, leading to more accurate predictions of their spatial distribution and thus the effective areas where vertebrate hosts are at risk of exposure to the pathogens they transmit.

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