West Nile virus activity in Chatham County, Georgia during 2011 by Robert A Moulis, Henry B Lewandowski, Jr, Jennifer D Russell, Jeffrey L Heusel, Laura F A W Peaty, Daniel G Mead and Rosmarie Kelly

West Nile virus (WNV) was first detected in Chatham County, Georgia from dead birds during the 2002 mosquito season. In all, 23 wild birds, 9 mosquito pools, and a horse were found positive for the virus that year. In 2003 a total of 27 wild birds, 6 sentinel chickens, 67 mosquito pools, and a horse were found positive for WNV. Additionally, nine human cases were diagnosed in 2003, including one fatality. WNV was also confirmed in Chatham County during 2004, 2006, and 2007. WNV was not detected in dead birds, mosquito pools, or humans between 2008 and 2010. Two sentinel chickens initially tested positive for the virus in 2009, but follow-up tests were negative. In 2011 WNV was recorded in 214 mosquito pools from 18 different sites, and 10 human cases were confirmed within our service area; see Table 1.

Prior to the arrival of WNV, Chatham County Mosquito Control (CCMC) conducted surveillance and control efforts primarily for nuisance mosquitoes (Aedes albopictus, Ae sollicitans and Ae taeniorhynchus) and vectors

of eastern equine encephalitis (EEE) (Culiseta melanura and Coquilletidia perturbans). However, during the 2002 season it became apparent that the primary vector of WNV in our region was Culex quinquefasciatus, a species of mosquito that was not targeted by our surveillance or control efforts. After the 2002 season, CCMC staff began a series of program modifications to augment our response to this newly emerging disease threat. Many of these program changes have previously been documented (Lewandowski and Moulis, 2008), although our approach is continuously reevaluated and refined as staff learn more about WNV and its ecology within our geographic region.

Most important to our WNV surveillance program is the use of gravid traps throughout the county to better assess *Cx quinquefasciatus* populations. Prior to the WNV threat, traps used by CCMC consisted solely of CDC light traps. A substantially larger number of *Cx quinquefasciatus* adults were available for arboviral testing at the University of Georgia's Southeastern Cooperative Wildlife Disease Study by using gravid traps. We have found that this testing is one of the best tools available for the early detection of virus. We further devised a system of thresholds based on the raw numbers of *Culex* captured in traps, which allowed us to treat areas prior to or early in the amplification stages of the WNV epizootic cycle in advance of laboratory confirmation of virus.

Secondly, we moved away from around ULV adulticide missions toward aerial applications, to provide "blanket" coverage of relatively large tracts of land in a very short time. We also began conducting missions closer to sunset, aligning with the peak activity of the local Cxquinquefasciatus population as indicated by timed collections in surveillance traps. Furthermore, we replaced malathion and permethrin-based products with naled adulticides, as susceptibility issues became apparent in our local Cx quinquefasciatus population. More recently (2011),

Comple Tune	Year									
Sample Type	2002	2003	2004	2005	2006	2007	2008	2009	2010	2011
Mosquito Pools	9	67	38	0	0	36	0	0	0	214
Wild Birds	23	27	0	0	0	1	0	0	0	0
Sentinel Chickens	0	6	0	0	0	0	0	2	0	0
Horses	1	1	0	0	0	0	0	0	0	0
Humans	0	9	1	0	1	0	0	0	0	10
Human Fatalities	0	1	0	0	0	0	0	0	0	0

Table 1: Occurrence of West Nile virus in Chatham County, GA 2002-2011.

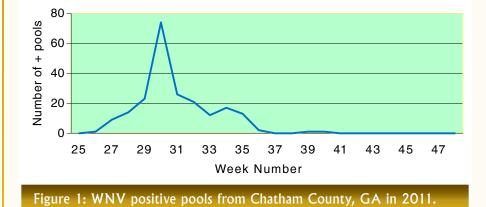
Year	Date of 1st pool collection	Date of last pool collection	Total pools	Blood-fed pools	Non- blood-fed pools	Total WNV positive pools	Collection date of 1st positive pool	Collection date of last positive pool
2001	09/25/01	12/06/01	137	8	129	0	N/A	N/A
2002	08/20/02	12/19/02	659	0	659	9	09/05/02	11/12/02
2003	04/01/03	11/17/03	2141	30	2111	67	07/11/03	09/23/03
2004	03/13/04	12/22/04	4144	502	3642	39	06/30/04	09/22/04
2005	01/04/05	10/24/05	6262	1085	5177	0	N/A	N/A
2006	06/19/06	12/28/06	2078	237	1841	0	N/A	N/A
2007	07/09/07	11/14/07	2981	149	2832	36	07/12/07	09/26/07
2008	03/26/08	11/17/08	3042	278	2764	0	N/A	N/A
2009	04/28/09	09/22/09	1010	38	972	0	N/A	N/A
2010	03/20/10	09/30/10	2123	132	1991	0	N/A	N/A
2011	02/22/11	11/14/11	3902	951	2951	214	06/20/11	09/27/11
Total	N/A	N/A	28479	3410	25069	365	N/A	N/A

Table 2: Summary of West Nile virus data from Chatham County, GA 2001-2011.

we moved away from fixedwing applications to an entirely rotary-winged adulticiding program, which enabled greater maneuverability and shortened application time.

We initiated an earnest storm drain larval treatment program that originally included catch basins located throughout the metro Savannah area, where the oldest infrastructure existed. This was expanded to include storm drains in the Savannah suburbs and surrounding municipalities as WNV was detected outside this core area. Originally, storm drains were treated with a 150 day product. This was eventually changed to a 30 day product to allow retreatment of storm drains on a monthly basis. From 2002 through early 2006 catch basins were treated with methoprene. In July of 2006 products containing *Bacillus sphaericus* (Bs) were used in catch basins, and we began alternating between Bs and methoprene products on a yearly basis. In August of 2011 when WNV positive pool numbers became staggeringly high, both products were used in storm drains until the end of October.

After the 2003 season we abandoned the use of sentinel chickens in our WNV surveillance program, as the turn-around time between confirmations from the laboratory on positive sentinels compared to that of positive mosquito pools was



approximately 3 weeks longer. However, we continued to use sentinels in our eastern equine encephalitis (EEE) surveillance program, as it is much more difficult to capture the primary vector (*Culiseta melanura*) of this virus in our area, and merely testing mosquitoes for EEE would not suffice.

CCMC recorded a total of 214 positive WNV mosquito samples during 2011. The first positive pool detected that year was collected on 20 June (week 26) which is 10 days earlier than any previous positive detection; see Table 2. The last positive pool was from a sample collected on 27 September (week 39). The number of WNV positive mosquito pools climbed quickly in 2011, peaking during week 30 (July 17-23) before gradually subsiding over the next several weeks; see Figure 1.

Positive pools were primarily recorded from trap sites located within the metro Savannah area (88%), although some virus was detected in suburban areas of Savannah, Tybee Island, Garden City, and Pooler. In addition, one positive pool was recorded from a rural area located in Jasper



Figure 2: Species make-up of WNV positive pools in 2011.

County, South Carolina. With the exception of two samples, all positive pools from 2011 were comprised of Culex species (x=97) or Cx quinquefasciatus (x=115) specimens; see Figure 2. One pool containing 7 Aedes albopictus and one pool containing a single blood-fed Aedes taeniorhynchus were also recorded. All positive mosquitoes collected in 2011 were captured in gravid traps, with the exception of the single salt marsh mosquito which was caught in a CDC light trap baited with dry ice.

Of the 18 sites where positive mosquito pools were collected during the 2011 season, the number of positive pools obtained through the course of the season varied. However, the majority of these sites (61%) recorded between one and five positive pools during the season. Two sites continued to produce positive samples over several weeks and tallied season totals of 36 and 79 positive pools. Weekly Minimum Infection Rates (MIR) at these locations were fairly high (4.78-38.81 and 9.09-60.61) throughout much of the summer; see Figure 3.

One site posted a MIR of 1000, and two other sites recorded

MIRs of 500. However, these inflated numbers are an obvious artifact of pooling only bloodfed mosquitoes from these sites, which included a single individual at the first site and two specimens at the other sites. One of these latter sites did produce more realistic MIRs in later weeks of 3.98 and 4.33, when all Culex collected from that site were analyzed for virus. It is noteworthy that of the 214 positive pools identified during 2011, eleven (5.1%) were composed of only a single blood-fed mosquito. Also of interest is that two WNV positive samples (one containing nine Culex species and the other 25 Cx quinquefasciatus) were simultaneously infected with Flanders virus.

In addition to some of the previously mentioned modifications we made to our program in 2011, we learned that a weekly treatment of at-risk areas is probably not sufficient to adequately reduce Cx quinquefasciatus numbers during an extremely active WNV year. Often aerial spray treatments did not reflect reductions in trap collections for several days. This is most likely due to a combination of factors during any spray event. First, on any given evening, only portions of the adult Cx quinquefasciatus population are active during the settling process of the ULV mist, while resting or dormant individuals avoid the aerosol. During a majority of the mosquito season all aquatic stages in the life cycle of Cx quinquefasciatus (egg through pupa) are present, allowing recruitment in the population to remain fairly constant. Furthermore, if the underground storm water system plays a major role in the life cycle of this species, as we assume, adult stages targeted by our adulticide work may only be susceptible to sprays during relatively short periods of time when outside this protected environment.

It is also important to mention that resistance issues involving our local populations of *Culex quinquefasciatus* came to light

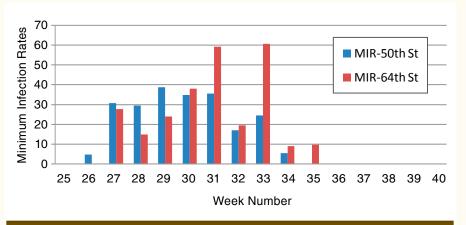


Figure 3: MIR by week at two metro Savannah locations in 2011.

when a series of susceptibility tests conducted by CDC indicated low susceptibility to many of the products available for mosquito control. No mortality was recorded at the diagnostic dosage ( $43\mu$ g/bottle) of permethrin after 30 minutes from specimens collected at four different sites in the county. Later tests indicated that resistance to resmethrin was 84% and 90% and to etofenprox was 94% and 88% at two of these sites, respectively. Testing of chloropyrifos and malathion on mosquitoes from these and one additional site showed little mortality even after two hours at the diagnostic dosage. Naled was the only chemical found to be effective on our local Cx quinquefasciatus (95% mortality at 15 minutes).

There are a number of possibilities that may explain the 2011 WNV resurgence in our area. First, our region experienced a fairly wet 2010-11 winter, but a hot and dry spring and summer. The winter rains provided ample water within the storm drain system that afforded local Cx quinquefasciatus populations ideal rookery conditions for egg deposition and larval development at the beginning of the season. The lack of rain during late spring and summer prevented storm drain systems from being flushed, resulting in large numbers of adult mosquitoes completing their larval stages between our current catch basin treatments, Furthermore, the local bird population had likely become susceptible to the virus during the previous 3-year WNV hiatus, despite a decline in the number of dead or dying birds reported by the public unlike in previous years; see Table 1.

In conclusion, our experiences with WNV have led to various changes in our approach to mosquito control in our region.

First, data collected through the last several years indicate that the primary vector of this virus in our region is Cx quinquefasciatus. The gravid trap is the superior device for collecting this species when used with adequately aged hay infusion. Naled is the only pesticide to which the local population of Cx quinquefasciatus is completely susceptible. It is also important to note that the testing and subsequent verification of virus in mosquito samples is paramount in the assessment of human risk in our area. Results from this work clearly revealed that the threat of WNV not only existed in 2011, but far exceeded any previous year on record. The total number of positive pools detected from Chatham County in 2011 was more than three times the amount seen in 2003 and five times the amounts in either 2004 or 2007. Overall, a total of 7622 mosquito pools were submitted for testing in the entire state of Georgia, and 397 of these were found positive for WNV in 2011. This represents an increase of approximately fourfold over the total number of positive pools detected in 2010, and indicates that the 2011 resurgence was not necessarily a localized problem. It further shows the importance of mosquito testing over a wide region is needed to adequately assess human health risk from one year to the next.

## ACKNOWLEDGMENTS

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