

Genetic Diversity of *Aedes vexans* (Diptera, Culicidae) from New Orleans: Pre- and Post-Katrina

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Abstract The floodwater mosquito *Aedes vexans*, a potential vector of West Nile virus, has a worldwide distribution that includes the continental United States and southern Canada. In order to determine the effect that Hurricane Katrina had on the temporal genetic variation of *Ae. vexans* from New Orleans, we compared genetic diversity of a portion of the mtDNA *ND5* gene of mosquito specimens collected during 2005 ($n = 99$) with specimens from 2006 ($n = 103$), after the hurricane. Average haplotype diversity (Hd) was high (>0.88) in 2005 and 2006 for both the parishes studied. It does not appear that Hurricane Katrina had any impact on genetic diversity, and despite the intense efforts to control mosquitoes in New Orleans, *Ae. vexans* has not undergone a population bottleneck. A bottleneck effect may be lacking because this species breeds outside the city and the adults migrate into the city.

Keywords *Aedes vexans* · Mosquito mtDNA · Genetic diversity · Haplotype · Hurricane Katrina

Introduction

Aedes vexans (Meigen), a floodwater mosquito present in both the New and Old World, is widespread through the North American continent (Horsfall 1972; Darsie

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and Ward 2005). It is an abundant nuisance mosquito that usually composes a high percentage of the total mosquito diversity and thrives in floodwater habitats where rain followed by water accumulation and recession is continuous. Species composition studies from Russia, Croatia, and the United States confirm its abundance (Merdic and Lovakovic 2001; Kent et al. 2003; Fyodorova et al. 2006). The widespread distribution of *Ae. vexans* may be due to its capacity for long-distance travel. Studies have documented that females can fly up to 10–17 km (Briegel et al. 2001), and adult females have been documented flying from south central Texas to New Orleans, Louisiana (M. Carroll pers. comm.).

Aedes vexans can competently transmit more than 30 viruses (Horsfall and Novak 1991). In the United States, viral diseases of veterinary and public health importance, such as West Nile virus (Goddard et al. 2002), St. Louis encephalitis, and eastern equine encephalitis (Cupp et al. 2004), are known to be transmitted by *Ae. vexans*. In Northern Africa, this mosquito has become an important vector in outbreaks of Yellow Rift Valley fever, which is characterized by abortion and death of young ungulates and hemorrhagic fever in cattle (Fontenille et al. 1998). Besides its ability to transmit viruses, *Ae. vexans* can competently transmit the L₃ stage of the nematode *Dirofilaria immitis*, causal agent of dog heartworm (Ludlam et al. 1970).

The study of population genetics can help to understand vector transmission, epidemiology, and control of diseases (Tabachnick and Black 1995). These studies can also assist in the identification of discrete populations across a determined area, determine their geographic distribution, and evaluate possible reproductive isolation (Lanzaro and Tripet 2003). Such studies are also important to estimate the rate of gene spread within and among populations at different spatial scales and to identify biological and physical features of the environment that may interfere with their movement (Lanzaro and Tripet 2003).

Currently, several different molecular regions are often used for population genetics studies. One of those regions is mitochondrial DNA (mtDNA), which is haploid and maternally inherited (Avisé 2004). Population genetics studies for mosquitoes of public health importance in the genus *Aedes*, *Anopheles*, and *Culex* within the last decade have relied primarily on mtDNA and microsatellite markers (Besansky et al. 1997; Birungi and Munstermann 2002; Kent et al. 2007; Venkatesan et al. 2007).

Most population genetics studies in general, and of mosquitoes in particular, are single-year studies, in which the main objective is to examine spatial differences among populations; very few studies have looked at temporal patterns of genetic structuring. Only a few of those temporal studies have looked at the effects of climate and weather changes on populations of mosquitoes. One study investigated the population structure differences of *Ae. aegypti* in Vietnam in relation to dry and rainy seasons (Huber et al. 2002), and another was a study that examined the population structure of *An. arabiensis* from Northern Africa across seasons (Kent et al. 2007).

The first informative study that examined the population structure of *Ae. vexans* from five states in the central United States based on *ND5* sequence data revealed high genetic differentiation at the local level, with only two haplotypes present

throughout all locations (Szalanski et al. 2006). Despite the importance of *Ae. vexans* in the transmission of both viral diseases and of the nematode *D. immitis*, the genetic structure of this mosquito and its variability across populations still needs to be elucidated in a smaller geographic area. It remains to be determined what changes are produced in genetic structure due to the impact of drastically changing climatic conditions or environmental factors.

Hurricane Katrina was a natural disaster that caused major human and economic losses in New Orleans, Louisiana, and other southern states. Flooding of the city may have had an impact on populations of insects and other invertebrates, because the city was inundated for several days, and ditches and levees were broken, allowing brackish water into city perimeters.

Our hypothesis is that Hurricane Katrina had an impact on populations of *Ae. vexans*, resulting in a genetic bottleneck, and therefore reducing the genetic diversity of haplotypes after the hurricane. The objective of this study was to characterize the genetic structure of *Ae. vexans* across a small geographic scale and determine the possible effect that Hurricane Katrina had on populations of this mosquito.

Materials and Methods

Collection of Samples

Female adult specimens of *Ae. vexans* were collected from Orleans, Jefferson, and St. Bernard parishes during the months of March through July for 2005 and 2006, utilizing CDC light traps baited with dry ice (Table 1; Fig. 1). Specimens were identified using the keys of Means (1979) and Darsie and Ward (2005) and stored at -20°C until processed for DNA extraction. Specimens were collected weekly, totaling approximately 500 specimens each year. Voucher specimens have been deposited in the Arthropod Museum, Department of Entomology, University of Arkansas, Fayetteville.

DNA Extraction, Amplification, and Sequencing

Genomic DNA from individual mosquitoes was extracted using either the Qiagen DNeasy extraction kit (Qiagen, Valencia, CA) or the Puregene DNA extraction kit (Gentra, Minneapolis, MN), according to the manufacturer's protocol. From the 2005 collection, 99 samples were subjected to genetic analysis, and 103 samples were used from the 2006 collection. A 423-bp region of the mitochondrial NADH dehydrogenase subunit 5 (*ND5*) gene was PCR-amplified in a Techne thermal cycler (Techne Inc., Burlington, NJ) using primers 6500 (5'-TCCTTAGAATAAAATCCCGC-3') and 7398 (5'-GTTTCTGCTTTAGTTCATTCTTC-3') (Birungi and Munstermann 2002), and the PCR protocol of Szalanski et al. (2000). Briefly, the PCR profile consisted of 35 cycles of 94°C for 45 s, 46°C for 45 s, and 72°C for 45 s. Amplicons were resolved using 1% agarose gels and photodocumented using a BioDoc-It Imaging System (UVP, Inc., Upland, CA). Afterward, PCR products

Table 1 New Orleans populations of *Aedes vexans* examined during 2005 and 2006

Parish and locality	Latitude Longitude	2005 Haplotypes (frequency)	2006 Haplotypes (frequency)
Orleans Parish		(55)	(87)
Midcity	29:57:16N 90:04:30W	1(2), 4(1), 31(2), 45(1)	–
New Orleans East	20:59:16N 90:01:17W	1(3), 2(1), 4(6), 50(1), 51(1), 62(1)	–
Lakeview	30:00:24N 90:06:28W	1(1), 46(1), 48(1), 49(1)	–
Algiers	29:56:41N 90:02:48W	4(1), 7(1), 19(1)	–
Seine St., Algiers	29:55:29N 90:01:42W	–	1(2), 4(3), 31(3), 53(1), 61(1), 68(1), 69(1), 70(1), 71(1), 72(1), 73(1), 74(1), 75(1)
Willow Dr., Algiers	29:54:53N 89:56:17W	–	1(3), 4(2), 25(1), 76(1), 77(1), 78(1), 79(1), 80(1), 81(1), 82(1), 83(1), 84(1)
Pelican, Algiers	29:57:11N 90:02:55W	–	4(1)
Tall Pine, Algiers	29:54:20N 89:59:55W	–	4(4), 25(1), 53(1), 81(1), 85(1), 86(1), 87(1), 88(1), 89(1), 90(1), 91(1)
Uptown	29:55:32N 90:06:03W	4(3), 19(1), 25(1), 37(1), 52(1), 57(1), 60(1), 64(1), 65(1), 66(1)	–
Audubon 200, Uptown	29:55:39N 90:07:59W	–	1(1), 4(6), 10(1), 93(1), 94(1), 95(1), 96(1), 97(1), 98(1), 99(1), 100(1), 101(1), 102(1), 103(1), 104(1)
Hillary St., Uptown	29:56:17N 90:07:57W	–	1(1), 4(2), 31(1), 37(1), 38(3), 47(1), 75(1), 105(1), 106(1), 107(1), 109(1), 110(1)

Table 1 continued

Parish and locality	Latitude Longitude	2005 Haplotypes (frequency)	2006 Haplotypes (frequency)
Gentilly	30:00:07N 90:01:46W	1(1), 2(1), 4(2), 37(1), 38(1), 43(1), 54(1)	4(2), 31(1), 92(1)
Cutoff	29:55:04N 89:58:40W	1(1), 4(2), 25(1), 31(1), 37(1), 42(1), 56(1)	–
Jefferson Parish		(35)	(16)
Metairie	29:59:02N 90:09:10W	4(4), 38(1), 46(1), 53(1)	–
Jefferson	29:44:00N 90:06:00W	1(1), 4(1), 25(1), 38(1), 41(1), 43(1), 63(1)	–
Kenner	29:59:38N 90:14:30W	4(3), 11(1), 38(1), 44(1), 47(1), 55(1), 59(1)	–
West Bank	29:52:41N 90:05:13W	1(2), 4(3), 25(1), 37(1), 38(1), 57(1)	1(1), 4(3), 31(1), 90(1), 108(1), 111(1), 112(1), 113(1), 114(1), 115(1), 116(1), 117(1), 118(1), 119(1)
Terrytown	29:54:14N 90:01:49W	25(1), 31(1), 51(1)	–
St. Bernard Parish		(9)	(0)
Chalmette	29:56:33N 089:57:48W	1(1), 4(2), 35(1), 38(2), 58(1), 61(1), 67(1)	–
Total		(99)	(103)



Fig. 1 New Orleans sampling sites for *Aedes vexans* during 2005 (before Hurricane Katrina) and 2006 (after Hurricane Katrina)

were purified using Microcon-PCR Filter Units (Millipore, Bedford, MA) and sent to the University of Arkansas Medical Sciences DNA Sequencing Core Facility in Little Rock for direct sequencing in both directions. *Aedes vexans* haplotypes new to this study were submitted to GenBank as accession nos. GU558773-GU558921.

Genetic Analysis

DNA sequences were aligned with Clustal W (Thompson et al. 1994), and consensus sequences were obtained using Bioedit (Hall 1999). The numbers of haplotypes and their frequencies were determined both visually and with the program DNAsp version 4.10.9 (Rozas et al. 2003). DNAsp also was used to estimate the parameters of haplotype diversity (H_d) and its variance (equations 8.4 and 8.12 of Nei 1987), nucleotide diversity (π) and its variance (equations 10.5 and 10.7 of Nei 1987), mean number of pairwise nucleotide differences (K , equation A3 of Tajima 1983), number of synonymous and nonsynonymous mutations ($S + NS$), and the parameters θ_s and θ_g . Nucleotide diversity was interpreted as the average proportion of nucleotide differences between all possible pairs of sequences in the sample (Hartl and Clark 1997). The parameter θ is the proportion of nucleotide sites that are expected to be polymorphic in any suitable sample from this region of the genome (Hartl and Clark 1997).

The program Arlequin version 2.0 was used to calculate nucleotide differentiation between parishes and years by estimating F_{ST} values (Schneider et al. 2000).

Analysis of molecular variance (AMOVA) and genetic distances were also estimated with Arlequin version 2.0 (Schneider et al. 2000), with the methodology described in Excoffier et al. (1992). Levels of gene flow were determined through the effective number of migrants (N_m) between locations using DNAsp with four different formulas (Nei 1973, 1982; Lynch and Crease 1990; Hudson et al. 1992). A Mantel test (Mantel 1967) was conducted to test for isolation by distance on Arlequin. This test examines if there is any relationship between genetic and geographic distance. It was based on 1,000 permutations when computing the correlation between the distance matrices. A genealogical relationship of haplotypes was calculated using the method of Templeton et al. (1992) in the program TCS version 1.21 (Clement et al. 2000), which represents the evolutionary steps that interrelate the observed haplotypes.

Population Expansion or Selection

In order to test for neutral mutation, Tajima's (1989) D , Fu and Li's (1993) D^* and F^* , and R_2 statistics were calculated. Tajima's D was calculated using the value of θ_s based on the number of segregating sites. In order to examine demographic stability, we used either Fu's F_s statistic (based on haplotype distribution) or R_2 . R_2 appeared to be more efficient for small samples (Ramos-Onsins and Rozas 2002). In addition, the raggedness statistic (rg) based on the mismatch distribution was calculated (Harpending 1994).

Results

Haplotypes and Polymorphism

From the 202 samples subjected to DNA sequencing analysis, we found 92 distinct haplotypes (Table 2). Haplotype diversity ranged from 0.887 to 0.975. Two haplotypes (designated 1 and 4) accounted for 12.1 and 28.3% of the observed haplotypes during 2005 and 7.8 and 22.3% during 2006 (Table 1). Haplotype 31 was the next most frequent (6.8% in 2006). Haplotype 4 was present at all locations during both years, except in Terrytown during 2005. Unique haplotypes totaled 26 in 2005 and 53 in 2006. Only eight haplotypes were shared between 2005 and 2006. On average, 340 nucleotide sites were nonsynonymous and 81 were synonymous (Table 2). Nucleotide diversity ranged from 0.006 to 0.013. Despite high haplotype diversity, the mean number of pairwise nucleotide differences ranged between 2.47 and 6.01.

Genealogy

The combined, 2-year *ND5* haplotype network, based on statistical parsimony with a 95% connection limit, exhibited two haplotypes (1 and 4) as the most abundant, with a difference of only 1 bp between them (Fig. 2). The base pair difference is due to an A at position 164 on haplotype 4, where haplotype 1 has a G. Haplotype 4 also

Table 2 *ND5* mtDNA polymorphism for *Aedes vexans* from New Orleans

Parish	Sequence ^a		Haplotypes ^b		Nucleotide diversity ^c				Population expansion ^d				
	<i>N</i>	NS + S	<i>N</i>	Hd ± SD	π	<i>K</i>	θ_s	θ_g	<i>D</i> ⁺	<i>F</i> ⁺	<i>F</i> _s	<i>S</i>	<i>D</i>
	Jefferson	35	339.4 + 80.7	18	0.887 ± 0.04	0.006	2.47	0.010	4.371	-1.97	-2.12	-11.99	1.0**
Orleans	55	353.1 + 84.9	26	0.903 ± 0.03	0.006	2.68	0.015	6.557	-2.58*	-2.80*	-20.77	1.0**	-1.94*
St. Bernard	9	355.6 + 85.4	7	0.944 ± 0.07	0.007	3.17	0.008	3.679	-0.66	-0.74	-2.39	0.98	-0.65
Total 2005	99	339.4 + 80.6	41	0.898 ± 0.02	0.006	2.63	0.018	7.547	-4.44*	-4.17*	-43.56	1.0**	-2.03*
Jefferson	16	355.7 + 85.3	14	0.975 ± 0.04	0.013	6.01	0.024	10.246	-2.30	-2.58*	-6.59	1.0**	-2.05*
Orleans	87	355.7 + 85.3	52	0.943 ± 0.02	0.007	3.49	0.019	7.941	-3.19*	-3.18*	-34.16	1.0**	-1.82*
Total 2006	103	355.7 + 85.3	62	0.945 ± 0.02	0.008	3.89	0.027	11.139	-4.86*	-4.53*	-80.41	1.0**	-2.26*
Combined	202	339.4 + 80.6	92	0.922 ± 0.01	0.007	3.22	0.032	12.749	-6.93*	-5.81*	-146.1	1.0**	-2.40*

^a *N* number of sequences; NS + S number of nonsynonymous and synonymous mutations

^b *N* number of haplotypes; Hd + SD haplotype diversity ± standard deviation

^c π nucleotide diversity; *K* mean number of pairwise nucleotide differences; θ_s theta per site; θ_g theta per gene

^d *D*⁺ and *F*⁺ (Fu and Li 1993); *F*_s Fu's *F*_s statistic, based on haplotype distribution; *S* (Strobeck 1987); *D* Tajima's *D* (Tajima 1989)

* *P* < 0.05

Table 3 Pairwise genetic differences (F_{ST}) among *Aedes vexans* populations of New Orleans during 2005 and 2006

Parish and year	Jefferson 05	St. Bernard 05	Orleans 05	Orleans 06	Jefferson 06
Jefferson 05	–				
St. Bernard 05	–0.021*	–			
Orleans 05	–0.006*	–0.003*	–		
Orleans 06	–0.001*	–0.010*	–0.001*	–	
Jefferson 06	0.004*	–0.009*	–0.001*	–0.009*	–

* Pairwise F_{ST} values not significant ($P = 0.05$)

Table 4 Number of migrants among *Aedes vexans* populations of New Orleans during 2005 and 2006

Source of N_m formula	2005			2006
	Orleans and St. Bernard	Orleans and Jefferson	Jefferson and St. Bernard	Orleans and Jefferson
Nei (1973)	32.3	–350.1	106.2	82.7
Nei (1982)	29.1	91.5	23.5	27.9
Lynch and Crease (1990)	–129.2	–42.3	–28.8	46.1
Hudson et al. (1992)	–125.7	–42.3	–28.8	47.4

among parishes within years only -0.81% ($df = 3$, $F_{SC} = -0.008$, $P = 0.85$). A Mantel test revealed no isolation by distance.

Population Expansion or Growth

The mismatch distribution of frequencies of pairwise nucleotide differences showed a unimodal curve, which is characteristic of population expansion (Fig. 3). Site frequency spectrum revealed an excess of unique haplotypes (Fig. 4). Harpending's raggedness statistic was not significant ($P > 0.05$) for any of the populations during 2005 and 2006. The values for the parishes in 2005 were 0.028 in Jefferson, 0.066 in Chalmette, and 0.018 in Orleans; in 2006, they were 0.025 in Orleans and 0.024 in Jefferson. Average numbers for Fu and Li's D^* (-6.93) and F^* (-5.81) and Fu's F (-146.1) were negative. Strobeck's S statistic values were high, and Tajima's D values were also negative and generally significant ($P = 0.05$).

Discussion

This is the first study that has examined the genetic variability of *Ae. vexans* in a limited geographic area (the city of New Orleans), over a two-year period. In addition to investigating genetic diversity at this scale, this study addressed a broader question: the impact that a major climatic catastrophe, such as Hurricane Katrina, can have on populations of *Ae. vexans*. Only one other study examined the

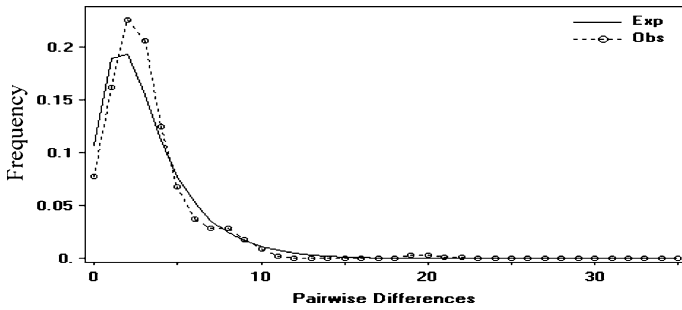


Fig. 3 Mismatch distribution of frequencies of pairwise number of differences among *Aedes vexans* New Orleans *ND5* haplotypes assuming population expansion. Unimodal curves are characteristic of populations that are in expansion (Michel et al. 2005)

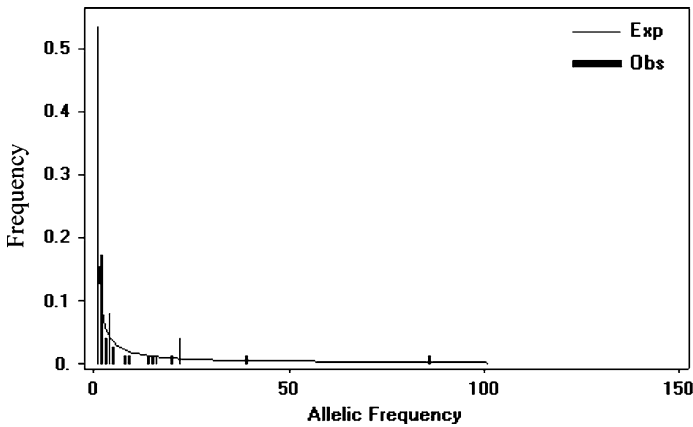


Fig. 4 Site frequency spectrum assuming constant population size indicates an excess of unique *Aedes vexans* New Orleans *ND5* haplotypes

genetic diversity of *Ae. vexans* and targeted populations from the central United States (Szalanski et al. 2006). Although genetic structure and variability have been studied for other mosquitoes of public health importance in small geographic areas, this is one of a few to study the population genetics of a floodwater mosquito species. The only other study accomplished on a floodwater species focused on *An. albifasciatus* Macquart in Argentina (de Souza et al. 1997).

We chose *ND5* mitochondrial sequence data for this study because it has proven to be an informative mtDNA marker for *Ae. vexans* (Szalanski et al. 2006). This marker, or portions of it, has also been widely used in *An. gambiae*, *An. funestus*, *An. arabiensis*, and *An. albopictus* (Besansky et al. 1997; Birungi and Munstermann 2002; Michel et al. 2005). Other NADH regions have been used for population genetics studies; *ND4* mtDNA data have been used for *Cx. tarsalis* (Venkatesan et al. 2007). Mitochondrial DNA coding gene markers are highly informative for population genetics studies because of their greater variability and rate of mutation, compared with DNA sequence data from nuclear genes. Recently, studies relying on

microsatellite loci have become increasingly popular. Microsatellites are short, repetitive sequences of DNA that are highly polymorphic, codominant, and neutral (Costa-Ribeiro et al. 2006). Future genetic studies on *Ae. vexans* using microsatellite markers would be informative for this species; however, no microsatellite markers currently exist for *Ae. vexans*.

In our study, observed haplotype diversity was high, ranging from 0.887 to 0.975, with a large number of unique haplotypes. Besansky et al. (1997) showed that *An. gambiae* and *An. arabiensis* also exhibited high levels of haplotypic diversity and a high number of unique haplotypes, based on mtDNA *cyt b*, *ND1*, and *ND5* sequence data. High haplotype diversity is generally evidence that a species is native, or it has been established for a long period of time (Hartl and Clark 1997). Birungi and Munstermann (2002) detected only nine haplotypes from almost 1,000 *An. albopictus*; this low haplotype diversity resembles a genetic bottleneck characteristic of introduced populations. Our mean number of pairwise nucleotide differences was 3.22 for all populations combined. In comparison, Besansky et al. (1997) found that *An. gambiae* or *An. arabiensis* sequences also did not differ by more than seven nucleotides.

We found noncorrespondence of haplotypes with geography, according to the genealogical parsimony network. In our study, we did not see the starlike pattern relationship that would indicate a close relationship among haplotypes. Genealogical relationships among haplotypes that were determined with TCS indicated that haplotypes 1 and 4 are the ancestral haplotypes, and the other haplotypes originated from these two. The genealogical tree for *Ae. vexans* from Kansas and neighboring states also presented haplotypes 4 and 1 as the ancestral haplotypes (Szalanski et al. 2006). Therefore, there is evidence of a shared ancestral origin of New Orleans populations with populations from the central United States. This shared origin concurs with active dispersal of this mosquito and strong flight capacity.

Dispersal capacity and genetic differentiation are related. According to previous studies, *Ae. vexans* can travel 10–24 km (Briegel et al. 2001). Our data also support a hypothesis of extensive traveling capacity, based on the low genetic differentiation seen between locations. Pairwise F_{ST} values were low and not significant from each other. In contrast, Gorrochotegui-Escalante et al. (2000) showed that there was genetic structuring and significantly different F_{ST} values when comparing populations of *Ae. aegypti* from the northeastern region of Mexico. *Aedes aegypti*, a small container breeder, is more likely to show genetic differences among populations even in relatively near geographic areas, because it does not need to travel long distances to find suitable sites for reproduction.

When F_{ST} values and geographic distances were correlated, no pattern of isolation by distance was found across the three parishes of New Orleans during both years, and there was no clustering of haplotypes by parish. Therefore, the results suggest there was no geographic barrier to movement of mosquitoes. Mosquito dispersal seems to be high enough to restrain genetic differences that could happen at the local level. The collection sites for Jefferson and Orleans parishes were 7.66 km apart, and the largest distance (between Jefferson and Chalmette) was 18.44 km. In addition, a relatively small population size might have influenced the nondetection of isolation by distance, as mentioned by Besansky et al. (1997).

Our study revealed no genetic structure in the populations from New Orleans. Variation was completely explained by populations within parishes (100%), which means variation was due to individual specimens. This pattern should be tightly linked with the great dispersal capacity of this mosquito. In contrast, there was a significant geographic structuring among the larger geographic scale sampled of populations from Kansas, South Dakota, Louisiana, and Texas (Szalanski et al. 2006). In our study, we found no evidence that Hurricane Katrina caused a genetic bottleneck; instead, we found high levels of genetic diversity.

Most population genetics studies take into account one-year sequence data, and generally geographic differences among populations are studied. Our study and only two other published studies have looked at temporal genetic variation in mosquito species. The first compared *Aedes aegypti* populations from Ho Chi Minh City in Vietnam and its suburbs, resulting in isozyme F_{ST} values that were 3 to 14 times higher in the dry season than in the rainy season (Huber et al. 2002). A second study looked at the genetic structure of *An. arabiensis* from southern Zambia, where no bottleneck effect was seen despite cycles of wet-drought-wet years (Kent et al. 2007).

Our study revealed high levels of gene flow. When gene flow is high, populations are said to be panmictic or randomly mating. In contrast, when gene flow is low, populations are said to undergo genetic drift. Our N_m values were high, which was evidence of gene flow between parishes.

When neutral mutation was tested, all values of Tajima's D were negative, and most of them were significant ($P < 0.05$). Negative values can reveal population expansion or selection. This statistic should be interpreted carefully, and to confirm that these values represent population expansion and not selection, the mismatch distribution should be taken into consideration. In *Ae. vexans* populations from five states in the central United States, Tajima's D and Fu and Li's parameters were significantly negative; indicating that a recent population expansion or growth had taken place (Szalanski et al. 2006).

We also observed a unimodal curve for the mismatch distribution of frequencies of pairwise nucleotide differences. Unimodal curves are characteristic of populations that are in expansion (Michel et al. 2005). The mismatch distribution is more likely to be smooth in an expanded population, because mutations are likely to be recent, causing segregating sites to occur predominantly in external branches of a genealogy. Segregating sites and corresponding sequence haplotypes are more likely singletons (Michel et al. 2005).

In our study, we found that Hurricane Katrina did not result in a change of genetic diversity of *Ae. vexans* from New Orleans. *Aedes vexans* breeds in freshwater temporary pools, retention ponds, woodland pools, and flooded fields (Andreadis et al. 2005). Mosquitoes from these areas outside the city would have flown to the city after water receded; therefore, no bottleneck effect was seen. Additional support for a lack of a genetic bottleneck in *Ae. vexans* is that the number of cases of West Nile virus in New Orleans increased after the hurricane (Caillouet et al. 2008).

Further studies should look into the genetic variability of *Ae. vexans* from different countries and continents. In addition, population genetics studies could be

coupled with vector competence studies (e.g., West Nile virus) of specimens from diverse geographic areas, because of its potential risk as a vector of West Nile and Rift Valley fever viruses.

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