Developing Models for the Forage Ratios of *Culiseta melanura* and *Culex erraticus* Using Species Characteristics for Avian Hosts

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ABSTRACT Studies of mosquito preferences for avian hosts have found that some bird species are at greater risk than others of being fed upon by mosquitoes. The ecological factors that determine this interspecific variation in avian host use by mosquitoes have been little studied, despite the possibility that such variation may influence spatial and temporal patterns of the occurrence of mosquito-borne pathogens. Our objective was to identify ecological variables associated with the avian host forage ratios estimated from a previous study of mosquito feeding patterns in Tuskegee National Forest, AL. We used species' characteristics derived from the literature to develop multiple linear regression models for the forage ratios of *Culiseta melanura* (Coquillett) and *Culex erraticus* (Dyar & Knab) for avian hosts. We found that habitat-edge association and body mass of avian host species were the best predictors of forage ratios of *Cx. erraticus* for avian hosts. Although no avian host traits were inferred to be strong predictors of forage ratios of *Cs. melanura*, body mass had the greatest importance weight among those considered. Our results suggest that characteristics of avian hosts may predict their levels of use by some mosquito species.

KEY WORDS forage ratio, avian host, *Culiseta melanura*, *Culex erraticus*

Differences in host use by mosquitoes may be influenced by intrinsic host factors (attractiveness of the host to mosquitoes or tolerance of the host to hematophagy) or extrinsic factors (host availability, geographic overlap of the host and mosquitoes, and climate) (Calisher 1994). A forage ratio for a host species is a measure of the level of use of that species corrected for availability to mosquitoes (Hess 1968); it is calculated as the ratio of the proportion of bloodmeals in a sample of blood-engorged mosquitoes that have that species as the source to the proportion of the community comprised of that species (Hess et al. 1968, Manly et al. 2002). Such availability-corrected measures of host use are necessary for the accurate estimation of elements in the next-generation matrices for vector-borne pathogens, which can be used to estimate R_0, the basic reproductive number (Hartemink et al. 2008, 2009). As such, accurate estimates of forage ratios for individual host species are important for accurate modeling of disease risk in mosquito-borne pathogen systems.

Birds are the primary reservoir hosts in many mosquito-borne pathogens (Stamm 1966, Calisher 1994, Gubler et al. 2001), so estimating mosquito forage ratios for individual bird species is particularly important for modeling some mosquito-borne diseases. However, accurate estimation of forage ratios for avian species poses logistical challenges. The rarer an avian species is or the lower its level of use by mosquitoes, the lower the probability that it will be detected in field-collected samples of blood-engorged mosquitoes. At the same time, rarer species are less likely to be detected in surveys of the avian community. As a consequence, the accuracy of estimates of forage ratios of rarer species or those with low levels of use are low, even though such species may be important to transmission dynamics if they have high or low reservoir competences (Komar et al. 1999). Currently, there are no means to predict the level of host use by mosquitoes for the bird species potentially involved in pathogen transmission except through intensive sampling over sites spread across a broad geographic area.

A potential solution to the challenge of predicting forage ratios for avian hosts is the development of a statistical model of forage ratios based on characteristics of the host species. Detailed descriptions of all North American birds, including life-history traits and habitat utilization, are published. If the probability that a mosquito will feed on a host of a particular species is associated with its species-level characteristics, then models of forage ratios based on such characteristics could be developed without the need for collection and analysis of blood-engorged mosquitoes and avian community surveys.

In the current study, we explored the development of such forage ratio models. We sought to identify

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those avian host characteristics for which data are available that are useful for predicting the forage ratios of two mosquito species that feed regularly on avian hosts in the southeastern United States and that transmit eastern equine encephalomyelitis virus (EEEV): *Culiseta melanura* (Coquillett) and *Culex erraticus* (Dyar & Knab). *Cs. melanura* is widely recognized as the primary enzootic vector of EEEV, a rare but dangerous mosquito-borne pathogen in humans for which birds are the primary reservoir hosts (Scott and Weaver 1989, Villari et al. 1995, Zacks and Paessler 2010). *Cx. erraticus* has been proposed to play a role as a bridge vector of EEEV in the southeastern United States (Cupp et al. 2003, Cohen et al. 2009). Insights gained from modeling forage ratios of these two mosquito species for avian species could potentially be used for predictive model development of EEEV transmission in this region.

Materials and Methods

Inferential Approach. We used a multi-model inferential approach (Anderson et al. 2000, Burnham and Anderson 2002) to identify host characteristics that could be used to predict forage ratios of *Cs. melanura* and *Cx. erraticus.* We first developed a set of candidate general linear models to predict the forage ratios of avian host species using characteristics of those species. We then examined model-averaged estimates of the coefficients of the predictor variables and 95% confidence intervals around those estimates to determine whether a host characteristic would be useful for predicting forage ratios. We used unconditional weighted standard errors in calculating confidence intervals of coefficient estimates. Predictor variables with confidence intervals that excluded null were inferred to be useful for prediction. The strengths of evidence for associations between predictor variables and forage ratios were also considered through examination of importance weights of each predictor variable and whether or not predictor variables were included in models that had substantial support, that is, $\Delta\text{AICc} \leq 2$ (Burnham and Anderson 2002).

Data Sources. We calculated forage ratios for avian host species identified in bloodmeals collected between 2001 and 2009 in a 28 km^2 study area in Tuskegee National Forest, AL (Estep et al. 2011). A forage ratio for an avian species was calculated as the ratio of the proportion of bloodmeals from that species to the relative abundance of that species in the avian community (Hess 1968, Manly et al. 2002). Forage ratios of avian species were calculated separately for *Cs. melanura* and *Cx. erraticus*, using bloodmeals derived from mosquitoes collected across all study years during sampling from March to October at 74 sampling sites located at regular intervals along transects radiating from the center of the study area (Estep et al. 2010). Mosquitoes were collected from natural resting sites, as well as artificial resting sites including fiber pots, resting boxes, and 50-gallon plastic cans located at sampling sites, with sampling device used dependent on year of sampling (Hassan et al. 2003, Estep et al. 2010). The relative abundances of host species in the avian community were calculated separately for *Cs. melanura* and *Cx. erraticus* using average estimated densities of avian hosts at all sites where individuals of either mosquito species were collected, respectively. These densities were estimated by applying models of density for each avian species to these sites (Estep et al. 2011, McClure et al. 2011). The number of sites where *Cx. erraticus* was present, and thus where avian densities were estimated for forage ratio calculations for this mosquito, was 56; avian densities were estimated at 14 sites to estimate the relative abundance of species in the avian community available to *Cs. melanura* (Estep et al. 2011). Bloodmeals derived from those vertebrate species for which density models were not available, for example, mammals and some avian species, were excluded from the sample of bloodmeals used in forage ratio calculations (Estep et al. 2011).

A large proportion of bird species that were recorded during censuses were not detected in any bloodmeal samples. These species have calculated forage ratios of zero. Inclusion of such zero-valued forage ratios would have caused significant violations of linear regression modeling. Therefore, we restricted our analysis to those avian species with forage ratios >0. In total, we used forage ratios of 15 avian species for development of the *Cs. melanura* model, and 22 avian species for development of the *Cx. erraticus* model (Estep et al. 2011, Appendix 1).

We identified seven host characteristics that could influence its level of host use by mosquitoes (Table 1). We chose these predictor variables partly due their potential association with host attractiveness, defensive behavior, or probability of encounter with mosquitoes, and partly due the availability of information for estimating species values in the ornithological literature, which included species accounts, published reviews, and online datasets. Full descriptions of each predictor variable and its predicted direction of association with the forage ratio variable are provided in Table 1. Estimated values of habitat-edge association of the barred owl (*Strix varia*) were not available in the primary literature, so we scored this species with a value of one (interior species) based on descriptions in Ehrlich et al. (1988). We also imputed the number of broods per season for brown-headed cowbird (*Molothrus ater*), a brood parasite with a wide range of hosts, using the average number of broods across all avian species detected during point count surveys in TNF. The beginning and end of the host-seeking seasons of *Cs. melanura* and *Cx. erraticus,* as detailed in Table 1, were estimated as the quarter-months during which the 2.5 and 97.5 percentiles of the dates of capture of engorged individuals of each species fell. Thus, the beginning and end of the host-seeking seasons were taken as first quarter of May and the second quarter of August for *Cs. melanura,* and the first quarter of April and the third quarter of September for *Cx. erraticus*, respectively.

Statistical Modeling. We used an all-subsets approach, such that each possible predictor variable combination was in a model in the candidate set for

Table 1. Predictor variables used in candidate forage ratio models of *Cs. melanura* and *Cx. erraticus*

Variable name	Description	Predicted direction: *Cs. melanura* model	Predicted direction: *Cx. erraticus* model	Data source
Availability	Availability status: indicator variable (0, present for only part of mosquito host-seeking season; 1, present for entire mosquito host-seeking season); assumed present from start of breeding to start of migration (rounded to quarter-month); see text for mosquito host-seeking season dates	Pos	Pos	Poole et al. 2005 (individual species accounts)
Cavity	Cavity nesting status: indicator variable (0, uses nest type other than cavity nest; 1, cavity-nesting)	Pos	Pos	Erhlich et al. 1988
Edge	Habitat-edge association: avg score from 3 sources, where: 1, forest interior species; 2, generalist species; 3, edge species	Neg	Pos	Whitcomb 1981, McIntyre 1995, Bender et al. 1998
Mass	Body mass (g)	Pos	Pos	Dunning 2008
Nestling-days	Nestling availability: expected no. of days breeding pair is with nestlings = nestling stage length*avg no. of broods (days)	Pos	Pos	Erhlich et al. 1988, Gough et al. 1998
Roost	Communal roosting status: indicator variable (0, does not roost communally on breeding grounds; 1, roosts regularly during breeding season or after breeding season, but before fall migration)		Pos	Poole 2005 (individual species accounts)
Ht	Nest ht: avg nest ht (m), midpoint of range of nest ht if avg not provided	Not predicted	Not predicted	Poole 2005 (individual species accounts)

each mosquito species. The communal roosting variable was not included in any models in the *Cs. melanura* candidate set because no species for which forage ratio estimates for *Cs. melanura* were available roost communally during the host-seeking season of this mosquito. We conducted analyses in the R software environment, specifying models with the glm function in the stats package and conducting multi-model inference calculations with functions in the MuMIn package (R Core Development Team 2008).

We used the Box-Cox method on the regression models for each mosquito species that included all predictor variables to determine the appropriate transformation of the response variable (forage ratio) necessary for alleviating potential heteroscedasticity (Kutner et al. 2005). As a result, we applied a *ln*-transformation to the forage ratio variable. We also examined residual plots of the *ln*-transformed forage ratios regressed on individual continuous predictor variables to determine whether transformations of predictor variables were necessary. As a result, we used a *ln*-transformation of the body mass variable and an inverse transformation of the nestling-days variable.

We compared the bias-corrected Akaike Information Criterion (AICc; Hurvich and Tsai 1989) between all models in the candidate set for each species and used this criterion in calculating importance weights of models and coefficient estimates, their unconditional confidence intervals, and importance weights for predictor variables in the forage ratio models (Burnham and Anderson 2002). All predictor variables had an equal probability of inclusion in models in the set used for calculating importance weights. We used the Breusch-Pagan test for heteroscedasticity and examined Variance Inflation Factors, DFFITS, Cook's distances, DFBETAS, and residual plots to check for potential modeling assumption violations in the model that included all predictor variables inferred useful for prediction (Kutner et al. 2005).

Results

***Cx. erraticus* Model.** The top-ranked model in the *Cx. erraticus* candidate set included habitat-edge association and body mass as predictor variables and had an importance weight of 0.26. One other *Cx. erraticus* forage ratio model received substantial support; it included body mass, habitat-edge association, and cavity nesting status as predictors and had an importance weight of 0.16. The other candidate predictors considered in model development-seasonal availability, communal roosting status, nest height, and nestling availability were not included in any models with substantial support. All models in the *Cx. erraticus* candidate set that received moderate to low support had importance weights ≤0.09 (Table 2).

Interpretation of model-averaged coefficient estimates and associated 95% unconditional confidence intervals suggested that habitat-edge association and body mass could both be used to develop predictive models of the *Cx. erraticus* forage ratio (Table 3). *Cx. erraticus* forage ratios increased with habitat-edge association ($\beta_{\text{edge}} = 1.35$, LCL = 0.44, UCL = 2.26; Fig. 1, Table 3), and habitat-edge association had the highest importance weight (0.98; Table 3) among all predictor variables considered.

Table 2. AIC table for *Cs. melanura* and *Cx. erraticus* forage ratio models that received moderate support (ΔAICc ≤ 7)

Model	Log(*L*)	AICc	K	Δ_i	w_i
Cs. melanura					
Mass	−18.97	46.11	3	0.00	0.22
(intercept only)	−20.70	46.39	2	0.28	0.20
Mass + edge	−18.55	49.09	4	2.98	0.05
Mass + cavity	−18.59	49.18	4	3.07	0.05
Availability	−20.55	49.28	3	3.16	0.05
Mass + ht	−18.65	49.29	4	3.18	0.05
Edge	−20.59	49.36	3	3.25	0.04
Cavity	−20.61	49.41	3	3.29	0.04
Ht	−20.64	49.46	3	3.34	0.04
Mass + nestling-days	−18.77	49.54	4	3.43	0.04
Nestling-days	−20.70	49.57	3	3.46	0.04
Mass + availability	−18.90	49.81	4	3.69	0.04
Mass + edge + nestling-days	−17.79	52.24	5	6.13	0.01
Ht + cavity	−20.27	52.55	4	6.43	0.01
Edge + cavity	−20.41	52.83	4	6.71	0.01
Nestling-days + availability	−20.42	52.84	4	6.73	0.01
Mass + nestling-days + availability	−18.15	52.96	5	6.85	0.01
Ht + availability	−20.49	52.98	4	6.86	0.01
Cavity + availability	−20.51	53.02	4	6.91	0.01
Edge + availability	−20.54	53.07	4	6.96	0.01
Cx. erraticus					
Mass + edge	−34.27	78.89	4	0.00	0.26
Mass + edge + cavity	−33.06	79.87	5	0.98	0.16
Mass + edge + cavity + nestling-days	−31.71	81.03	6	2.14	0.09
Mass + edge + nestling-days	−33.86	81.47	5	2.59	0.07
Mass + edge + ht	−34.23	82.21	5	3.33	0.05
Mass + edge + availability	−34.26	82.27	5	3.39	0.05
Mass + edge + roost	−34.26	82.28	5	3.39	0.05
Mass + edge + cavity + availability	−32.65	82.90	6	4.02	0.04
Mass + ht + edge + cavity	−33.04	83.68	6	4.79	0.02
Mass + edge + cavity + roost	−33.05	83.70	6	4.81	0.02
Mass + ht + edge + nestling-days	33.47	84.54	6	5.66	0.02
Mass + ht + edge + cavity + nestling-days	−31.49	84.97	7	6.09	0.01
Mass + edge + cavity + nestling-days + availability	−31.58	85.15	7	6.27	0.01
Mass + edge + nestling-days + availability	−33.83	85.26	6	6.37	0.01
Mass + edge + nestling-days + roost	−33.83	85.27	6	6.38	0.01
Mass + edge + nestling-days + cavity + roost	−31.71	85.42	7	6.54	0.01

AICc, bias-corrected Akaike Information Criterion; K, no. of parameters estimated; Δ_i, difference in AICc from the model that minimized the AICc; w_i, model importance weight.

The direction of association between *Cx. erraticus* forage ratios and body mass was also positive (β_{mass} = 0.52, LCL = 0.07, UCL = 0.97; Fig. 2, Table 3), and the body mass variable had an importance weight of 0.94. Cavity nesting status ranked third in terms of importance weights with importance weight = 0.41, but this variable was not interpreted as being useful for prediction because of its 95%

Table 3. Results of model averaging for predictor variables in *Cs. melanura* and *Cx. erraticus* forage ratio models and importance weights of all variables considered in analyses

Model	Variable	Importance weight	Model-averaged estimate	Weighted unconditional SE	95% confidence interval	
					Lower	Upper
Cs. melanura	Mass	0.51	0.22	0.28	−0.34	0.78
Cs. melanura	Edge	0.16	0.04	0.23	−0.41	0.50
Cs. melanura	Cavity	0.15	−0.02	0.44	−0.89	0.86
Cs. melanura	Availability	0.14	0.04	0.28	−0.51	0.59
Cs. melanura	Nestling-days	0.14	0.63	5.38	−9.91	11.17
Cs. melanura	Ht	0.14	0.00	0.03	−0.07	0.06
Cs. melanura	Intercept		−0.23	1.16	−2.50	2.05
Cx. erraticus	Edge	0.98	1.35	0.47	0.44	2.26
Cx. erraticus	Mass	0.94	0.52	0.23	0.07	0.97
Cx. erraticus	Cavity	0.41	0.55	0.89	−1.19	2.29
Cx. erraticus	Nestling-days	0.27	3.13	7.83	−12.22	18.48
Cx. erraticus	Availabilty	0.15	−0.02	0.32	−0.64	0.60
Cx. erraticus	Roost	0.15	0.04	0.47	−0.88	0.97
Cx. erraticus	Ht	0.14	0.00	0.03	−0.06	0.06
Cx. erraticus	Intercept		−4.03	1.44	−6.85	−1.20

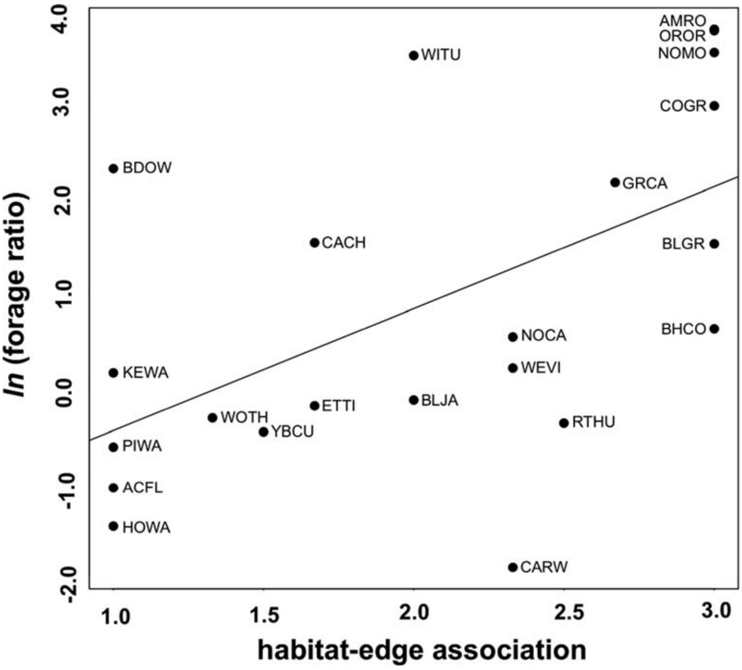

Fig. 1. Scatterplot of *Cx. erraticus* forage ratios for avian host species (*ln*-transformed) versus avian species' habitat-edge associations, with best-fit line from simple linear regression overlaid ($n = 22$). Avian species codes follow *Appendix* 1. The model-averaged estimate for the slope of *ln* (forage ratio) regressed on habitat-edge association was 1.35, with an unconditional 95% confidence interval of (0.44, 2.26). The coefficient of determination for the simple linear regression model of *ln* (forage ratio) versus habitat-edge association was 0.32.

confidence interval, which included null ($\beta_{\text{cavity}} = 0.55$, LCL = −1.19, UCL = 2.29). The fourth-ranked importance weight was assigned to nestling availability, but this variable also was not interpreted as being useful for prediction: ($\beta_{\text{nestling-days}} = 3.13$, LCL = −12.22, UCL = 18.48) All predictor variables considered in modeling *Cx. erraticus* forage ratios other than habitat-edge association, body mass, and nestling availability had relatively low importance weights (≤0.15; Table 3).

Diagnostic tests for the model of forage ratios of *Cx. erraticus* that included body mass and habitat-edge association as predictor variables suggested there were no assumption violations. Residual plots revealed no discernible residual patterns in this model. The maximum absolute value of the DFFITS among all data points was 0.72 and thus, less than the cutoff criterion of 1. The maximum absolute value of the DFBETAS was 0.18 for habitat-edge association and 0.08 for body mass, both of which are below the cutoff criterion of $2/\sqrt{n} = 0.43$. The maximum Cook's distance for observations was 0.15, which is less than the cutoff criterion of the 50th percentile of the F(p,n-p) distribution = 0.81. VIF for both variables were 1.00 and thus, below the cutoff criterion of 2. The null hypothesis of homoscedasticity of error variance was not rejected using the Breusch-Pagan test (BP = 1.99, df = 2, $P = 0.37$).

***Cs. melanura* Model.** The top-ranked *Cs. melanura* forage ratio model included body mass as the sole predictor variable and had an importance weight = 0.22. The model for the mean (no predictor variables) was the only other model that received substantial support, with a ΔAICc value = 0.28; it had an importance weight = 0.20. All other models in the *Cs. melanura* candidate set had ΔAICc values >2 and relatively low importance weights (≤0.05; Table 2).

Unconditional 95% confidence intervals for all coefficient estimates for predictor variables in the *Cs. melanura* candidate model included null. The predictor variable receiving the highest support in the *Cs. melanura* models was body mass (importance weight = 0.51). The direction of association of body mass with *Cs. melanura* forge ratios was positive, as it was in the *Cx. erraticus* forage ratio model. All other predictor variables in the *Cs. melanura* forage ratio models had relatively low importance weights (≤0.16; Table 3).

Discussion

Our results offer the first evidence that the level of use of an avian species by mosquitoes as a bloodmeal source can be predicted from its species-level characteristics. Associations between mosquito levels of use of host species and host characteristics are to be expected because the ecology of the host determines whether a mosquito successfully locates a host and acquires a bloodmeal from it (Calisher 1994). The implications of demonstrating that specific host characteristics predict hemotophagy are significant because elucidation of the patterns that control host-

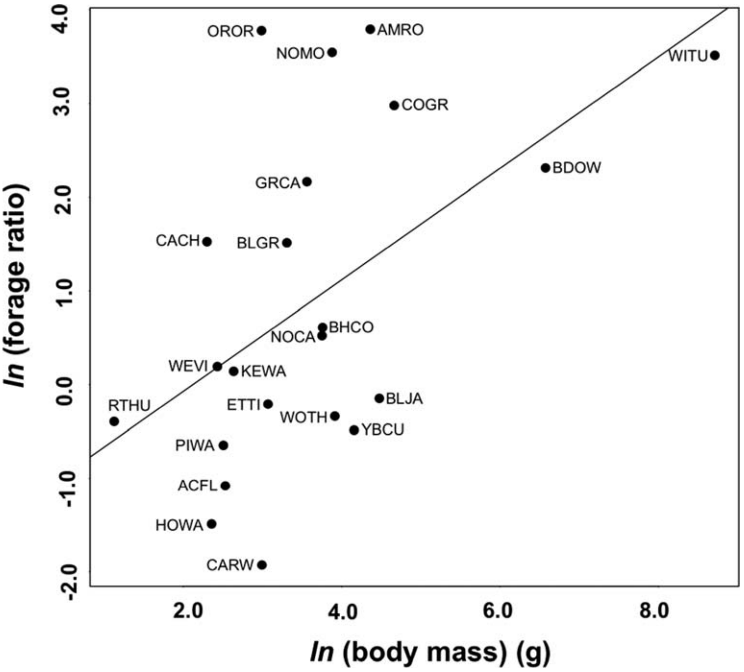

Fig. 2. Scatterplot of *Cx. erraticus* forage ratios for avian host species (*ln*-transformed) versus *ln* (body mass), with best-fit line from simple linear regression overlaid ($n = 22$). Avian species codes follow *Appendix* 1. The model-averaged estimate for the slope of *ln* (forage ratio) regressed on *ln* (body mass) was 0.52, with an unconditional 95% confidence interval of (0.07, 0.97). The coefficient of determination for the simple linear regression model of *ln* (forage ratio) versus *ln* (body mass) was 0.28.

vector interactions should allow for more accurate predictions of disease risk in multi-host systems (Hartemink et al. 2009).

Habitat is an obvious environmental factor for linking vectors and hosts, so it was not surprising that habitat edge association was the highest-ranked predictor variable in the forage ratio models of *Cx. erraticus.* What was unexpected was that the habitat associations of birds did not appear to be useful for predicting *Cs. melanura* forage ratios. We had predicted that habitat-edge association would be positively associated with forage ratio in the *Cx. erraticus* model and negatively associated with forage ratio in the *Cs. melanura* model. The basis for these predictions was a previous study of mosquito microhabitat associations (Bidlingmayer 1971), which showed different strengths of habitat-edge association in wooded swamp habitats between *Cs. melanura* and species in the *Culex* (*Melanoconion*) subgenus, of which *Cx. erraticus* is a member (Darsie and Ward 2005). Specifically, the average number of *Cs. melanura* captured per trap night was higher in traps located in swamps compared with traps located at swamp edges, field edges, or in fields. Based on these observations, Bidlingmayer (1971) suggested that *Cs. melanura* is a swamp interior species that avoids swamp edges and the higher levels of illumination associated with them. Conversely, the highest average trap counts of *Culex* (*Melanoconion*) species were at field edges, followed by traps in fields, at swamp edges, or in swamps (Bidlingmayer 1971).

We predicted that if microhabitat associations of *Cs. melanura* and *Cx. erraticus* in TNF were similar to those described in Bidlingmayer (1971) then swamp interiors would have had the highest densities of *Cs. melanura.* If *Cs. melanura* concentrated their activities in swamp interiors the result would be inflated forage ratios for swamp interior bird species because of greater availability to *Cs. melanura* than assumed (Estep et al. 2011). Conversely, *Cx. erraticus* were expected to encounter swamp- and field-edge avian species at a higher rate than assumed, such that the forage ratio of habitat-edge associated avian species would be inflated.

Although the predicted positive association between habitat-edge association and forage ratio was observed for avian species in the *Cx. erraticus* model, the predicted negative relationship was not observed for avian species in the *Cs. melanura* model. One explanation for why we did not find habitat-edge association to be a useful predictor of *Cs. melanura* forage ratios is that any negative effect of this variable was too small for us to detect, given the limited sample size of the *Cs. melanura* dataset. Alternatively, our classification of edge habitat may have been different than that of Bidlingmayer (1971). In Bidlingmayer's study, edge was defined as an ecotone—the transition between very different habitat types such as pasture and woodland. In contrast, species that are classified as edge species may recognize edge habitat at a much finer scale, such as a small opening in a forest. As a result, *Cs. melanura* may avoid field-swamp edges but nonetheless use marsh-swamp edges in host-seeking, such that there would be no net biological influence on *Cs. melanura* forage ratio, or potentially a slight positive one, as our results indicate.

Our predictions and interpretation of results rests on the assumption that the habitat-edge association variable used in this study is positively associated with the likelihood that a species occurs at a habitat edge and, conversely, that it is negatively associated the likelihood that a species occurs in the interior of habitat patches. We expect this assumption to hold for species with strong affiliation for either edge or habitat interiors. The assumption is more problematic for generalist species, but we can still make predictions. The "jack-of-all-trades-master-of-none" hypothesis posits that generalists will be poor competitors relative to specialists in specific habitats (Kawecki 1994, Whitlock 1996, Julliard et al. 2006, Verberk et al. 2010), such that generalists will be numerically subordinant to specialists within those habitats (Sala et al. 2006). As such, we assumed that generalist species were less likely than edge specialists to occur in edge habitats, and that generalist also had a lower occupancy rates of patch interiors than interior specialists.

Evidence of inflated forage ratios for edge-associated species underscores the importance of accurate estimation of availability of vertebrate hosts used in forage ratio calculations. For example, when a uniform distribution of hosts is assumed in calculating host availabilities over areas where both hosts and mosquitoes are nonuniformly distributed because of habitat associations, resulting forage ratios may be biased, such that they do not accurately estimated probability of use given equal availability of hosts in each species class (Manly et al. 2002). Nonetheless, and perhaps more importantly, such biased estimates of forage ratios would perhaps be more useful for predicting avian host use than unbiased estimates if they are used to predict use over areas where the edge-habitat associations of mosquitoes and hosts will influence their rates of encounter.

The least surprising results of our analysis were the positive associations between forage ratio and avian host body mass in both the *Cx. erraticus* and *Cs. melanura* models, and the inference that avian body mass is a useful predictor of *Cx. erraticus* forage ratios for avian hosts. Birds with larger body masses have higher rates of oxygen consumption (Grubb 1983), and greater rates of carbon dioxide output. Carbon dioxide is one of the primary biochemical attractants to vertebrate hosts identified for mosquitoes (reviewed in Nicolas and Sillans 1989), so larger birds would be expected to attract questing mosquitoes at a higher rate compared with smaller birds. In a recent study, the body mass of birds in baited mosquito traps was positively associated with the number of mosquitoes captured at the trap (Suom et al. 2010), suggesting that indeed carbon-dioxide output of individual host species could influence their contact rate with mosquitoes. However, birds with greater body masses also occupy a greater volume of space, such that mosquitoes are more likely to encounter larger birds. Larger birds may also have higher tolerances to hematophagy or might attract more mosquitoes than smaller birds through greater heat production (Brown 1966, Edman and Scott 1987). Any one of these factors, or the combined effect of them, offers a clear biological explanation for the observed association between forage ratios and body mass observed in the current study.

Other than habitat-edge association and body mass, predictor variables considered in model development of *Cx. erraticus* forage ratios did not receive much support. For *Cs. melanura* forage ratios, no variables other than body mass received significant support in the models. In the *Cx. erraticus* forage ratio model, even though cavity-nesting status was not inferred to be useful for prediction, it may be worth some consideration. The estimated coefficient of cavity-nesting status was in the direction we predicted from results of a previous study that showed unexpectedly high seroprevalences of EEEV antibodies in cavity-nesting species (Crans et al. 1994). Given the data showing that cavity-nesting species tend to have a higher level of use by *Cx. erraticus* than species that do not nest in cavities, this variable may be useful for prediction, but because of limited samples sizes the effect was too weak to be detected in our study.

The total number of days that nestlings are available, calculated as product of the average number of broods and average nestling stage length, ranked fourth in importance weight in the forage ratio model for *Cx. erraticus.* The coefficient of this variable was positive, and thus, in the opposite direction based on the hypothesis that nestlings may be particularly vulnerable to hematophagy by mosquitoes (Blackmore and Dow 1958, Kale et al. 1952, Griffing et al. 2007, Burkett-Cadena et al. 2010) and that avian species that score high for this variable have nestlings available more frequently than species that score low. While we predicted a positive association and nestling availability, our application of an inverse transformation applied to this variable resulted in an quantitative prediction of a negative coefficient of this variable in model results.

Although cavity nesting status and nestling availability both had moderate importance weights in the forage ratio models of *Cx. erraticus*, they all had uniformly low importance weights in forage ratio models of *Cs. melanura.* Moreover, two variables (seasonal availability and nest height) had uniformly low importance weights in forage ratio models for both species of mosquito. The low importance weight of seasonal availability suggests that there was likely little bias introduced in the calculation of forage ratios using bloodmeals collected between March and September of each year, rather than those collected strictly between the dates when the avian community may be most stable (1 May to 15 August) (Estep et al. 2011).

Mosquitoes often exhibit vertical niche partitioning within habitats (Snow 1955). As such, we expect mosquitoes to encounter the avian host species overlapping their individual vertical niches more often than those that nest in strata outside of their niche. The foraging height of *Cx. erraticus* was studied in South Carolina by comparing the fractions of *Cx. erraticus* in mosquito samples from traps suspended at heights of 1.5, 5, and 10 m. The fractions of *Cx. erraticus* in the samples at these different heights were 0.72, 0.20, and 0.07, respectively (Swanson and Adler 2010). Studies of the vertical distribution of *Cs. melanura* demonstrated a high degree of inter-site variability, with either no association between abundance and trap height detected, or the direction of the detected association dependent upon habitat type (reviewed in Nasci and Edman 1981). Although such variability in vertical distribution among habitats precluded formation of specific predictions about the direction of association between average nest height and forage ratios of avian host species, we did indeed expect to detect that it would be useful for predicting forage ratios. However, our results suggest that nest height has a relatively weak influence on the contact rate between mosquitoes and avian hosts compared with other variables considered.

Communal roosting status received a low importance weight in the forage ratio models for *Cx. erraticus,* the only set of models in which it was included. The inferred lack of an effect of communal roosting status on forage ratios of mosquitoes considered in this study is counter to our prediction that the patchy distribution of communal roosts (Diuk-Wasser et al. 2010) makes roosting birds more easy to locate (Reynolds et al. 2009), such that their availability for forage ratio calculations was underestimated using simple relative abundances derived from avian density modeling.

Our objective in this study was to identify variables that can be used to predict the levels of mosquito use of avian host species. To this end, we developed models of forage ratios calculated from a field study of mosquito feeding patterns of avian hosts (Estep et al. 2011). Because these forage ratios were calculated using only those bird species that were detected during avian surveys—excluding nocturnal species, most wading birds, and all nonavian hosts—an important caveat of our study is that inference derived from results our analysis may not extend to those species not included in forage ratio calculations (McClure et al. 2011). A direction for future research is to examine the extent to which habitat-edge association and body mass serve as useful predictors of level of use by mosquitoes for avian species that were not included in our study, or more generally, other vertebrate hosts.

Although habitat-edge association and body mass appear to be useful for predicting forage ratios for *Cx. erraticus,* identification of variables that could be used in a predictive model for *Cs. melanura,* the putative primary enzootic vector of EEEV, were less successful. Body mass was the only predictor in the *Cs. melanura* model that received strong support, but this variable was not inferred to be useful for prediction. It is possible that body mass and all host trait variables that were deemed not useful for prediction may indeed influence level of use of a species by mosquitoes but that their effects were too small for use to detect in this study given limited sample sizes for both candidate model sets. Alternatively, biological variables that influence avian host use may not be related to the avian host traits considered in our analyses, but instead biochemical cues (e.g., pheromones and metabolic byproducts) or defensive behaviors that vary between host species and more difficult to estimate. Given the important role of *Cs. melanura* as an enzootic vector of EEEV in the northeast and potentially through the range of the virus in North America, there remains a clear need for more estimates *Cs. melanura* forage ratios for avian hosts.

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Appendix 1. Forage ratios and host-trait data used in model development

Species	Code	Forage ratio[a] (*Cs. melanura*)	Forage ratio[a] (*Cx. erraticus*)	Availability (*Cs. melanura*)	Availability (*Cx. erraticus*)	Cavity	Edge	Mass	Nestling-days	Roost	Height
Acadian flycatcher *Empidonax virescens*	ACFL	1.11 (0.77); 2	0.34 (0.34); 1	0	0	0	1.00	12.6	28.0	0	5.88
American robin *Turdus migratorius*	AMRO		44.27 (11.1); 14	1	1	0	3.00	78.5	37.5	1	3.60
Barred owl *Strix varia*	BDOW	8.12 (4.53); 3	10.08 (3.69); 7	1	1	1	1.00	716.5	42.0	0	9.77
Blue grosbeak *Passerina cerulea*	BLGR		4.55 (3.19); 2	1	0	0	3.00	27.4	18.0	0	3.98
Blue jay *Cyanocitta cristata*	BLJA		0.86 (0.49); 3	1	1	0	2.00	88.0	57.0	0	15.50
Blue-gray gnatcatcher *Polioptila caerulea*	BGGN	1.03 (0.72); 2		1	1	0	2.00	5.8	22.0	0	8.50
Brown-headed cowbird *Molothrus ater*	BHCO		1.84 (1.29); 2	1	1	0	3.00	43.0	19.2	1	1.50
Carolina chickadee *Poecile carolinensis*	CACH		4.6 (1.07); 16	1	1	1	1.67	10.0	15.0	0	2.80
Carolina wren *Thryothorus ludovicianus*	CARW	0.35 (0.35); 1	0.15 (0.15); 1	1	1	0	2.33	20.0	32.5	0	1.80
Common grackle *Quiscalus quiscula*	COGR		19.65 (9.66); 4	1	1	0	3.00	106.1	36.0	1	2.51
Common yellowthroat *Geothlypis trichas*	COYE	7.73 (4.32); 3		1	1	0	2.33	9.5	20.0	0	0.00
Eastern towhee *Pipilo erythrophthalmus*	EATO	3.28 (3.25); 1		1	1	0	2.67	40.1	27.5	0	0.00
Gray catbird *Dumetella carolinensis*	GRCA	7.06 (6.98); 1	8.69 (4.95); 3	1	1	0	2.67	35.3	21.0	0	1.50
Hooded warbler *Wilsonia citrina*	HOWA	1.12 (0.77); 2	0.23 (0.22); 1	0	0	0	1.00	10.6	8.5	0	0.64
Kentucky warbler *Oporornis formosus*	KEWA		1.15 (1.15); 1	1	0	0	1.00	14.0	9.0	0	0.00
Louisiana waterthrush *Parkesia motacilla*	LOWA	2.82 (2.79); 1		0	0	0	1.00	19.9	10.0	0	0.82
Northern cardinal *Cardinalis cardinalis*	NOCA	4.18 (0.70); 20	1.69 (0.32); 22	1	1	0	2.33	42.7	18.8	0	2.05
Northern mockingbird *Mimus polyglottis*	NOMO		34.59 (9.05); 13	1	1	0	3.00	48.5	30.0	0	2.00
Orchard oriole *Icterus spurius*	OROR		43.63 (21.44); 4	0	0	0	3.00	19.9	12.5	0	8.95
Pine warbler *Dendroica pinus*	PIWA		0.52 (0.52); 1	1	1	0	1.00	12.3	15.0	0	12.69
Red-eyed vireo *Vireo olivaceus*	REVI	1.04 (0.58); 3		1	0	0	1.67	16.8	22.0	0	3.40
Ruby-throated hummingbird *Archilochus colubris*	RTHU		0.67 (0.67); 1	1	0	0	2.50	3.1	52.5	0	6.00
Tufted titmouse *Baeolophus bicolor*	ETTI	0.67 (0.46); 2	0.81 (0.32); 6	1	1	1	1.67	21.6	33.0	0	11.50
White-eyed vireo *Vireo griseus*	WEVI	0.61 (0.60); 1	1.21 (0.53); 5	1	0	0	2.33	11.4	20.0	0	0.85
Wild turkey *Meleagris gallapavo*	WITU		33.49 (14.65); 4	1	1	0	2.00	6050.0	8.0	1	0.00
Wood thrush *Hylocichla mustelina*	WOTH		0.71 (0.71); 1	1	0	0	1.33	50.2	18.0	0	2.30
Yellow-billed cuckoo *Coccyzus americanus*	YBCU	0.94 (0.52); 3	0.62 (0.3); 4	0	0	0	1.50	64.0	15.0	0	1.83
Yellow-throated vireo *Vireo flavifrons*	YTVI	2.24 (2.22); 1		0	0	0	2.00	18.0	14.0	0	10.50

[a] Forage ratio estimate (SE of estimate); no. mosquitoes collected during TNF surveys engorged with blood from this avian species (Estep et al. 2011).