West Nile Virus Vector Larval Mosquito Monitoring Report - 2014

March 2015

Toronto and Region Conservation for The Living City®

Acknowledgements

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Region of Peel

DURHAM

York Region

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Executive Summary

West Nile virus (WNV) is primarily a bird pathogen that first appeared in Ontario in 2001. Evidence suggests that two key mosquito species, *Culex pipiens* and *Culex restuans,* are primarily responsible for spreading the disease to humans in Ontario (Kilpatrick *et al*. 2005; Hamer *et al*. 2009). Toronto and Region Conservation Authority's (TRCA's) data show that *Culex pipiens*—an urban mosquito species, is the most important vector species within our jurisdiction due to their sheer number. Vector mosquitoes are species that are capable of carrying and transmitting WNV. Their population dynamics are influenced by biological and environmental factors; therefore, forecasting an outbreak is difficult. WNV management strategies undertaken collectively by the provincial and regional health agencies in Ontario focus on prevention through education and mosquito control measures.

The number of human WNV case fluctuates annually. In 2014, a total of 11 human cases were reported in Ontario. In the Greater Toronto Area (GTA), 3 human WNV cases were reported in the City of Toronto (Public Health Ontario, 2014).

The WNV Larval Mosquito Surveillance and Monitoring Program was established in 2003 as a measure of due diligence and at the request of TRCA's regional public health partners. The program has a three-pronged approach, which includes public education and communication, collaboration with regional public health units, and larval mosquito monitoring. The two objectives of the program are to:

1) Reduce WNV risk to residents and conservation area visitors
2) Protect wetlands

In 2014, these objectives were achieved by identifying WNV hotspots and taking appropriate intervention measures, through public education, and through collaboration with regional public health partners. Wetland habitats are traditionally considered mosquito-friendly habitats. However, monitoring data collected by TRCA since 2003 have shown that healthy-functioning wetlands generally do not support large vector mosquito populations. When a WNV vector mosquito hot spot is detected, appropriate control measures can be taken to eliminate mosquito larvae if warranted.

Larval mosquito monitoring was undertaken in 45 sites across TRCA jurisdiction from June 3 to August 21 in 2014. In total, 6956 mosquito larvae were collected, of which 6243 larvae were identified, including 5840 larvae from 39 wetlands and 403 larvae from 6 stormwater management ponds (SWMPs). The rest of larvae died prematurely during the rearing process, thus the numbers were not included in risk assessment or analyses. Although most mosquitoes were collected from wetlands, higher concentrations of vector mosquito larvae were collected in SWMPs. In wetlands, 40% of mosquito larvae collected were vectors; in SWMPs, vector mosquito larvae represented 46% of larvae collected.

In total, 13 mosquito species including 7 WNV vector species and 6 non-vector species were identified. The most widespread species was *Culex territans.* The two key vectors, *Culex pipiens* and *Culex restuans,* were only found at 12 (27%) and 8 (18%) sites respectively.

Six sites were identified as hot spots for vector species larvae in 2014: Grenadier Pond in High Park, Albion Hills Pond 2, Granger Wetland South Pond, Woodland Pond in Eglinton Flats, Goldfish Pond in Tommy Thompson Park, and an unnamed wetland in Vaughan. TRCA‘s regional public health unit partners provide assistance in treating these hotspots.

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1. Introduction

This report provides an overview of activities conducted by The Toronto and Region Conservation Authority (TRCA) through its West Nile virus (WNV) vector larval mosquito monitoring program in 2014. WNV primarily exists between birds and bird-biting mosquitoes. The virus transmits to humans through the bite of an infected mosquito which had fed on infected birds. Humans are considered dead-end hosts whereby people can be infected with the virus, but do not spread it. The majority of people who become infected with WNV will have no symptoms or only mild flu-like symptoms. Severe cases of WNV, including the development of meningitis and encephalitis, are extremely rare but can be fatal.

Mosquito species that are capable of carrying and transmitting WNV are referred to as **vector** species. The species that do not transmit the virus are **non-vector** species. There are 57 mosquito species in Ontario, of which only 13 species are WNV vectors. Studies (Kilpatrick *et al*. 2005; Hamer *et al*. 2009) suggested that *Culex pipiens* and *Culex restuans* are not only the primary species in spreading the disease among birds, but also the primary species that spread the disease into the human populations. Most other mosquito species do not pose serious WNV threats and their larvae are important food sources for fish and other predatory aquatic organisms.

TRCA owns over 17,000 hectares of land, including natural and constructed wetlands, woodland pools, reservoirs, and ponds. These aquatic ecosystems have been considered "mosquito friendly" as a result of the permanent availability of standing water (Knight *et al*. 2003; Gingrich *et al*. 2006; Rey *et al*. 2006). The WNV Surveillance and Monitoring Program was initiated in 2003 as a measure of due diligence, and at the request of TRCA's Regional Public Health partners (Regions of Peel, York, Durham and the City of Toronto). Selected natural habitats (collectively referred to as "wetlands" in this report) and stormwater management ponds (SWMPs) have been monitored for the presence of mosquito larvae in the summer since the launch of the program. Data collected have been used to identify sites of potential concern or vector mosquito "hot spots", which may require follow-up with appropriate management actions.

The objectives of the WNV Vector Mosquito Larval Monitoring and Surveillance Program are to reduce WNV risk and protect wetlands on TRCA properties through the following approaches:

- **Public Education and Communication:** to respond to public inquiries on WNV related issues and address standing water complaints

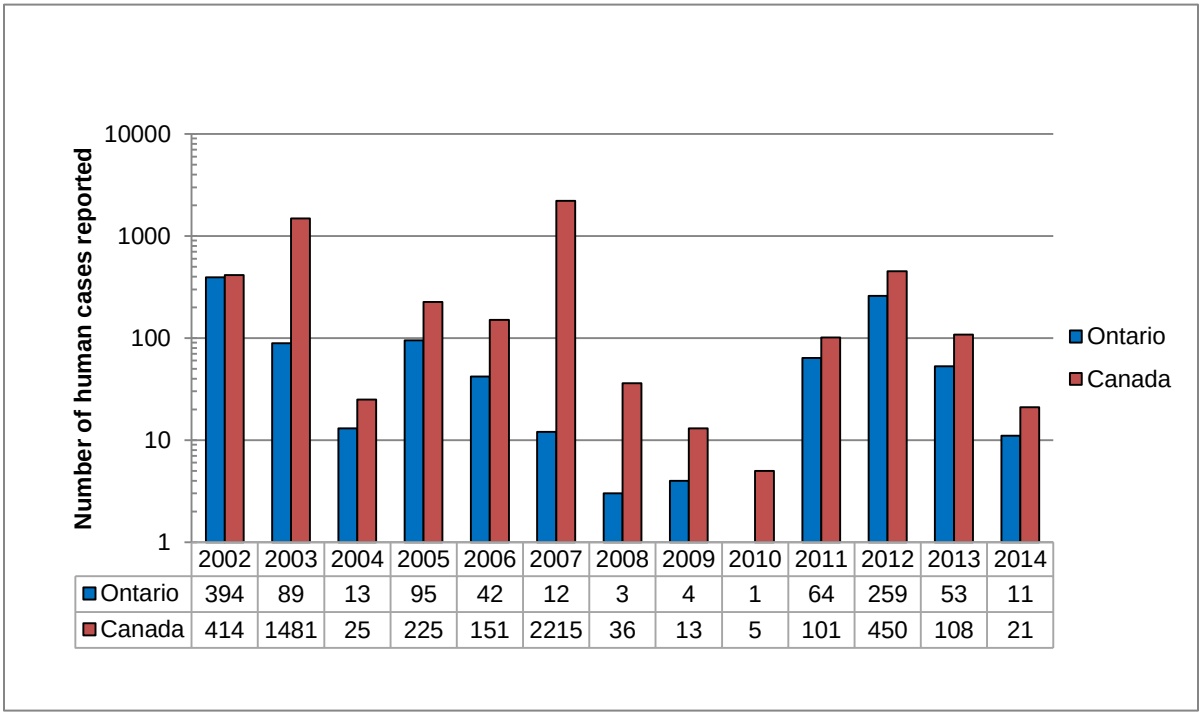
- **Collaboration with Regional Health Units:** to participate in WNV advisory committees and share WNV related information and data

- **Monitoring and Surveillance:** to identify sites of WNV concern through larval mosquito monitoring, and take appropriate control measures if deemed necessary.

In Canada, the number of human WNV cases fluctuates annually, driven by complex environmental and biological factors (Figure 1). In 2014, a total of 21 human cases were reported from three provinces: Ontario – 11, Quebec – 5, and Manitoba – 5 (Public Health Agency of Canada, 2014). Within TRCA's jurisdiction, three human WNV cases were reported; all occurred in the City of Toronto (Public Health Ontario, 2014). In 2014, Ontario's provincial and regional health agencies continued to monitor numbers of adult mosquitoes, larval mosquitoes, and human cases as part of the WNV surveillance programs. Adult mosquitoes monitoring is crucial for determining the immediate risk of humans contracting WNV. Larval mosquito surveillance provided information allowing Regional Public Health Units to eliminate/reduce mosquito larvae through larvicide application. Human surveillance information is used in a number of important ways. Knowing that West Nile virus is in an area puts doctors and the general public on special alert. It also provides more clues about who may be at highest risk for serious health effects from West Nile virus.

When West Nile virus first came to North America, bird surveillance was used as an early indicator of the presence of the virus in animals. Experience from past outbreaks showed that crows, jays, magpies and ravens were highly susceptible to West Nile virus. Infected dead birds are a good indicator to determine whether people in particular areas are at risk. The Province of Ontario no longer conduct dead bird surveillance. Instead, The Canadian Wildlife Health Cooperative tests dead birds for West Nile virus in collaboration with provincial laboratories and The National Microbiology Laboratory in Winnipeg.

Figure 1. Human West Nile virus cases in Ontario and Canada, 2002 - 2014



	2002	2003	2004	2005	2006	2007	2008	2009	2010	2011	2012	2013	2014
Ontario	394	89	13	95	42	12	3	4	1	64	259	53	11
Canada	414	1481	25	225	151	2215	36	13	5	101	450	108	21

2. Public Education and Communication

One of TRCA's WNV management approaches is to focus on prevention through increasing public awareness and to deal with standing water concerns on TRCA properties.

2.1 Increasing public awareness of West Nile virus

In 2014, TRCA continued to increase public awareness of WNV by:

- sharing tips on personal protection against mosquito bites, reminding the public to perform good housekeeping practices, and making the latest WNV program annual reports available on TRCA website **(**http://www.trca.on.ca/protect/monitoring/west-nile-virus-monitoring-program.dot).
- Providing up-to-date WNV related information on TRCA website.
- Providing WNV monitoring program updates in CreekTime e-Newsletter.
- reminding staff the importance of personal protection against WNV, and providing the latest WNV monitoring program and regional WNV status.
- displaying posters with WNV information in TRCA offices and Conservation Areas.

2.2 Standing Water Complaints

2.2.1 Standing Water Complaint Procedure

Complaints or enquiries from the public or staff regarding standing water or mosquito activities were addressed according to TRCA's Standing Water Complaint Procedure (Appendix A). The procedure includes the following steps:

1. Acquire background information (location, name of the complainant, contact information, and the nature of the complaint).

2. Evaluate the location for its proximity to a routine WNV sampling station, and the sensitivity of the area.

3. TRCA's Finance and Business Services Division and Planning and Development Division are consulted to review property ownership, management agreements and land regulation information.

4. For non-TRCA property or property under management agreement, the respective regional public unit is notified. For TRCA properties, if deemed necessary, monitoring activity following the methods described in Section 4.1: mosquito larval collection and identification and WNV risk assessment is undertaken.

5. When a potential hotspot is identified, and if larviciding is deemed appropriate, the following agencies are notified:
   - Respective regional public health unit.
   - Manager and Director at TRCA – for approval to proceed with the larvicide treatments.
   - The Ministry of the Environment and Climate Change (MOECC) – to obtain the permit for larviciding.
   - The Ministry of Natural Resources and Forestry (MNRF) – to review the sensitivity of the area.

6. Notify the complainant with the results of the investigation.

2.2.2 Standing Water Complaint Sites

In 2014, TRCA monitored three sites with standing water complaints. None of these sites was identified as a WNV hotspot. Most of mosquitoes collected at these three sites were non-vector mosquitoes.

3. Collaboration with Regional Health Units

The collaboration efforts with our regional public health partners involved notification of vector mosquito hot spots, and participation in advisory committees. TRCA also provided larval mosquito identification training to Durham, Halton, and the City of Hamilton Public Health staff. The participants of the training workshop learned to identify mosquito larvae commonly found in Southern Ontario (Figure 2). In addition, TRCA also received an Order from the Peel Region Medical Officer under the *Health Protection and Promotion Act*, R.S.O. 1990, c. H.7 to facilitate all mosquito reduction activities within the Heart Lake Wetland Complex in Brampton.

Figure 2. Mosquito larval identification workshop hosted by TRCA, 2014

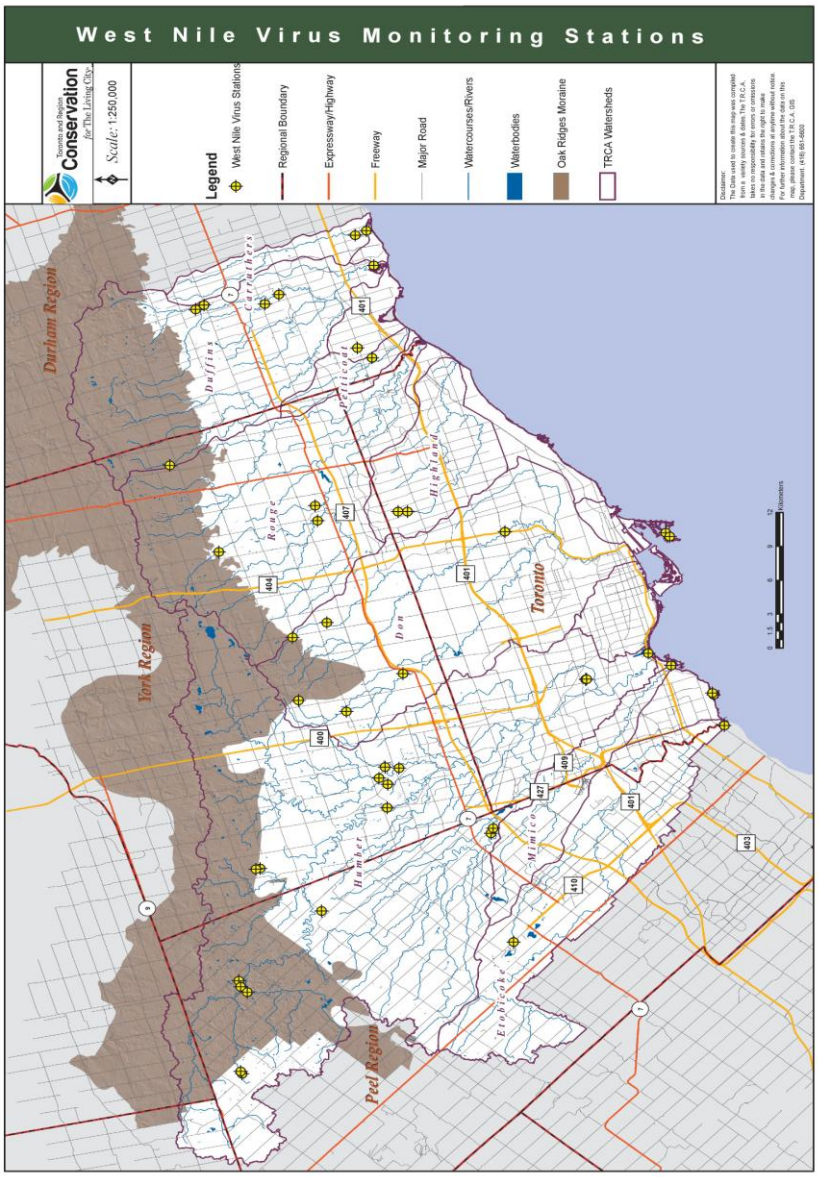
4. Larval Mosquito Monitoring

4.1 Methods

4.1.1 Monitoring Site Locations

The 2014 larval mosquito monitoring program began on June 3, and it covered 39 wetlands and 6 SWMPs across TRCA's jurisdiction (Figure 3). The monitoring stations remained unchanged from 2013.

Figure 3. Location of West Nile virus monitoring sites, 2014



4.1.2 Collection and identification

Each monitoring station was sampled five times in approximately two- week intervals between June and August with a standard mosquito dipper (diameter = 13 cm). Each site was divided into four comparatively equal quadrants, and one sample was taken within each quadrant. Each sample consisted of 10 dips of the mosquito dipper. During sampling, field technicians used several dipping techniques to ensure that all types of potential mosquito habitats were sampled (Figure 4a). Samples were not collected during a rain event because raindrops disturb the water surface and consequently cause mosquito larvae to disperse (O'Malley, 1995).

Collected mosquito larvae were taken back to the lab (Figure 4b), enumerated, and reared in rearing chambers until they reached maturity (fourth instar stage). The larvae were then preserved in 70% ethyl alcohol. Mosquito larvae were identified to species under a dissecting microscope using mosquito taxonomic keys (Wood *et al.,* 1979; Darsie and Ward, 2005). Those larvae that died before reaching maturity were not identified.

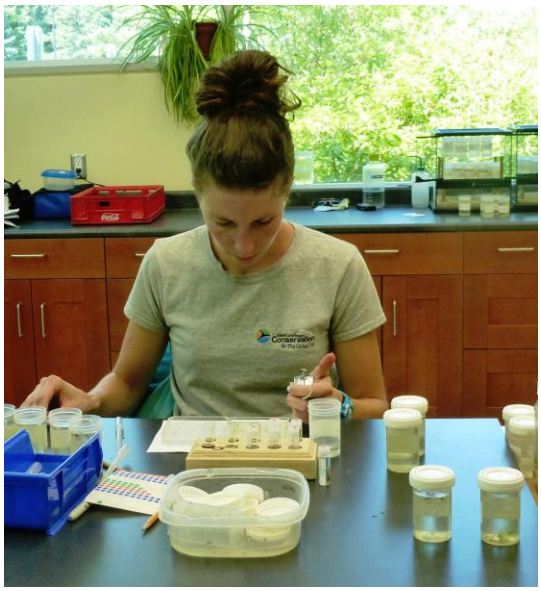


Figure 4. a) (left) Field technician sampling with a standard mosquito dipper b) (below) Mosquito Larvae were brought back to the lab for rearing and identification.

4.1.3 WNV Risk Assessment

WNV risk ranking was assessed for each site based on the number of vector larvae found in a sample after each site visit, according to the modified Wada's method of ranking (Wada, 1956):

- Sites with no vector larvae were ranked as "***Nil***" risk;
- Sites with <2 vector larvae per 10 dips were ranked as "***Low***" risk;
- Sites with 2 - 30 vector larvae per 10 dips were ranked as "***Moderate***" risk;
- Sites with >31 vector larvae per 10 dips were ranked as "***High***" risk sites.

Risk ranking was applied to each vector species independently, instead of the cumulative number of vector larvae found due to species variation in WNV transmission abilities.

Sites with "high" risk ranking or vector hot spots were addressed, the respective regional health unit was informed and if warranted, the sites were treated with larvicide.

Since mosquitoes only carry WNV after biting an infected bird, mosquito larvae do not need blood meals thus do not carry the virus. When a site is ranked as high-risk, it does not imply that the virus is present and poses immediate threat to the public. The risk ranking merely indicates the presence of vector mosquito species which could potentially spread WNV to human populations after they emerge as adult mosquitoes, not the presence of the virus.

4.2 Results

4.2.1 Mosquito diversity

In total, 6956 mosquito larvae representing 13 species were collected in 2014 from 45 routine monitoring stations. Larval mortality during the rearing process remained low at 9%. Mosquito larvae that died prematurely were not identified to species, thus excluded from the analyses and risk assessment in the following sections. The identified larvae included 5840 larvae from wetlands and 403 larvae from SWMPs.

The species collected included six non-vector species (*Culex territans*, *Culiseta inornata, Culiseta morsitans, Psorophora ferox*, *Anopheles earlei*, and *Uranotaenia sapphirina)* and seven WNV vector species (*Aedes vexans*, *Anopheles punctipennis, Anopheles quadrimaculatus, Culex pipiens*, *Culex restuans*, *Culex salinarius, and Ochlerotatus trivittatus)*. The most widespread species was *Culex territans,* which inhabited 39 of the 45 (87%) monitoring sites. Two key WNV vectors, *Culex pipiens* and *Culex restuans,* were found at 12 (27%) and 8 (18%) of the sampled sites respectively. *Culex pipiens* occurred in fewer sites in 2014 compared to 2012 and 2013, in both years, *Culex pipiens* were found at 15 sampled sites. *Culex restuans* occurrence remained unchanged.

In 2014, TRCA recorded three *Culiseta morsitans* larvae from two sites for the first time. Even though *Culiseta morsitans* is not a WNV vector, it is closely related to *Culiseta melanura*, the

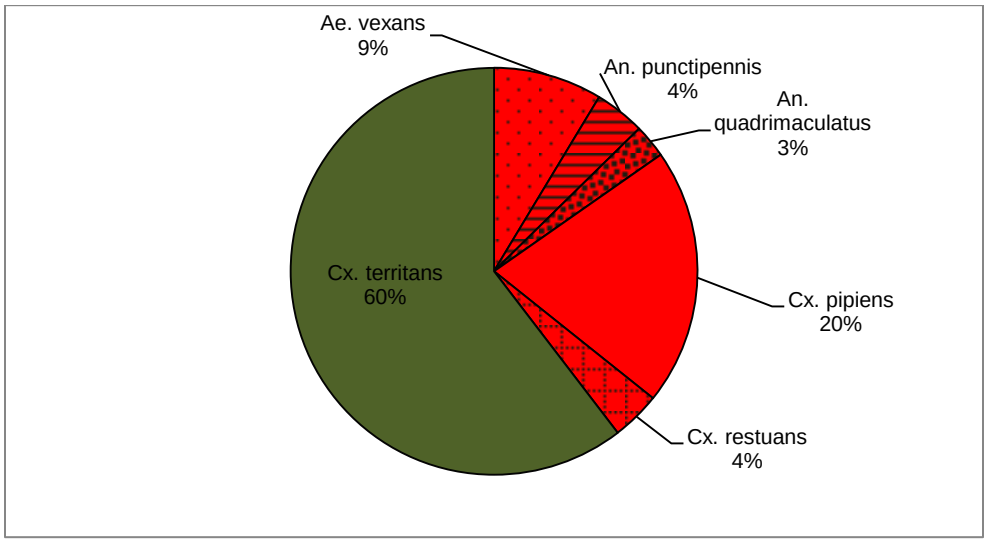
primary vector for Eastern equine encephalitis virus. Eastern equine encephalitis virus (EEEV), like WNV is a mosquito-borne virus. In North America, EEEV is restricted to areas east of the Mississippi River in the United States and southeastern Canada. The first human case occurred in Massachusetts in 1938, yet EEEV was initially recognized about a century earlier in horses. While EEEV activity has been noted in Canada, no human cases have been reported so far (Public Health Ontario, 2014). In the US, 41 human EEEV cases occurred from 2008 through 2013. Researchers (Molaei *et al*. 2006) suggested that *Culiseta morsitans* can be a potential vector for EEEV, therefore its presence will be closely monitored in 2015.

4.2.2 Wetlands

In total, 5840 mosquito larvae were identified to species. Similar to the findings from previous years, non-vector mosquito species, namely *Culex territans* dominated wetland habitats (Figure 5). Thirteen mosquito species were collected in wetlands. The predominant non-vector species was *Culex territans* (60%), and the predominant vector species was *Culex pipiens* (20%) *(*Figure 5). As in previous years, higher mosquito diversity was observed in wetlands compared to SWMPs. This finding is probably attributed to the facts that more wetland sites were sampled, and wetlands generally provide more diverse habitats and shelter.

Figure 5. Mosquito species composition in wetlands in 2014.

(non-vector species are indicated in green and vector species are indicated in red)



Note: Other 7 uncommon species collectively represented less than 1% of the mosquito collected, therefore were excluded from the figure.

Monitoring results showed that most wetlands posed minimal risk for harbouring WNV vector mosquitoes, however isolated vector mosquito hot spots continued to occur. Environmentally friendly larvicide, *Bacillus thuringiensis israelensis* (Bti) was used to treat the hot spots identified. Bti is a bacterium found naturally in soils, and since 1982, it has been used successfully worldwide as a biological pest control agent to combat mosquitoes and black flies (Health

Canada 2011). The pest control contractor displayed signs to notify the public prior and during larvicide treatments. The six identified hot spots were: Grenadier Pond in High Park, Woodland Pond in Eglinton Flats, Goldfish Pond in Tommy Thompson Park, Albion Hills Pond 2, Granger Wetland South Pond at the TRCA Restoration Services Centre, and a floodplain in Vaughan. Possibly due to the cooler summer, these hot spots occurred later in the season compared to previous years (Figure 6). Full mosquito monitoring risk assessment results for each monitoring station can be found in Appendix B-1 to B-4.

Figure 6. Identified WNV Vector Mosquito Hot Spots, TRCA – 2014.

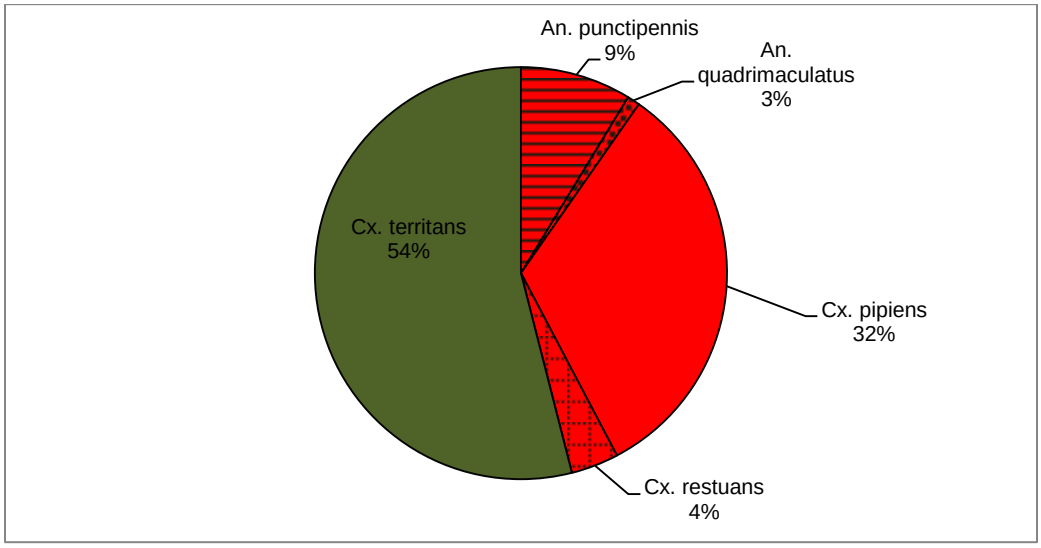
Site	Region	Vector species	Identified on
High Park – Grenadier Pond	Toronto	*Culex pipiens*	July 24, 2014
Eglinton Flats – Woodland Pond	Toronto	*Culex pipiens*	August 13, 2014
Tommy Thompson Park – Goldfish Pond	Toronto	*Culex pipiens*	August 11, 2014
Albion Hills Conservation Area – Pond 2	Peel	*Aedes Vexans*	July 31, 2014
Restoration Services Centre – Granger South Pond	York	*Culex pipiens*	July 29, 2014
Floodplain (un-named)	York	*Aedes vexans*	July 29, 2014

4.2.3 Stormwater Management Ponds

From the 6 SWMP monitoring sites, 403 mosquito larvae were identified, which consisted of 186 (46%) vector and 217 (54%) non-vector mosquito species larvae. The most abundant mosquito species was *Culex territans* (non-vector), and *Culex pipiens* only represented 32% of the larvae collected (Figure 7). In previous years, *Culex pipiens* had been the most abundant mosquito species in SWMPs, because L'Amoreaux Park North Pond had yielded large numbers of *Culex pipiens* consistently. In 2014, L'Amoreaux Park North Pond was not identified as a *Culex pipiens* hotspot. The City of Toronto Public Health staff also noticed a reduction in numbers of mosquito collected in their adult mosquito trap in the area. The reason for this reduction in *Culex pipiens* presence is not clearly known. None of the other sampled SWMPs were identified as hot spots for WNV vector mosquitoes this year.

Figure 7. Mosquito species composition in stormwater management ponds, 2014.

(non-vector species are indicated in green and vector species are indicated in red)

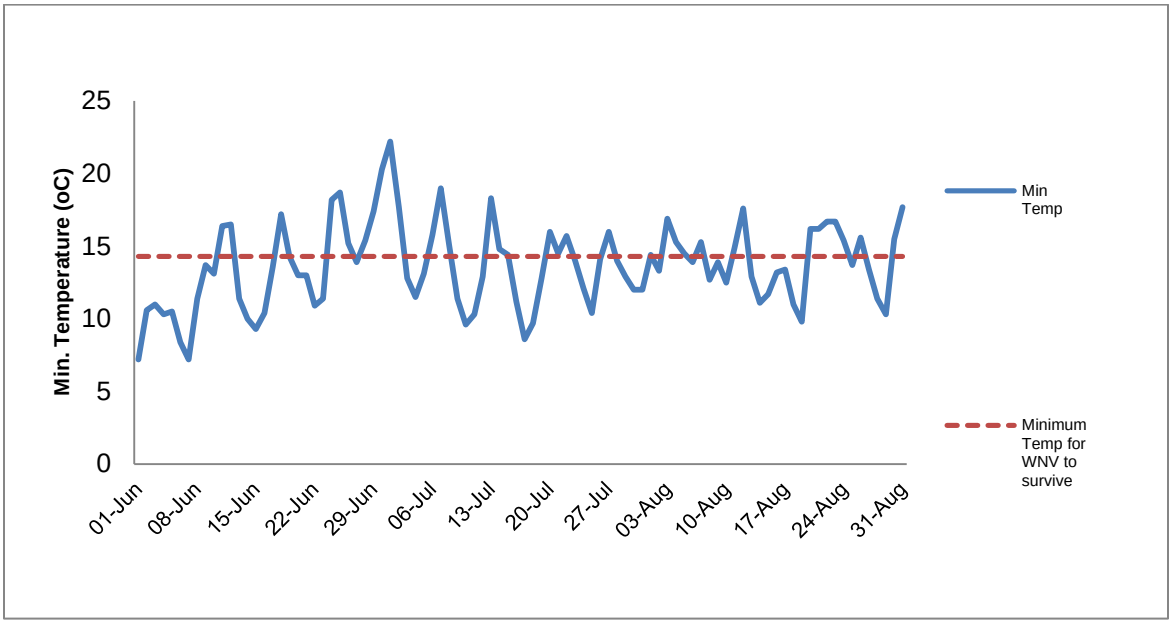


4.3 Temperature and its potential influence on the spread of WNV

In 2014, number of WNV human cases dropped significantly from 2012 and 2013 in Toronto and throughout Ontario. Fewer *Culex pipiens* larvae were collected in 2014 by TRCA, and regional public health units also reported a reduction in numbers of adult mosquitoes that tested positive for WNV. The results could potentially be linked to the lower than normal summer temperatures in 2014. Researchers at the Center for Vectorborne Disease at the University of California suggested that WNV needs temperatures to stay above 14.3 ^{o}C or it cannot survive. The cooler summer temperatures (Figure. 8) may play a key role in limiting the development and survival of WNV, leading to fewer human WNV cases in 2014.

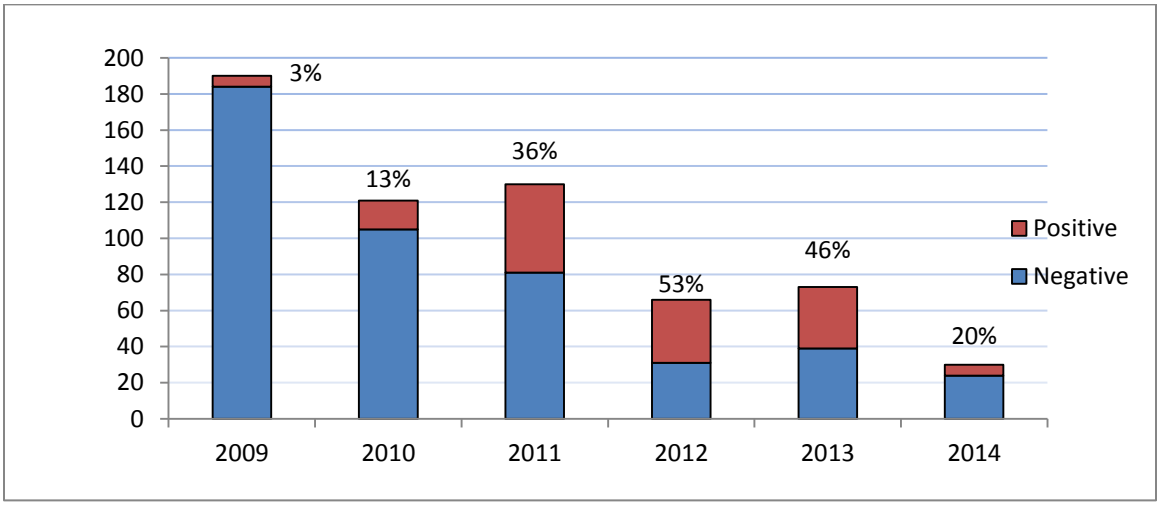
The Canadian Wildlife Health Cooperative coordinates national surveillance project to monitor WNV in dead birds across Canada, with approximately 300 birds per year being tested for WNV since 2009. Interestingly, dead bird surveillance data also showed that in 2014, lower percentage (20%) of birds were tested positive for WNV comparing 2012 (53%) and 2013 (46%) (Figure 9).

Figure 8. Minimum daily temperature, June – August 2014.



* Weather data source: Historical Climate Data. Environment Canada (http://climate.weather.gc.ca/)

Figure 9. Percentage of dead birds tested positive for WNV in Ontario, 2009-2014.



* Data source: The Canadian Wildlife Health Cooperative (http://www.cwhc-rcsf.ca/data_products_wnv.php)

5. Conclusions

The results from the 2014 program supported the findings from the previous TRCA studies. Generally, wetlands do not pose threats of WNV transmission due to the low percentage of vector larvae present.

Compared to 2012, a significant reduction in WNV infection rate in humans was observed in Toronto and throughout Ontario. The cooler summer temperature in 2014 might have not only slowed the development of *Culex pipiens* larvae and but also the virus itself. However, WNV vector hotspots continued to occur; six hot spots were detected and treated through TRCA's larval mosquito monitoring program. The ability to detect hot spots, and subsequently take appropriate control measures continue to highlight the importance of regular and continuous seasonal monitoring of wetlands and SWMPs. TRCA addressed two standing water concerns associated with TRCA properties as per TRCA's Standing Water Complaints Procedure.

Collaboration with Regional Public Health units is crucial in managing WNV vector hot spots in a timely manner on TRCA properties. In 2014, the City of Toronto Public Health, York Region Public Health, and Peel Region Public Health assisted TRCA in treating identified WNV hot spots. Jurisdictionally, *Culex pipiens* abundance were the highest in the City of Toronto compared to the Regions of Peel, Durham, and York. The number of WNV positive mosquito pools and the number of human cases showed the same trend. TRCA's data are valuable for regional public health partners to use as a tool in predicting the emergence of vector species adult mosquitoes and the WNV risk in the human population.

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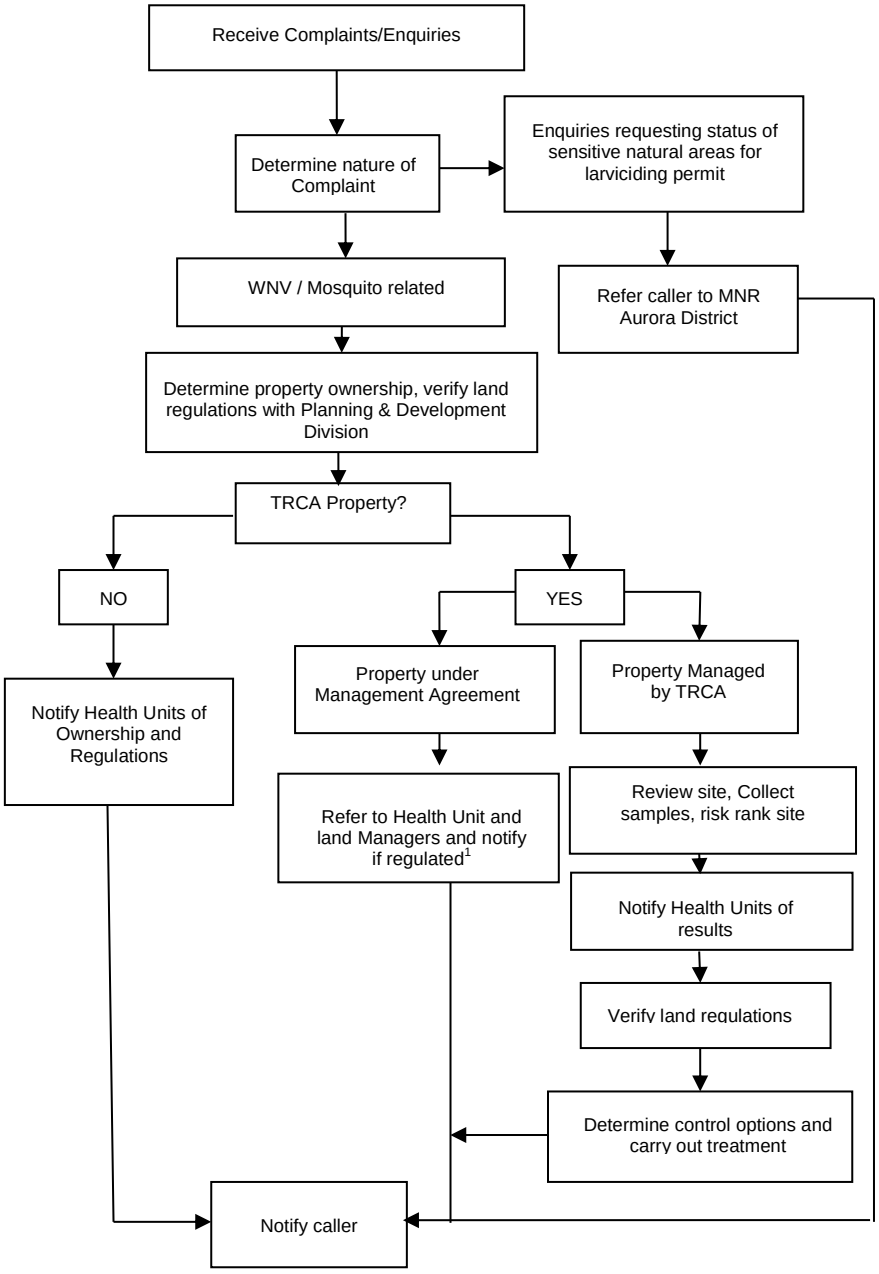
Appendices

Appendix A. TRCA Standing Water Complaint Procedure



Appendix B-1 Monitoring and Risk Assessment Results in Durham Region - 2014

Sites with no vector larvae were ranked as "***Nil***" risk; sites with <2 vector larvae per 10 dips were ranked as "***Low***" risk; sites with 2 - 30 vector larvae per 10 dips were ranked as "***Moderate***" risk; and sites with >31 vector larvae per 10 dips were ranked as "***High***" risk.

Site	Sampling Event	*Ae. vexans*	*An. punctipennis*	*An. quadrimaculatus*	*Cx. pipiens*	*Cx. restuans*	*Oc. trivittatus*
Altona Forest	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Low	Nil	Nil	Nil	Nil
	3	Nil	Low	Nil	Nil	Nil	Nil
	4	Nil	Low	Nil	Nil	Nil	Nil
	5	Low	Moderate	Nil	Nil	Nil	Nil
Carruthers Swamp Complex	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Low	Low	Nil	Nil	Nil	Nil
Claremont Wetland-1	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Low	Nil	Nil	Nil	Nil
	3	Nil	Low	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Nil	Low	Nil	Nil	Nil
Claremont Wetland-2	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Moderate	Low	Nil	Nil	Nil
	4	Nil	Nil	Moderate	Nil	Nil	Nil
	5	Nil	Nil	Moderate	Nil	Nil	Nil
Frenchman's Bay Promenade	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Nil	Nil	Nil	Nil	Nil
Greenwood Marsh	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Moderate	Nil	Nil	Nil	Nil	Nil
Greenwood Pond	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Nil	Nil	Nil	Nil	Nil
Lower Duffins	1	Nil	Nil	Nil	Nil	Moderate	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Nil	Nil	Nil	Nil	Nil

Appendix B-2 Monitoring and Risk Assessment Results in Peel Region - 2014

Sites with no vector larvae were ranked as "***Nil***" risk; sites with <2 vector larvae per 10 dips were ranked as "***Low***" risk; sites with 2 - 30 vector larvae per 10 dips were ranked as "***Moderate***" risk; and sites with >31 vector larvae per 10 dips were ranked as "***High***" risk.

Site	Sampling Event	*Ae. vexans*	*An. punctipennis*	*An. quadrimaculatus*	*Cx. pipiens*	*Cx. restuans*	*Oc. trivittatus*
Albion Hills Pond-1	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Nil	Nil	Nil	Nil	Nil
Albion Hills Pond-2	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Low	Nil	Nil	Nil	Nil	Nil
	4	High	Low	Nil	Nil	Nil	Nil
	5	Nil	Nil	Nil	Nil	Nil	Nil
Albion Hills Pond-4	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Low	Nil	Nil	Nil	Nil
Claireville Wetland-1	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Low	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Nil	Nil	Nil	Nil	Nil
Claireville Wetland-2	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Nil	Nil	Nil	Nil	Nil
Glen Haffy Trout Pond-1	1	Nil	Moderate	Nil	Nil	Nil	Nil
	2	Nil	Low	Nil	Nil	Nil	Nil
	3	Nil	Moderate	Nil	Nil	Nil	Nil
	4	Nil	Moderate	Low	Nil	Nil	Nil
	5	Nil	Moderate	Moderate	Nil	Nil	Nil
Glen Haffy Trout Pond-2	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Moderate	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Nil	Nil	Nil	Nil	Nil
Heart Lake	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Nil	Nil	Nil	Nil	Nil
Marie Curtis	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Low	Nil	Nil	Nil
	5	Nil	Nil	Low	Nil	Nil	Nil
SWMP-174	1	Nil	Moderate	Nil	Nil	Nil	Nil
	2	Nil	Low	Nil	Nil	Nil	Nil
	3	Nil	Moderate	Nil	Nil	Nil	Nil
	4	Nil	Low	Low	Nil	Nil	Nil
	5	Nil	Moderate	Nil	Nil	Nil	Nil

Appendix B-3 Monitoring and Risk Assessment Results in Toronto - 2014

Sites with no vector larvae were ranked as "***Nil***" risk; sites with <2 vector larvae per 10 dips were ranked as "***Low***" risk; sites with 2 - 30 vector larvae per 10 dips were ranked as "***Moderate***" risk; and sites with >31 vector larvae per 10 dips were ranked as "***High***" risk.

Site	Sampling Event	*Ae. vexans*	*An. punctipennis*	*An. quadrimaculatus*	*Cx. pipiens*	*Cx. restuans*	*Oc. trivittatus*
Col. Samuel Smith Main Pond	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Nil	Nil	Nil	Nil	Nil
Col. Samuel Smith Mini Pond	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Low	Low	Moderate	Nil	Nil
	5	Nil	Nil	Moderate	Moderate	Nil	Nil
High Park Grenadier Pond	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Moderate	Moderate	Nil
	3	Nil	Nil	Low	Moderate	Moderate	Nil
	4	Nil	Nil	Low	High	Moderate	Nil
	5	Nil	Nil	Moderate	High	Moderate	Nil
L'Amoreaux North Pond	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Moderate	Moderate	Nil
	4	Nil	Nil	Nil	Moderate	Moderate	Nil
	5	Nil	Nil	Nil	Nil	Nil	Nil
L'Amoreaux South Pond	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Nil	Nil	Nil	Nil	Nil
Milne Hollow	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Low	Low	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Nil	Nil	Nil	Nil	Nil
Mimico Amphibian Pond	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Nil	Nil	Nil	Nil	Nil
Topham Pond	1	Nil	Nil	Nil	Low	Low	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Moderate	Low	Moderate	Nil	Nil
TTP Goldfish Pond	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Moderate	Nil	Nil
	5	Nil	Nil	Nil	High	Nil	Nil
TTP Tri-Pond	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Nil	Nil	Nil	Nil	Nil
Woodland Pond	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Low	Nil	Nil
	3	Nil	Nil	Nil	Moderate	Nil	Nil
	4	Nil	Low	Nil	Moderate	Nil	Nil
	5	Nil	Nil	Nil	High	Nil	Nil

Appendix B-4 Monitoring and Risk Assessment Results in York Region - 2014

Sites with no vector larvae were ranked as "***Nil***" risk; sites with <2 vector larvae per 10 dips were ranked as "***Low***" risk; sites with 2 - 30 vector larvae per 10 dips were ranked as "***Moderate***" risk; and sites with >31 vector larvae per 10 dips were ranked as "***High***" risk.

Site	Sampling Event	*Ae. vexans*	*An. punctipennis*	*An. quadrimaculatus*	*Cx. pipiens*	*Cx. restuans*	*Oc. trivittatus*
Boyd Conservation Area	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Low	Nil	Nil	Nil	Nil
	5	Nil	Nil	Nil	Nil	Nil	Nil
Bruce's Mill	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Moderate	Moderate	Nil	Nil	Nil
	4	Low	Low	Moderate	Nil	Nil	Nil
	5	Nil	Nil	Moderate	Nil	Nil	Nil
Cold Creek Pond	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Nil	Nil	Nil	Nil	Nil
Cold Creek Wetland	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Low	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Low	Moderate	Nil	Nil	Nil
Earth Rangers	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Low	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Nil	Low	Nil	Nil	Nil
Granger Wetland South	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Low	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Low	High	Nil	Nil
	5	Nil	Moderate	Moderate	Nil	Nil	Nil
Keffer Marsh	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Nil	Nil	Nil	Nil	Nil
Killian Lamar	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Nil	Nil	Nil	Nil	Nil
Kortright Centre Marsh	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Low	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Low	Low	Nil	Nil	Nil

Appendix B-4 Monitoring and Risk Assessment Results in York Region – 2014 (Continued)

Sites with no vector larvae were ranked as "***Nil***" risk; sites with <2 vector larvae per 10 dips were ranked as "***Low***" risk; sites with 2 - 30 vector larvae per 10 dips were ranked as "***Moderate***" risk; and sites with >31 vector larvae per 10 dips were ranked as "***High***" risk.

Site	Sampling Event	*Ae. vexans*	*An. punctipennis*	*An. quadrimaculatus*	*Cx. pipiens*	*Cx. restuan s*	*Oc. trivittatus*
Stouffville Reservoir	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Nil	Nil	Nil	Nil	Nil
Toogood Pond	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Nil	Nil	Nil	Nil	Nil
Un-named wetland - Vaughan	1	Moderate	Nil	Nil	Nil	Nil	Nil
	2	Nil	Moderate	Nil	Nil	Nil	Nil
	3	Moderate	Low	Nil	Moderate	Low	Moderate
	4	High	Nil	Nil	Nil	Nil	Moderate
	5	Nil	Nil	Nil	Nil	Nil	Nil
Un-name Wetland 1	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Moderate	Nil	Nil	Nil	Nil	Nil
Un-named Wetland 2	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Low	Nil	Nil	Nil	Nil
	5	Nil	Nil	Nil	Nil	Nil	Nil
SWMP-88.2	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Nil	Nil	Nil	Nil	Nil
SWMP-139	1	Nil	Nil	Nil	Nil	Nil	Nil
	2	Nil	Nil	Nil	Nil	Nil	Nil
	3	Nil	Nil	Nil	Nil	Nil	Nil
	4	Nil	Nil	Nil	Nil	Nil	Nil
	5	Nil	Nil	Nil	Nil	Nil	Nil