Am. J. Trop. Med. Hyg., 74(1), 2006, pp. 174–179

CHANGES IN SEROPREVALENCE OF WEST NILE VIRUS ACROSS ILLINOIS IN FREE-RANGING BIRDS FROM 2001 THROUGH 2004

TARA A. BEVEROTH, MICHAEL P. WARD, RICHARD L. LAMPMAN, ADAM M. RINGIA, AND ROBERT J. NOVAK*
Medical Entomology Laboratory, Illinois Natural History Survey, University of Illinois, Champaign, Illinois; U.S. Fish and Wildlife Service, Bronx, New York

Abstract. Of the 5,236 birds sampled for antibodies to West Nile virus (WNV) in Illinois from 2001 through 2004, 348 (6.6%) birds were seropositive. Our multiple year surveillance identified several avian species that had particularly high percentages of seropositive individuals. The importance of these species in the enzootic and/or epizootic transmission of WNV is discussed relative to their regional abundance and literature on host competency. The species with the highest exposure rates to WNV differed both temporally and regionally. In general, birds that bred or were born in Illinois were more likely to have antibodies than transient birds. There was also a significant difference in the seroprevalence between adults (12.1%) and juveniles (5.5%), indicating that the acquired antibody response from previous years is a critical concern when interpreting seroprevalence rates in wild-caught birds. The most common hosts for St. Louis encephalitis virus were also the most common hosts for WNV, which strongly supports the role of similar vectors for both flaviviruses. Avian species with high WNV seroprevalence rates tended to be those that bred throughout the year, have open cup nests, and live in close proximity to humans.

INTRODUCTION

West Nile Virus (WNV) is a mosquito-borne flavivirus (family Flaviviridae) that was first isolated in Africa in 1937.[1] It was first reported in North America in New York City in 1999 and in just five years spread throughout most of North and Central America.[2] In Illinois, WNV was first reported in September 2001 with an initial distribution limited to seven counties, primarily in the northeastern section of the state.[3] Since then, WNV has been detected in all but 2 of the 102 counties in Illinois.[3] In 2002, Illinois had the highest number of WNV human cases (884) and deaths (66) in the United States. Although WNV poses a serious long-term health threat, little is known about the impact WNV has on birds, the primary reservoir hosts, and the role that specific bird species have on the distribution and amplification of WNV transmission.[4] Since seropositive/seroprevalence rates are a measure of exposure and survival, they can be interpreted in conjunction with ancillary information. Using seropositive/seroprevalence rates, we present the findings of a four-year study that documents the spatial and temporal changes in avian seroprevalence within Illinois.

Although almost 300 bird species have been reported as WNV positive, a much smaller number are likely to be reservoir hosts.[5] The significance of a species or group of species in WNV transmission is related to their host competency, their natural exposure rates to mosquito vectors, host serology, and their temporal and spatial availability (e.g., relative abundance) to the mosquitoes.[1,6–9] For example, Apperson and others found that *Culex pipiens* (the species most often reported as the principal zoonotic vector of WNV in the east-central United States) has a broad avian host range, but this vector species did not randomly feed on bird species based on their abundance as predicted by breeding bird surveys.[6] In addition, viremic transient birds have been suggested as the means by which WNV has rapidly spread across North and Central America,[10–12] while resident breeding birds are probably critical to maintain and amplify the zoonotic cycle.[1,13,14]

The presence of seropositive birds may not be an indication of current WNV activity, but is an indication of exposure to WNV-carrying mosquitoes and survivorship, as seen in research with St. Louis encephalitis.[14] Recent research suggests that persistence of antibody to WNV was at least 60 weeks in rock pigeons.[8] Other studies on St. Louis encephalitis virus (SLEV), a closely related virus, found that birds may retain antibodies throughout their life.[4,15] Therefore, after the initial introduction of WNV to an area, juvenile rather than adult seroprevalence data may provide a better index of current WNV activity in an area.

Seroprevalence in adult avian species can be used to determine the initial spread of WNV across a region, an approach taken by a study on seroprevalence during the spread of the virus across Illinois in 2002.[14] We can now determine if seroprevalence rates have changed over time between regions and species. The number of human cases of WNV in Illinois decreased by more than 10-fold after 2002; however, reports of infected birds and mosquitoes remained relatively high.[3] This study investigates the annual distribution and extent of WNV exposure in avian species after the initial outbreak in humans as enzootic cycles appear to become established within the wildlife of Illinois. With these seroprevalence data, we can begin to address the long-term effect of WNV on birds and to identify what makes specific species more likely to be exposed to WNV.

MATERIALS AND METHODS

Field collection. Wild birds were captured at 60 different sampling sites from September 2001 through November 2004. Not all 60 sites were sampled every year. Nine of the sites were sampled two or more of the four years. Of these nine, two were in northern Illinois, four in central Illinois, and three in southern Illinois. These three regional divisions represent a latitudinal gradient that corresponds to climatic and ecologic differences. Sampling sites were changed yearly based on how successful we were at catching birds at them, coordination with mosquito sampling, and differences in the amount of personnel available. In addition, game birds were only sampled over a short time span, in conjunction with activities of the Illinois Department of Natural Resources. All other

* Address correspondence to Robert J. Novak, Medical Entomology Laboratory, Illinois Natural History Survey, 1910 Griffith Drive, North 104 Annex, Champaign, IL 61820. E-mail: rjnovak@uiuc.edu

sites were sampled every two weeks. Birds were primarily captured using mist nets and potter traps (small four-chambered box traps). At some sites, birds were captured year-round; however, at other sites birds were only captured during the breeding season. Captured birds were identified by species and, if possible, by sex and age.[16–18] Individuals were classified as adults (those born the previous breeding seasons) and as juveniles (those born during the current breeding season). In addition, species were classified as those that were thought to be migrating through Illinois (transients) versus individuals believed to have bred within the area. Individuals that were captured during the breeding season (May–August) and that were captured in suitable habitat were considered breeding individuals. Individuals that are known not to breed in the state or to only have a small breeding area, not near where the individuals were captured, were considered transients. Once captured, a blood sample was taken from the jugular or brachial vein, depending on the size of the bird, using a 27-gauge ½-inch needle attached to a syringe. Depending on the size of the bird, quantities of blood taken from each bird ranged from 0.1 to 0.2 mL. Birds were banded with U.S. Geological Survey aluminum leg bands (Permit no. 06507) to ensure that a bird was only used once in the analysis, and to obtain data for future research on how long antibodies persist within an individual.

Statistical analysis. To determine what percentage of each species were seropositive in each region, we divided the number of seropositive individuals of a species by the total number of seropositive individuals. When comparing the most seropositive species in each region, we used only species that had a sample size of 15 (14 species). All other comparisons were analyzed using either chi-square or Fisher's exact tests (Statview, SAS, Cary, NC). When determining the abundance of particular species, we used spring bird count data.[19] Other studies have used a breeding bird survey[6]; however, these censuses are typically not conducted in urban areas. Therefore, we believe they do not represent the true regional abundance. The spring bird count is conducted throughout the entire state by more than 1,000 volunteers early in the breeding season and in all habitats. Thus, we believe it provides a better representation of the relative species abundance.[19]

Laboratory testing. After capture and bleeding of the birds, blood samples were brought back to the laboratory to be tested for antibodies to WNV. An epitope-blocking enzyme-linked immunosorbent assay was used to detect the presence of antibodies to WNV using three different monoclonal antibodies (MAbs): 3.1112G, 2B2, and 6B6C-1.[14] Monoclonal antibody 3.1112G is specific for NV, while MAbs 2B2 and 6B6C-1 can react with other viruses, including WNV. For a serum sample to be considered positive for antibodies to WNV, it had to block the binding of all three MAbs by > 30% relative to the negative control, normal chicken serum (Vector Laboratories, Burlingame, CA), which readily allows the binding of MAbs.[7] Serum from WNV-infected horses was used as a positive control.[20]

RESULTS

Over the course of this study, 5,236 wild, free-ranging birds belonging to 11 orders, 36 families, and 145 species were sampled. Positive wild birds represented 6 orders, 19 families, and 38 species (Table 1). Despite the diversity of species exposed to WNV, the top five species (of those with more than 15 individuals sampled) throughout the state, in terms of the percentage that were seropositive, were Wild Turkey (40.5%, n = 37), Mourning Dove (38.1%, n = 97), Northern Cardinal (24.7%, n = 389), American Robin (15.2%, n = 341), and House Sparrow (9.6%, n = 1,207). Several species never tested positive for antibodies to WNV, including, Downy Woodpecker (n = 39), Tufted Titmouse (n = 49), and Black-Capped or Carolina Chickadee (because of difficulty in identifying chickadee species, we combined the data for the two; n = 76). The species with the highest exposure rates differed both temporally and regionally. In 2002, data (i.e., both adult and juveniles) from Ringia and others[14] showed that the species with the highest seroprevalence was Northern Cardinal (12.4%), while in 2003 Wild Turkey and Mourning Dove (40.5%) were the highest, and in 2004 Mourning Dove (42.1%) was the highest (Table 1).

The exposure rates within species differed regionally (Table 2). There was a significant difference in the exposure rates of Northern Cardinals ($\chi^2 = 515.51$, n = 485, $P < 0.01$), American Robins ($\chi^2 = 144.30$, n = 393, $P < 0.01$), and Mourning Doves ($\chi^2 = 271.23$, n = 134, $P < 0.01$) between collection sites in the north, central, and southern regions. These differences are probably not due to differences in abundance because these species are abundant throughout the state.[21] An evaluation of juvenile seroprevalence rates

TABLE 1
West Nile virus seropositive birds tested from September 2001 to November 2004*

Common/scientific name	2002†		2003		2004		2002–2004 Total sampled
	Total	Ab+ (%)	Total	Ab+ (%)	Total	Ab+ (%)	
American Goldfinch/*Carduelis cristis*	10	0	25	0	38	1 (2.6)	73
American Robin/*Turdus migratorius*	79	3 (3.8)	184	39 (21.2)	78	10 (12.8)	341
Blue Jay/*Cyanocitta cristata*	18	0	15	1 (6.7)	13	0	46
Brown Thrasher/*Toxostoma rufum*	9	2 (10)	17	3 (17.6)	12	1 (8.3)	38
Brown-headed Cowbird/*Molothrus ater*	58	0	64	8 (12.5)	38	0	160
Canada Goose/*Branta Canadensis*	253	3 (1.2)	58	3 (5.2)	175	9 (5.1)	320
Carolina Wren/*Thryothorus ludovicianus*	9	0	4	2 (50)	16	0	29
Cedar Waxwing/*Bombycilla cedrorum*	5	1 (20)	5	1 (20)	4	0	14
Chipping Sparrow/*Spizella passerina*	8	0	4	0	15	1 (6.7)	27
Common Grackle/*Quiscalus quiscula*	46	0	175	5 (2.9)	57	2 (3.5)	278
Eastern Bluebird/*Sialia sialis*	7	0	10	3 (30)	10	0	27
Eastern Wood-Pewee/*Contopus virens*	9	0	14	1 (7.1)	8	0	31
European Starling/*Sturnus vulgaris*	3	0	14	1 (7.1)	7	0	24
Field Sparrow/*Spizella pusilla*	4	0	15	1 (6.7)	14	0	33
Gray Catbird/*Dumetalla carolinensis*	72	6 (8.3)	57	9 (15.8)	58	1 (1.7)	187
House Finch/*Carpodacus mexicanus*	2	0	28	4 (14.3)	20	1 (5)	50
House Sparrow/*Passer domesticus*	185	21 (11.4)	722	77 (10.7)	207	18 (8.7)	1,114
Indigo Bunting/*Passerina cyanea*	28	1 (3.6)	38	1 (2.6)	31	1 (3.2)	97
Mallard/*Anas platyrhynchos*	27	0	–	–	–	–	27
Mourning Dove/*Zenaida macroura*	9	1 (11.1)	69	28 (40.5)	19	8 (42.1)	97
Northern Cardinal/*Cardinalis cardinalis*	129	16 (12.4)	138	41 (29.7)	122	39 (32)	389
Northern Flicker/*Colaptes auratus*	3	0	9	1 (11.1)	9	0	21
Northern Mockingbird/*Mimus polyglottos*	2	0	6	2 (33.3)	1	0	9
Northern Waterthrush/*Seiurus noveboracensis*	4	0	7	0	11	1 (9.1)	22
Ovenbird/*Seiurus aurocapillus*	32	1 (3.1)	26	2 (7.7)	12	1 (8.3)	70
Red-headed Woodpecker/*Melanerpes erythrocephalus*	1	0	4	1 (25)	1	0	6
Red-winged Blackbird/*Agelaius phoeniceus*	39	3 (7.7)	28	0	18	1 (5.6)	85
Rose-breasted Grosbeak/*Pheucticus ludovicianus*	–	–	8	1 (12.5)	6	2 (33.3)	14
Song Sparrow/*Melospiza melodia*	13	0	24	1 (4.2)	12	0	49
Swainson's Thrush/*Catharus ustulatus*	32	1 (3.1)	10	1 (10)	33	0	75
White-breasted Nuthatch/*Sitta carolinensis*	5	0	9	1 (11.1)	7	0	21
White-throated Sparrow/*Zonotrichia albicollis*	99	0	73	0	25	1 (40)	197
Wild Turkey/*Meleagris gallopavo*	–	–	37	15 (40.5)	–	–	37
Wood Duck/*Aix sponsa*	140	5 (3.6)	66	1 (1.5)	90	2 (2.2)	296
Wood Thrush/*Hylocichla mustelina*	1	0	7	2 (28.6)	–	–	8
Yellow-bellied Cuckoo/*Coccyzus americanus*	5	0	1	1 (100)	–	–	6
Yellow-breasted Chat/*Icteria virens*	1	0	14	2 (14.3)	5	2 (40)	20
Yellow-rumped Warbler/*Dendroica coronata*	25	0	44	0	5	1 (20)	74

* AB = antibody.
† Ringia and others.[14]

DISCUSSION

Over the course of this study, a large number of avian species were exposed to WNV throughout northern, central, and southern Illinois. However, not all species had the same seroprevalence, and the species-specific exposure rates varied between year and region. The regional difference is illustrated by the variability in the species that compose the highest percentage of seropositive birds (Table 2). Although the north and central regions have similar species, the most common seropositive species in both regions are Mourning Doves and Northern Cardinals (Table 2). Several species were also only found to be seropositive in one region; additional research is needed to determine why species seroprevalence differs regionally. Research should address whether habitat features, species assemblages, or differences in mosquito species account for these differences.

The temporal variation in seroprevalence from 2001 to 2004 in Illinois may reflect the spread and establishment of WNV in Illinois. Although the WNV epidemic occurred in 2002, our data suggest a WNV epizootic among birds occurred in 2003 followed by a marked decrease in WNV activity in 2004. There are two plausible explanations for the decrease in 2004. The first is that climatic factors resulted in lower mosquito abundance and WNV activity in mosquitoes.[24] The second is that the increase in seroprevalence in 2003 reduced the number of available hosts in 2004, thus regulating the intensity of transmission. This feedback loop, where the increasing seroprevalence decreases the likelihood of an infectious vector finding a susceptible host, is only valid if the rate of turnover is relatively low.[25] Although this may be unlikely, given the usually high turnover in most bird species, we believe it does deserve additional attention.

Although the data suggest that adults are more likely to be seropositive than juveniles, juvenile data provides a much better index of the temporal and regional transmission levels because antibodies persist for at least one year.[4,8] Therefore, the use of seroprevalence data from adult birds may suggest that birds are being exposed at a greater rate at one site than another, but these differences in exposure may not represent the year from which they were sampled. This is because in many species adults exhibit high site fidelity.[26] Therefore, one outbreak year may result in adult seroprevalence being relatively high for several years, whereas juvenile seroprevalence

TABLE 2
Seroprevalence for West Nile virus of species by region

Species	North		Central		South	
	No. of birds sampled	No. of birds antibody + (%)	No. of birds sampled	No. of birds antibody + (%)	No. of birds sampled	No. of birds antibody + (%)
American Robin	62	8 (12.9)	241	43 (17.8)	38	1 (2.6)
Brown Thrasher	3	0	20	4 (20)	25	2 (8)
Brown-headed Cowbird	29	2 (6.9)	126	6 (4.8)	–	–
Canada Goose*	215	14 (6.5)	139	1 (.7)	132	0
Common Grackle	92	0	162	6 (3.7)	24	1 (4.2)
Gray Catbird	63	3 (4.8)	101	13 (12.9)	23	0
House Finch	9	0	31	4 (12.9)	10	1 (10)
House Sparrow†	358	35 (9.8)	755	81 (10.7)	–	–
Indigo Bunting	–	–	27	0	62	3 (4.8)
Mourning Dove	10	2 (20)	83	35 (42.2)	4	0
Northern Cardinal	23	8 (34.8)	140	55 (39.3)	226	33 (14.6)
Red-winged Blackbird	60	4 (6.7)	4	0	20	0
Wood Duck*	65	5 (7.7)	69	2 (2.9)	162	1 (.6)
Yellow-breasted Chat‡	–	–	–	–	20	4 (20)

* Species was only sampled during a 1–2-week period week during the summer of any given year.
† Species was not sampled in the southern portion of the state due to trapping locality.
‡ Due to the range and habitat of this species it was only captured in the southern region.

data would provide a better index of transmission levels that year.

Our research suggests that breeding birds, in addition to juveniles, should also be used when accessing WNV activity. Although transient birds have been implicated as a means by which WNV rapidly spreads across regions,[11,12,22] several recent studies have suggested that transient birds may play a relatively small role in the dispersal of viruses.[4,10,27] Although this remains one of the major epidemiologic questions still to be answered about WNV,[28] we were not investigating the role transient birds play in the spread of the virus. Our data only suggest that few transients were exposed to WNV relative to that of breeding birds. A possible explanation for the low seroprevalence of transients is that fewer transient were exposed to WNV because most breed farther to the north where there are decreased levels of WNV activity.

Seropositive individuals are not necessarily good reservoir hosts. Laboratory studies reported by Komar and others[2] and Reisen and others[9] suggested that certain species are more competent (higher and longer viremia) than others as reservoir hosts for WNV. Reisen and others[9] found that although corvids produce the highest viremias, House Sparrows and House Finches, which are more evenly distributed, may also be important hosts for effective WNV transmission. We found that many of the species that Komar and others[2] and Reisen and others[9] found to be possible competent hosts have high WNV seroprevalence. Whereas some birds, such as corvids, exhibit a high mortality from WNV and thus tend to have a low seroprevalence rate, it seems reasonable to assume that birds with high seroprevalence rates experience a much lower mortality from the virus. For example, Reisen and others[9] found that although only 16% of House Sparrows died of exposure to WNV, 63% of House Finches died. Three of our top five WNV seroprevalent species (American Robin, Mourning Dove, House Sparrow) were also considered by Komar and others[2] and Reisen and others[9] to be competent hosts. In our study, the Northern Cardinal and the Wild Turkey both exhibited high WNV seroprevalence rates, but their competency were not determined in the studies of Komar and others[2] or Reisen and others.[9] Because of the high seroprevalence of Northern Cardinals, we believe that this species should receive additional attention to determine its host competency to evaluate its potential role in WNV zoonotic cycles in Illinois. Although Komar and others[2] considered the Mourning Dove to be a poor candidate as a reservoir host because of its short viremia, the high seroprevalence rates in our study indicate that this species has some behavioral characteristic(s) that result in it being frequently exposed to infected mosquitoes. In Illinois, the high seroprevalence of Mourning Doves, coupled with high abundance, may offset their low competency in regard to their importance in as a reservoir host. Species abundance must be taken into account when determining how important species are in the zoonotic cycle.[29] This is best illustrated by House Sparrows. Although they are not experiencing the highest exposure (9.6%) in Illinois, their high abundance, particularly in urban areas, may result in them being a very important species in the zoonotic cycle, as seen in research with SLEV.[26]

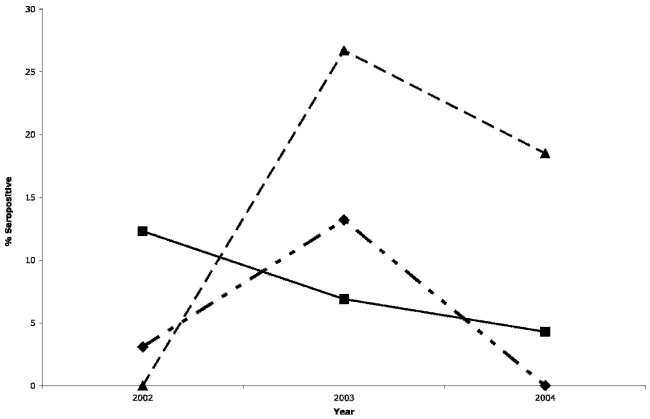

FIGURE 1. Species whose juveniles have the highest West Nile virus seroprevalence from 2002 through 2004. Line with ▲ = American Cardinal; Line with ■ = House Sparrow; Line with ♦ = American Robin.

Many of the characteristics of the WNV epizootic are remarkably similar to what has been reported during outbreaks

TABLE 3
Characteristics of bird species with the highest seroprevalence for West Nile virus

Rank	Species	No.	Seropositive	Broods per year	Association with human	Timing of breeding
1	Red-winged Blackbird	85	4 (4.7)	Single	Weak	Early
2	Common Grackle	278	7 (2.5)	Single	Medium	Early
3	European Starling	24	1 (4.2)	Multiple	Strong	Throughout
4	American Robin	341	52 (15.2)	Multiple	Strong	Throughout
5	House Sparrow	1,210	116 (9.6)	Multiple	Strong	Throughout
6	Canada Goose	487	15 (3.1)	Single	Medium	Early
8	American Goldfinch	73	1 (1.4)	Single	Medium	Late
9	Mourning Dove	97	37 (38.1)	Multiple	Strong	Throughout
10	Northern Cardinal	389	96 (24.7)	Multiple	Strong	Throughout
11	Brown-headed Cowbird	162	8 (4.9)	Brood parasite	Medium	Throughout
15	Blue Jay	46	1 (2.2)	Single	Medium	Throughout

of SLEV. The activity of both viruses peak in late summer, their major vector in the midwest is *Cx. pipiens*, there is regional variation in preferred bird hosts, and the primary hosts are most often in the orders Columbiformes (i.e., doves) and Passeriformes (e.g., sparrow, robins, cardinals, etc.).[15] In the Midwest, the common hosts for SLEV are House Sparrow, Blue Jay, American Robin, Northern Mockingbird, and Northern Cardinal.[30] In the 1975 SLEV epidemic in Chicago, America Robins were implicated as the major reservoir host for SLEV.[15] It is interesting that four of the five birds with the greatest exposure to WNV in our study were also important species in SLEV epizootics. The obvious difference between the two viruses is that bird mortality associated with WNV appears to be much greater. This high mortality may result in WNV persisting in an area longer than SLEV. When birds are removed from an area other, non-territorial, birds rapidly move in. This replacement of removed birds by new birds has been observed in 63 experiments with 53 species.[31] The replacement can occur as fast as a few minutes or take up to two days.[31] Therefore, although birds in an area infected with SLEV might acquire immunity, the mortality associated with WNV might result in a net influx of susceptible hosts, possibly perpetuating the epizootic cycle.

There appears to be common natural history traits among the species with high seropositive rates and those with low seropositive rates. Four of the five species with the highest seroprevalence rates have commonalities among them. As previously stated, they are all multiple brooded, breed throughout the breeding season, and have a strong association with humans due to their habitulization in residential/urban areas.[21,23,31] Also, all of the species are residents, except American Robins, which are short-distance migrants.

A good example of the differences in seroprevalence between species is illustrated by Common Grackles and American Robins. These species nest in close proximity, often in the same tree; however, Common Grackles nest much earlier.[23] This difference in breeding phenology may explain why the proportion of grackles with antibodies to WNV is approximately 3%, while approximately 12% of Robins have antibodies to this virus. This difference may be related to how lethal WNV is to a species, but it may also be due to the behavior of the species, particularly when the species nests and form communal roosts. Grackles breed extremely early (April), form large communal roosts in mid-summer, and begin migrating in mid to late summer. American Robins breed throughout the summer months and form communal roosts late in the summer.[21,23,31] Because roosting probably occurs when birds are exposed to WNV-carrying mosquitoes (i.e., *Cx. pipiens*), the roosting behavior of birds, especially when communal roosts are formed, may provide an explanation for the difference in seroprevalence between species.[15] Grackles may not be exposed to mosquitoes that carry WNV because they breed when fewer infected mosquitoes are present, and their roost behavior in Illinois does not predispose them to being exposed to WNV, while the communal roosts of American Robins late in the summer may attract large numbers of mosquitoes.

Because WNV is new to North America it is important to determine how the avifauna acclimates to its presence. Long-term studies are needed to characterize the enzootic cycle of WNV. However, it is also important to determine if the species that are not seropositive are in areas where other species that are experiencing high exposure rates are refractory, not being exposed, or dying after being exposed. For example, several cavity nesting species never tested positive for antibodies to WNV in our study, including, Downy Woodpecker, Tufted Titmouse, and Black-capped or Carolina Chickadee, despite a reasonable capture rate for each species. Although, seroprevalence data provide an insight into how WNV activity differs both temporally and spatially, and which species may be most important in an enzootic cycle, more research is needed on the transmission dynamics of WNV.

Received March 22, 2005. Accepted for publication July 14, 2005.

Acknowledgments: We thank the Forest Preserve Districts of Illinois, the Illinois State Parks, Southern Illinois University, and the University of Illinois at Champaign-Urbana for use of research sites; the field crew (Brad Danner, Evette Vlach, Stephanie Eicholz, Bill Anderson, Brett Amdor, Joe Heisinger, Jennifer Wise, Arlo Raim, Sarah Yaremych, Christine LaPointe, Bill Stewart, Derrick Robertson, and Amanda Kamradt) for their help with this study; and Weidong Gu and David Enstrom for reviewing the manuscript. This study was conducted under animal use (collecting) permit no. 04010.

Financial support: This study was supported by grants to Robert J. Novak from the Illinois Department of Natural Resources Waste Tire Act, the Centers for Disease Control and Prevention (PHS U50/CCU 52051), and the U.S. Department of Agriculture Cooperative State Research, Educational, and Extension Service (PHS U50 CCU52051: West Nile Virus, Illinois).

Authors' addresses: Tara A. Beveroth, Michael P. Ward, Richard L. Lampman, and Robert J. Novak, Medical Entomology Laboratory, Illinois Natural History Survey, 1910 Griffith Drive, North 104 Annex, Champaign, IL 61820, Telephone: 217-333-1186, Fax: 217-333-2359, E-mails: beveroth@uiuc.edu, mpward@uiuc.edu,

rlampman@uiuc.edu, and rjnovak@uiuc.edu. Adam M. Ringia, U.S. Fish and Wildlife Service, 1590 Williamsbridge Road, Bronx, NY 10461, Telephone: 718-415-3451, E-mail: ringia@yahoo.com.

REFERENCES

1. Centers for Disease Control and Prevention. West Nile virus activity. Available from http://www.cdc.gov/ncidod/dvbid/westnile/background.htm. Accessed December 2004.
2. Komar N, Langevin S, Hinten S, Nemeth N, Edwards E, Hettler D, Davis B, Bowen R, Bunning M, 1999. Experimental infection of North American birds with the New York 1999 strain of West Nile virus. *Emerg Infect Dis 9:* 311–322.
3. Illinois Department of Public Health. West Nile virus. Available from http://www.idph.state.il.us/envhealth/wnv.htm. Accessed December 15, 2004.
4. Reisen WK, Chiles VM, Martinez YF, Green EN, 2003. Experimental infection of California birds with western equine encephalomyelitis and St. Louis encephalitis viruses. *J Med Entomol 40:* 968–982.
5. Centers for Disease Control and Prevention. West Nile virus activity. Available from http://www.cdc.gov/ncidod/dvbid/westnile/birdspecies.htm. Accessed February 2005.
6. Apperson CS, Harrison BA, Unnasch TR, Hassan HK, Irby WS, Savage HM, Aspen SE, Watson DW, Rueda LM, Engber BR, Nasci RS, 2002. Host-feeding habits of *Culex* and other mosquitoes (Diptera: Culicidae) in the Borough of Queens in New York City, with characters and techniques for identification of *Culex* mosquitoes. *J Med Entomol 39:* 777–785.
7. Blitvich GJ, Marlenee NL, Hall RA, Calisher CH, Bowen RA, Roehrig JT, Komar N, Langevin SA, Beaty BJ, 2003. Epitope-blocking enzyme-linked immunosorbent assays for the detection of serum antibodies to West Nile virus in multiple avian species. *J Clin Microbiol 41:* 1041–1047.
8. Gibbs SEJ, 2005. WNV antibody persistence in pigeons. *Southeastern Cooperative Wildlife Disease Study Briefs 20:* 3–4.
9. Reisen WK, Fang Y, Martinez M, 2005. Avian host and mosquito (Diptera: Culicidae) vector competence determine the efficiency of West Nile and St. Louis encephalitis virus transmission. *J Med Entomol 42:* 367–374.
10. Rappole JH, Derrickson SR, Hubalk Z, 2000. Migratory birds and spread of West Nile virus in the western hemisphere. *Emerg Infect Dis 6:* 319–328.
11. Peterson AT, Vieglais DA, Andreasen JK, 2003. Migratory birds modeled as critical transport agents for West Nile virus in North America. *Vector Borne Zoonotic Dis 3:* 27–37.
12. Dupuis AP II, Marra PP, Kramer LD, 2003. Serologic evidence of West Nile virus transmission, Jamaica, West Indies. *Emerg Infect Dis 9:* 860–863.
13. Reisen WK, Lundstrom JO, Scott TW, Eldridge BF, Chiles RE, Cusack R, Martinez VM, Lothrop HD, Gutierrez D, Wright SE, Boyce K, Hill BR, 2000. Patterns of avian seroprevalence to western equine encephalomyelitis and St. Louis encephalitis viruses in California, USA. *J Med Entomol 37:* 507–527.
14. Ringia AM, Blitvich BJ, Koo H, van de Wyngaerde M, Brawn J, Novak RJ, 2004. Antibody prevalence of West Nile virus in birds, Illinois, 2002. *Emerg Infect Dis 10:* 1120–1124.
15. McLean RG, Bowen G, 1980. Vertebrate hosts. Monath TP, eds. *St. Louis Encephalitis*. Washington, DC: American Public Health Association, 381–450.
16. Pyle P, 1997. *Identification Guide to North American Birds: Part I, Columbidae to Ploeidae*. Bolinas, CA: Slate Creek Press.
17. Peterson RT, 1980. *A Field Guide to the Birds of Eastern and Central North America.* Fourth edition. Boston: Houghton Mifflin Co.
18. Sibley D, 2000 *The Sibley Guide to Birds*. First edition. New York: Alfred A. Knopf, Inc.
19. Kleen VM, 2004. The 2004 Illinois statewide spring bird count. *Meadowlark 13:* 132–141.
20. Blitvich BJ, Bowen RA, Marleen NL, Hall RA, Bunning ML, Beaty BJ, 2003. Epitope-blocking enzyme-linked immunosorbent assays for detection of West Nile virus antibodies in domestic mammals. *J Clin Microbiol 41:* 2676–2679.
21. Kleen VM, Cordle L, Montgomery RA, 2004. *The Illinois Breeding Bird Atlas*. Champaign, IL: Illinois Natural History Survey Special Publication.
22. Komar O, Robbins MB, Klenk K, Blitvich BJ, Marlenee NL, Burkhalter KL, Gubler DJ, Gonzálvez, Peña CJ, Peterson AT, Komar N, 2003. West Nile virus transmission in resident birds, Dominican Republic. *Emerg Infect Dis 9:* 1299–1302.
23. Bohlen DH, 1989.*The Birds of Illinois*. Bloomington, IN: Indiana University Press.
24. Ahumada JA, Lapointe D, Samuel MD, 2004. Modeling the population dynamics of *Culex quinquefasciatus* (Diptera: Culicidae), along an elevational gradient in Hawaii. *J Med Entomol 41:* 1157–1170.
25. John TJ, Samuel R, 2000. Herd immunity and herd effect: new insights and definitions. *Eur J Epidemiol 16:* 601–606.
26. Greenwood PJ, Harvey PH, Perrins CM, 1979. The role of dispersal in the great tit (*Parus major*): the causes, consequences and heritability of natal dispersal. *J Anim Ecol 48:* 123–142.
27. Komar N, Burns J, Dean C, Panella NA, Dusza S, Cherry B, 2001. Serologic evidence for West Nile virus infection in birds in Staten Island, New York, after an outbreak in 2000. *Vector Borne Zoonotic Dis 1:* 191–196.
28. Marra PP, Griffing S, Caffrey CA, Kilpatrick AM, McClean R, Brand C, Saito E, Dupuis AP, Kramer L, Novak R, 2004. West Nile virus and wildlife. *Bioscience 54:* 393–402.
28. Lord RD, Calisher CH, Doughty WP, 1973. Assessment of bird involvement in three urban St. Louis encephalitis epidemics. *Am J Entomol 99:* 364–367.
29. Monath TP, 2000. The Arboviruses: Epidemiology and ecology. *Evidence from Field Studies*. Boca Raton: CRC Press Inc., 127–128.
30. Newton I, 1998. *Population Limitation in Birds*. San Diego: Academic Press, Inc.
31. Ehrlich PR, Dobkin DS, Wheye D, 1988. *The Birder's Handbook: A Field Guide to the Natural History of North American Birds*. New York: Simon and Schuster Inc.