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Executive Summary

The following report provides information about the transmission and health effects of the vector-borne diseases of concern in Peel, as well as details and results of surveillance and risk reduction activities undertaken in 2009.

In 2008, the Ontario Ministry of Health and Long-term Care (MOHLTC) reduced funding to health units by over twenty percent and changed its funding structure by expanding the West Nile Virus (WNV) program to include additional vector-borne diseases. Consequently, larval surveillance activities in Peel were postponed by one month and the WNV Prevention Plan was enhanced to include two other vector-borne diseases of concern in Ontario: Lyme disease and Eastern equine encephalitis (EEE). The WNV Prevention Plan changed its name to the Vector-Borne Disease (VBD) Prevention Plan.

Though WNV continues to feature prominently in this, the first Vector-Borne Disease Report, surveillance activities and results of Lyme disease and EEE will also be discussed briefly. This report follows the 2008 WNV Prevention Plan which was adopted by Peel Council.¹

Like in previous years, Peel Public Health in 2009 continued to monitor the risk of WNV transmission to humans through surveillance and to reduce it through control efforts and education.

Weather conditions in 2009 were similar to 2008 and consisted mainly of high precipitation and lower than normal temperatures. Under such conditions mosquito larvae are slow to mature and replicate, leading to reduced incidence of WNV. This was demonstrated through surveillance of mosquito populations in 2009, which showed a reduction in WNV activity over the previous year. There were no human cases of WNV in the Region of Peel in 2009.

Mosquito larval surveillance was undertaken at 2,073 potential breeding sites in Peel Region, a decrease from 2009 due to the delayed program start. Larval mosquito reduction activities involved the application of; four rounds of methoprene (Altosid®) pellets to 363,795 roadside catch basins; 2,314 methoprene briquets to non-roadside catch basins, private backyards and public parks; and 656 treatments to four hundred and five surface water sites.

Increased temperatures associated with climate change could increase the survival or replication rates of vector mosquitoes and may contribute to higher incidence of disease in the future. Climate changes could also expand the habitat and infectivity of other disease-carrying insects, increasing the potential for transmission of diseases such as Lyme disease and Eastern equine encephalitis in Peel Region. As a result, it is prudent to continue surveillance programs for these vector-borne diseases.

Introduction

The West Nile Virus (WNV) program went through some considerable changes in 2009. The Ontario government reduced the WNV budget allocated to health units by twenty-three per cent relative to 2007 levels and changed its funding structure by expanding the WNV program to include additional vector-borne diseases. As a result, Peel's larval surveillance activities were postponed by one month, starting in June instead of May, and the Region of Peel WNV Prevention Plan was enhanced to include Lyme disease and Eastern equine encephalitis (EEE). The Ontario Ministry of Health and Long-term Care (MOHLTC) also announced in 2008 that it would cease funding of the dead bird surveillance program, so Peel Public Health stopped collecting dead birds in 2009. These service changes impact the comparability of 2009 data to previous years.

A vector-borne disease is a disease that is transmitted to humans or other animals by an insect or other arthropod. In Ontario, WNV and Lyme disease are the two most common vector-borne diseases to transmit infection to humans. Due to concern that regional environmental change is making it more favorable for these vector populations to increase, there is mounting pressure on health units in Ontario to develop surveillance programs.

Another vector-borne disease of concern in Ontario is Eastern equine encephalitis (EEE). Although rarer, Eastern equine encephalitis is a serious viral disease spread by mosquitoes that has the potential to affect people and horses. The EEE virus has been detected in Ontario in horses and mosquitoes; so far no human cases have been reported.

Peel Public health initiated surveillance of Lyme disease and EEE and changed its name from West Nile Virus Surveillance program to the Vector-Borne Disease (VBD) Surveillance program.

Increased temperatures associated with climate change could increase the survival or replication rates of vector mosquitoes and may contribute to higher incidence of disease in the future. Climate changes could also expand the habitat and infectivity of other disease-carrying insects, increasing the potential for transmission of diseases such as Lyme disease and Eastern equine encephalitis in Peel Region. As a result, it is prudent to continue surveillance programs for these vector-borne diseases.

The following report provides information about the transmission and health effects of the vector-borne diseases of concern in Peel, as well as details and results of surveillance and risk reduction activities in 2009.

West Nile Virus

West Nile Virus (WNV), a virus transmitted primarily through the bite of infected female mosquitoes, was first detected in North America in 1999 when an outbreak was experienced in New York City. Since then, WNV has rapidly spread across all other continental U.S. states and the majority of Canada's provinces.

In early spring, the amplification of WNV begins after infected adult mosquitoes overwinter and/or infected migratory birds return to a region. *Culex pipiens* and *Culex restuans*, two mosquito species that feed primarily on birds, are the main vectors for the virus in Ontario and have been estimated to be responsible for up to 80% of WNV human infections in the north-eastern United States, an environment similar to Peel Region.²

Infected mosquitoes feed on birds and the virus is transmitted back and forth resulting in an increase in the number of birds and mosquitoes infected. Later on in the season, typically late July, there is a "spill over point" where the virus bridges out of the mosquito-bird cycle via bridge vectors. The bridges are mosquito species, like *Aedes vexans*, that feed on humans and other mammals in addition to birds.

The species type of WNV vector mosquito will vary with geography. For example, the species responsible for the 2007 increase in human cases in the Prairie Provinces and the central United States, *Culex tarsalis*, is not found in significant abundance in Ontario.

An estimated one in five people who are bitten by a mosquito infected with WNV will develop symptoms. Most people who are infected have either no symptoms or mild illness, such as West Nile fever. The incubation period is estimated to be three to 14 days with symptoms lasting on average three to six days. Cases are classified as West Nile Virus Neurological Syndrome (WNNS) or West Nile Virus Non-Neurological Syndrome (WN Non-NS).

WNV fever is described as a sudden onset of fever that is often accompanied by malaise, headache, nausea, vomiting, anorexia, eye pain, myalgia and less commonly, rash and/or swollen lymph nodes. This is typically classified as WN Non-NS.⁴

In about 1% of infected individuals, WNV can cause serious illness including severe neurological disease which is classified as WNNS. Additional symptoms among those with severe disease include muscle weakness and a change in mental status.⁴

Long-term health effects of WNV infection are possible but are less well understood. They can include physical (long-term muscle weakness and paralysis,

fatigue and headache), cognitive (depression, confusion, and memory loss) and functional effects (difficulty with meal preparation and shopping).⁴

In 2001, WNV was first detected in birds and mosquitoes in Peel Region. Locally acquired human illness of WNV first occurred in Peel Region in 2002. Twenty-one of 37 cases required hospitalization. In 2002, there were two deaths in Peel due to WNV infection, which have been the only deaths to occur in Peel due to this cause.

Ontario Regulation 199/03⁵ (Control of West Nile Virus), under the *Health Protection and Promotion Act*, requires that the local Medical Officer of Health (MOH) conduct a risk assessment of the conditions pertaining to WNV in their health unit. The risk assessment relies on surveillance of human and mosquito infections. This guides the MOH with respect to appropriate WNV risk reduction activities, including the need for mosquito reduction measures. Provincial regulation also requires the MOH to record, investigate and report any adverse or unintended human health effects attributed to mosquito reduction actions and to report any non-human environmental adverse effects to the Ministry of Environment and/or other relevant local or provincial authorities. WNV is both a reportable and communicable disease under Regulations 558/91⁶ and 559/91⁷, respectively, requiring physicians and laboratories to report human cases to the local MOH.

The goal of the WNV component of the Region of Peel's Vector Borne-Disease (VBD) Prevention Plan is to minimize the impact of WNV with a regional surveillance program involving humans and mosquitoes. The surveillance program guides the integrated pest management activities which include mosquito larvae reduction and prevention, and public education and community outreach activities.

Human Case Surveillance

The human case surveillance program for WNV is intended to detect illness in the population of Peel Region. All probable or confirmed cases identified by hospitals and physicians are reported to the local public health department. The Ontario Ministry of Health and Long-term Care (MOHLTC) has developed case definitions and diagnostic test criteria (refer to Appendix A).⁸

Peel Public Health staff investigate all reported probable and confirmed cases of WNV among Peel residents. Demographic and medical information including symptoms and risk factors (e.g. travel history, blood products recipient) are collected and entered into the integrated Public Health Information System (iPHIS).

Human Surveillance Program - 2009

In 2009, there were no confirmed human cases of WNV in Peel Region.

In 2002, 57 probable and confirmed cases of WNV were reported based on the case definitions at the time. However, the case definition has changed since 2002. If the present day definition was applied there would have been 18 confirmed human cases in 2002. The only WNV-related deaths in Peel occurred in 2002.

Comparison with Other Ontario Health Units

Across Ontario, there were three confirmed WNV human cases in 2009 compared to four in 2008 and 15 in 2007.⁹ Only two of the 36 health units reported human cases in 2009, Winsor-Essex and Middlesex-London.

Comparison with Other Provinces

Across Canada, there were a total of eleven human cases of WNV and no deaths in 2009 compared to 38 cases and no deaths in 2008, and 2,355 cases and two deaths in 2007. This was the lowest number of human cases reported in Canada since surveillance activities began.

The highest number of human cases of WNV was reported in British Columbia with three. It was the first time cases of WNV had been locally acquired in BC.

The United States saw a reduction in the number of human cases with 663 human cases and 30 deaths in 2009 compared to 1,370 cases and 37 deaths in 2008.

Adult Mosquito Surveillance

Mosquito surveillance programs serve to monitor the mosquito population both for their abundance and the species present. Certain species of mosquitoes are more likely to transmit WNV to humans. Therefore, it is important to monitor their occurrence in order to assess the potential human health risk. In Ontario, the species of particular interest due to their WNV transmission risk continues to be the *Culex* species.

Historically, *Culex* species have consistently been responsible for the majority of the WNV-positive mosquito batches in Peel Region. However, in 2009 only one of the four (25%) WNV-positive mosquito batches were from *Culex* species.

Other mosquito species are also known to carry WNV. So while reducing the abundance of the *Culex* species remains a priority for the Region of Peel, monitoring the abundance and WNV potential of various species remains very important.

For more information about the methodological details associated with species identification, sorting and viral testing, refer to the 2006 West Nile Virus in the Region of Peel report.¹⁰ available at:

<http://www.peelregion.ca/health/westnile/resources/reports.htm#control06>

Thirty-one fixed CDC light traps were distributed by Regional ward, with a minimum of one trap per ward across Peel: 17 in the City of Mississauga, 9 in the City of Brampton and five in the Town of Caledon. Map 1 shows the locations for the fixed traps set in Peel Region in 2009.

Map 1 Location of Mosquito Traps by Municipal Ward, Region of Peel, 2009

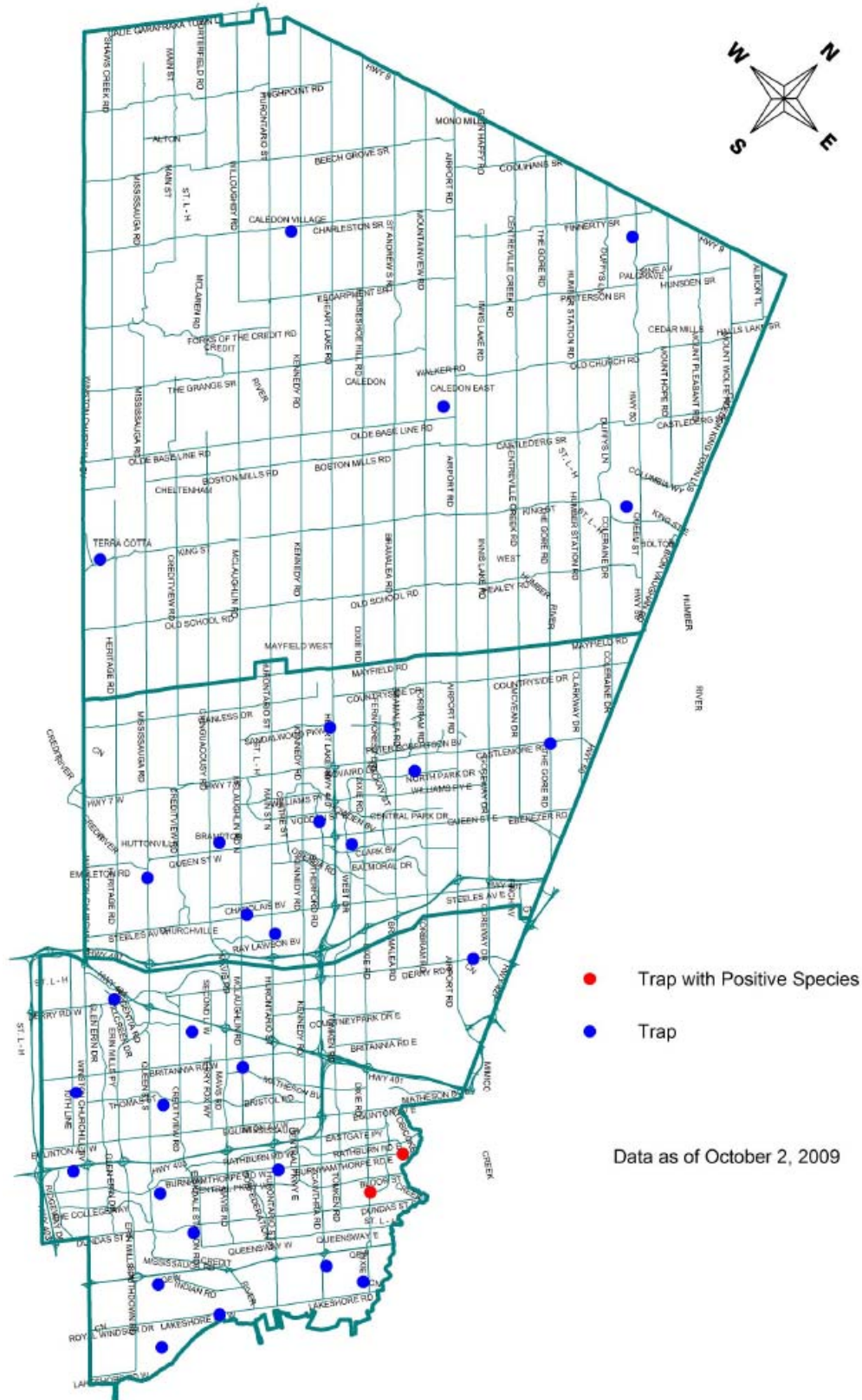


Table 1 presents the estimated number of mosquitoes collected by species in 2009. These estimates are based partially on actual counts, where the pool size was less than 50 mosquitoes, and partially on estimating methods when the pool size exceeded 50 mosquitoes.¹¹ An estimated 64,393 mosquitoes were collected between June 14 to October 3 of 2009 and identified by species.

Table 1 Estimated Number of Female Adult Mosquitoes Collected and Identified by Species, Region of Peel, 2009*

Vector Species	All Peel	Brampton	Caledon	Mississauga North	Mississauga South
<i>Cx. pipiens/restuans</i>	8,862	4,404	284	1,655	2,519
<i>Cx. salinarius</i>	5	2	0	1	2
<i>Ae. vexans vexans</i>	18,358	8,242	699	3,752	5,663
<i>Ae. vexans nipponii</i>	11	2	1	2	6
<i>An. punctipennis</i>	577	167	98	155	157
<i>An. walkeri</i>	14	4	5	0	5
<i>An. quadrimaculatus</i>	65	39	17	3	6
<i>Oc. japonicus</i>	1,414	196	10	410	797
<i>Oc. triseriatus</i>	1,125	48	49	396	632
<i>Oc. trivittatus</i>	5,102	2,321	377	833	1,572
<i>Oc. stimulans</i>	3,913	792	1,991	509	621
<i>Oc. canadensis</i>	3,050	1,745	269	124	911
Non-vectors					
<i>Ae. cinereus</i>	329	128	91	24	85
<i>An. barberi</i>	1	0	0	0	1
<i>An. earlei</i>	4	3	1	0	0
<i>Cq. perturbans</i>	17,575	7,425	2,030	299	7,820
<i>Cs. inornata</i>	1	1	0	0	0
<i>Cs. minnesotae</i>	22	20	2	0	0
<i>Cs. morsitans</i>	16	0	15	1	0
<i>Cx. territans</i>	11	2	0	2	7
<i>Oc. black-legged</i>	136	26	75	17	18
<i>Oc. broad-banded</i>	317	9	243	14	50
<i>Oc. dorsalis</i>	27	10	6	5	6
<i>Oc. excrucians</i>	58	4	54	0	0
<i>Oc. provocans</i>	10	0	10	0	0
<i>Ps. ferox</i>	93	1	2	14	76
<i>Ur. sapphirina</i>	3	1	0	1	1
Males (not identified)	3,294	1,173	103	625	1,394
Totals	64,393	26,766	6,433	8,842	22,351
% Peel Total		41.6%	10.0%	13.7%	34.7%

Source: Sum of weekly reports provided to the Region of Peel from Cosray Labs, 2009
* estimates based on CDC light trap surveillance data

In 2009, like in 2008, *Aedes vexans* was the dominant mosquito species representing 29% (18369/64393) of the total estimated mosquitoes captured during the season. Other mosquito species detected in 2009 included *Coquillettidia perturbans* (27% species abundance), and *Culex pipiens-restuans* (14% species abundance).¹² *Culex* species overall accounted for 14% of the total species found.

Ochlerotatus japonicus (*Oc. japonicus*) a highly competent WNV-vector that is becoming more established in Ontario accounted for 2.2 per cent of species captured. Approximately thirty-one per cent of the mosquitoes were unclassifiable and were placed in an “other species” grouping (not included in the table above).

The breakdown of the percentage of mosquitoes collected by municipality was Mississauga (48%), Brampton (42%) and Caledon (10%).

Oc. japonicus continues to increase in actual counts and trapping events. Laboratory studies indicate that *Oc. japonicus* is a very efficient vector of WNV. This invasive mosquito is native to Asia. It was first identified in North America in New Jersey in 1998. Since then, it has spread rapidly throughout most of eastern North America. Several batches were positive for WNV in the United States in 2000, 2001, and 2002.¹³ In 2007, the first WNV positive *Oc. japonicus* was reported in Ontario (Chatham-Kent).¹⁴

As illustrated in Figure 1, the number of *Oc. japonicus* captured and individually counted has doubled each year since 2007, from 225 in 2007, to 551 in 2008, and 1325 in 2009. The percentage of estimated female *Oc. japonicus* caught relative to other species also increased from 0.76% in 2008 to 2.2 % in 2009. This is a much higher percentage than all years prior to 2008, which remained around 0.4%.

Figure 1 *Ochlerotatus japonicus* (*Oc. japonicus*) abundance (based on actual counts), Region of Peel, 2002 – 2009

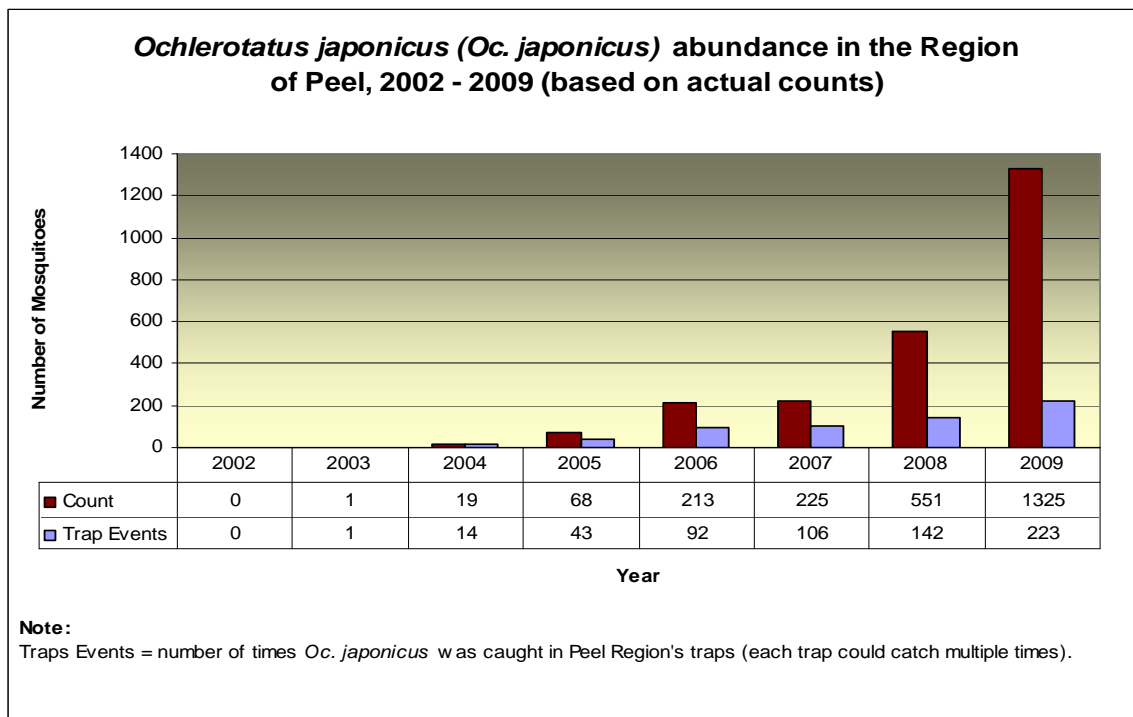


Table 2 presents the number of WNV-positive mosquito trapping events for 2002, 2007, 2008 and 2009 by area municipality. In 2009, there were 4 separate events where a mosquito trap tested positive, compared to 21 in 2008. All four positive trap events occurred in Mississauga. Since the beginning of the WNV surveillance program in the Region of Peel, 2002 remains to be the year with the highest number of positive trapping events (128).

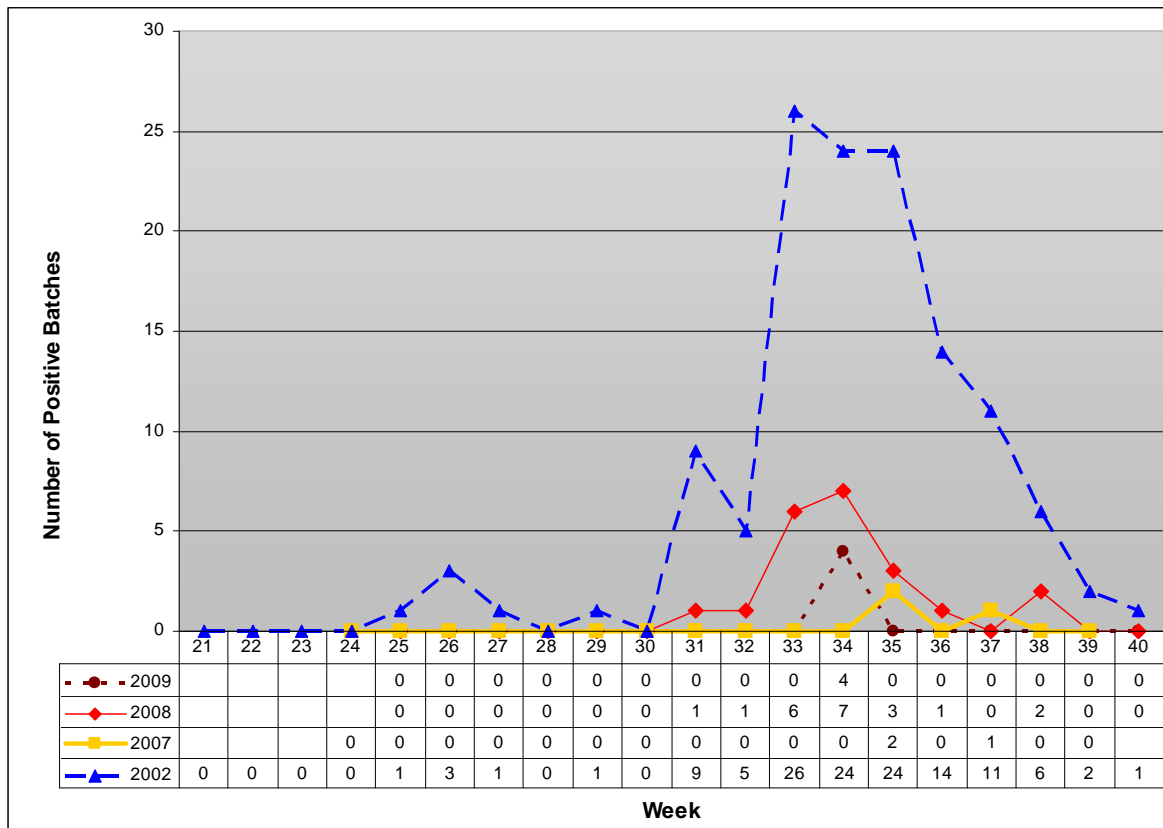
Table 2 Number of Positive Trapping Events by Municipality, Region of Peel, 2002, 2007-2009

Year	Region of Peel	Mississauga	Brampton	Caledon	Date of First Positive Event
2002	128	106	22	0	June 20, 2002
2007	3	1	2	0	August 28, 2007
2008	21	10	11	0	July 29, 2008
2009	4	4	0	0	August 26, 2009

Figure 2 compares the yearly total number of positive batches per week for the 2002 baseline, 2007-2009. Based on previous years' data, batches found with positive mosquitoes are likely to occur anytime after June. There are occasions when no positive mosquitoes are found at sites during the weeks tested. The year to year onset and peak of WNV-positive mosquito batches vary. This is likely due to a range of factors including weather (temperature and rainfall) and the effectiveness of the multifaceted prevention program.

All four positive trapping events in 2009 occurred the week of August 23-29.

Figure 2 WNV Positive Mosquito Batches by Week of Collection, Region of Peel, 2002, 2007-2009



Culex species mosquitoes are of particular interest because they have been the predominant species to carry WNV in Peel Region. Table 3 presents an annual comparison of the number of female *Culex* mosquitoes. In 2009, the absolute number of mosquitoes (all species) captured continued to increase over all other years. Similarly, the absolute number of *Culex* species captured was the highest ever recorded. The relative percentage of *Culex* species to total mosquitoes captured remained high compared to previous years with about 14% in 2009.

However, unlike previous years, 2009 showed the lowest percentage of positive batches containing *Culex* species (25%). The remaining positive batches consisted of two *Aedes vexans* and for the first time ever one *Oc. japonicus* mosquitoes.

Table 3 Total Number and Percentage of Female Adult *Culex pipiens/restuans* Mosquitoes in all Batches and all Positive Batches, Region of Peel, 2002, 2007-2009

Year	All Batches			Positive Mosquito Batches		
	Total Number of mosquitoes	Number of <i>Culex</i> mosquitoes	% <i>Culex</i> in all batches	Total Number of mosquitoes	Number of <i>Culex</i> mosquitoes	% <i>Culex</i> in all positive batches
2002 [†]	24,269	7,278	30.0%	128	98	76.6%
2007 ^a	64,450	4,482	7.0%	3	2	66.7%
2008	78,214	8,431	10.8%	21	20	95.2%
2009 [±]	64,392	8,862	13.8%	4	1	25.0%

[†] Source: 2007 West Nile Virus in the Region of Peel, 2008

^a Source: 2008 West Nile Virus in the Region of Peel, 2009

[±] Source: Cosray Labs, 2009

The Minimum Infection Rate (MIR)

The minimum infection rate (MIR) is used as an indicator of the prevalence of WNV transmission intensity, and therefore the risk for human disease. The MIR is the number of positive batches of infected mosquitoes of a given vector species divided by the total number of mosquitoes of a given vector species that were tested for the presence of the virus, expressed per 1,000.

Table 4 presents the MIR for the *Culex* species, grouped by municipality in Peel Region. The table also includes the MIR in Peel for *Oc. japonicus*. Higher MIRs are usually indicative of greater WNV activity among a given species but can be unreliable when the sample size is less than one thousand. In 2009, the MIRs for *Culex* species were much lower throughout the Region than in 2008. What was unique in 2009 is that the *Oc. japonicus* species showed a higher MIR than *Culex* (0.84 per 1,000 vs. 0.12 per 1,000, respectively).

Table 4 Minimum Infection Rates* of *Culex* Species in Each Municipality in Region of Peel, 2007-2009

Municipality	Vector Species	2009 Actual # Tested	2009 Positive Batches	2009 MIR*	2008 MIR*	2007 MIR*
Mississauga	<i>Culex pipiens/restuans</i>	4015	1	.25	2.12	0.40
Brampton	<i>Culex pipiens/restuans</i>	4215			2.01	0.73
Caledon	<i>Culex pipiens/restuans</i>	284			-	-
Peel	<i>Culex pipiens/restuans</i>	8514	1	.12	1.99	0.50

* The Minimum Infection Rate (MIR) is calculated as the number of positive batches of infected mosquitoes of a given species divided by the total number of mosquitoes of a given vector species that were tested for the presence of the virus, expressed per 1,000.

As shown in Table 5, three positive mosquito batches were attributed to non-*Culex* species in 2009. The MIR for the City of Mississauga was 0.34 for *Ae. vexans* and 0.20 for Region of Peel as a whole. For the first time, *Ae. vexans* showed a higher MIR than *Culex* species compared to any previous year.

Table 5 Minimum Infection Rates of non-*Culex* Species in Each Municipality, Region of Peel, 2008 and 2009

Municipality	Vector Species	2009 Actual Number Tested [†]	2009 Positive Batches	2009 MIR*	2008 Actual Number Tested	2008 Positive Batches	2008 MIR*
Mississauga	<i>Ae. vexans</i>	5862	2	0.34	6993	1	0.14
Brampton	<i>Ae. vexans</i>	3690	-	-	5369	-	-
Caledon	<i>Ae. vexans</i>	575	-	-	842	-	-
Peel	<i>Ae. vexans</i>	10127	2	0.20	13204	1	0.08
	<i>Oc. japonicus</i>	1184	1	.85		-	

* The Minimum Infection Rate (MIR) is calculated as the number of positive batches of infected mosquitoes of a given species divided by the total number of mosquitoes of a given vector species that were tested for the presence of the virus, expressed per 1,000.

† MIRs based on numbers tested < 1000 are less reliable than those based on numbers > 1000.

Adult Mosquito Surveillance in Other Ontario Health Units

The summer of 2009 was a relatively quiet season for WNV activity in Ontario, largely due to weather. The number of positive mosquito pools across the province totalled just 14 (compared to 62 in 2008). Peel Region had the second most positive pools in Ontario (4) after Windsor's 5. Toronto, York and Halton Region's were the health units adjacent to Peel Region that reported positive batches.

Adult Mosquito Surveillance Across Canada

During the 2009 WNV season, thirty-seven (37) WNV positive mosquito pools were identified in Canada [British Columbia (10), Saskatchewan (11), Manitoba (2) and Ontario (14)]. This is down from a total of 122 across Canada in 2008. It was the first time WNV was detected in mosquitoes in British Columbia.

Larval Mosquito Surveillance

Larval surveillance is useful in guiding WNV prevention and reduction activities. It is used to determine the location, species and population densities of mosquitoes. Larval surveillance activities are vital for predicting adult emergence and establishing optimal times for implementation of larval reduction measures.

As a result of funding cut backs, seasonal staff surveyed a variety of aquatic habitats for the presence of mosquito larvae one month later than previous years, from June to September. These potential breeding sites were identified by referring back to breeding site information collected in previous years and by stagnant water complaints received through the Environmental Health Contact Centre or on-line reporting form. Refer to the 2006 WNV in the Region of Peel report for details on the methods used for larval surveillance.¹⁵ Available at: <http://www.peelregion.ca/health/westnile/resources/reports.htm#control06>

In 2009, larval surveillance was undertaken at 2,093 potential mosquito breeding sites on publicly owned lands across Peel Region. Table 6 breaks down the number of surface water sites monitored by municipality and compare this to previous years. The total number of sites monitored across Peel Region decreased in 2009 due to the delayed program start. Overall 58% of the sites were in Mississauga, 21% in Brampton, and 22% in Caledon.

Table 6 Number of Surface Water Sites Monitored by Municipality, Region of Peel, 2002, 2007-2009

Year	Region of Peel	Mississauga	Brampton	Caledon
2002	278	152	106	20
2007	2,400	1,689	451	260
2008	3,479	1,904	784	791
2009*	2,093	1,206	441	446

Note: These are the number of breeding sites identified by WNV students
2009* Monitoring season ran from June to September
2002 - 2008 Monitoring season ran from May - September each year

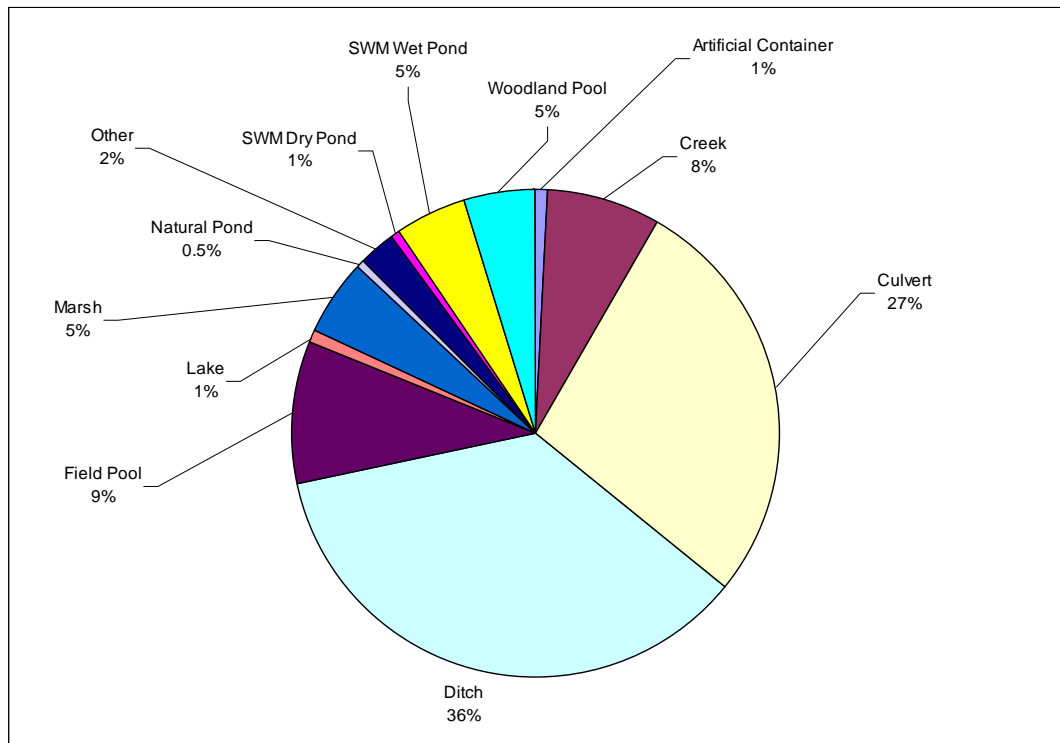
Species Identification – Larval Analysis

In 2009, a total of 5,111 mosquito larvae were identified from early June to the end of September at our in-house lab. This was slightly over double the number of larvae identified in 2008 (2,501). The notable increase in the number of larvae identified can be attributed to having four trained personnel on-site that had received larval identification training.

Fourteen different species of larvae were identified; 61% (3,132) were the two *Culex* species, *pipiens* and *restuans*, the primary WNV vector. This figure is similar to the percentage of identified larvae from these two species in 2008 (61%) and 2007 (60%). Fewer species were identified through larval surveillance in 2009 compared to 2008 and 2007 (21 and 19, respectively)

Figure 3 presents the larval surveillance results by breeding site type (habitat) in Peel Region. Ditches, culverts and field pools make up the greatest proportion (72%) of sites with larvae present. These are also the locations that are most difficult to contain mosquito populations using control measures because of their relative abundance and effectiveness at holding standing water.

Figure 3 Types of Sites Found to Contain Mosquito Larvae, Region of Peel, 2009



Larval Mosquito Reduction

A major part of the Region of Peel WNV Prevention Program is aimed at reducing the number of vector mosquitoes. This is done by eliminating or altering mosquito breeding habitats (source reduction) and by applying pesticides to common habitats during the larval stage of development.

Although public education is used to increase awareness about the importance of reducing mosquito breeding sites by removing standing water around homes, it alone is not enough to effectively reduce vector populations. Therefore, a larviciding program is necessary to control mosquito populations.

The most common breeding sites for mosquito vectors include roadside catch basins, ditches, discarded tires, unused swimming pools and containers left outdoors. These sites are ideal conditions for breeding, providing standing or slow-moving water and decaying organic matter which serves as food for the larvae.

Catch basin networks are extensive in urban and suburban environments. They retain a small amount of water and organic matter in the form of sediment that collects in the sump of the catch basin. The majority of catch basins in Peel Region have been found to contain larvae. Surface water breeding sites are many in number and type and can change from year to year requiring a systematic approach to their surveillance and treatment.

Habitat modification, which includes altering the habitat to eliminate standing water, can also reduce the potential to breed mosquitoes. Peel Public Health staff work with municipal departments to pursue all effective measures to achieve this outcome.

Larvicides

Mosquito larviciding remains a key component of this vector management strategy, and in particular the abatement of mosquitoes in the extensive network of storm-water catch basins and in surface waters located on municipal properties.

Methoprene, a synthetic insect growth regulator, interferes with mosquito larvae development. It has been approved by Health Canada's Pest Management Regulatory Agency (PMRA) for mosquito larviciding. It is effective against the *Culex* species, degrades rapidly in water and is low in toxicity for non-target species. The Ministry of the Environment (MOE) found that methoprene did not harm streams, rivers and drinking water in treated areas and that it was effective in reducing mosquito larvae.¹⁶

In catch basins, contractors for Peel Public Health use either methoprene pellets/briquets (Altosid®) or *Bacillus sphaericus* (VectoLex® WSP – water soluble pouches). Methoprene pellets were used in the majority of roadside catch basins. Methoprene briquets were used in non-roadside catch basins such as those located in public parks and Region of Peel-owned or operated buildings. *Bacillus sphaericus* was used in catch basins draining into Environmentally Sensitive Areas (ESAs). Surface water treatment involved the use of *Bacillus thuringiensis var. israelensis* (VectoBac 1200L or 200G). *Bacillus sphaericus* has a longer residual effect than *Bacillus thuringiensis var. israelensis* and is effective in organic environments.

Pestalto Environmental Health Services Inc. was contracted by Peel Public Health to carry out larviciding of catch basins and surface water sites across the region. Permit applications were prepared by Peel Public Health staff, in consultation with Pestalto, and submitted to the MOE. Three permits were issued in 2009 by the MOE to allow treatment for the following site types: catch basins, surface water and sensitive areas. Notices of larviciding were placed in local newspapers before application began.

Catch Basin Treatment

A total of 361,480 treatments were made to roadside municipal catch basins over four applications from mid-June to early September across Peel Region in 2009. Non-roadside catch basins, such as ones located in parks, private backyards, day cares, government buildings, social housing and long-term care facilities received 2,315 treatments.

Table 7 summarizes all the types and number of catch basin treatments made across the region in 2009.

Table 7 Summary of Catch Basin Treatments, Region of Peel, 2009

Location	Product	Phase	Start Date	End Date	Quantity	Treatments	
Roadside	Altosid® Pellets	1	Jun-15	Jul-06	63.09 kg	90,130	
	Altosid® Pellets	2	Jul-06	Jul-27	63.26 kg	90,371	
	Altosid® Pellets	3	Jul-27	Aug-13	63.14 kg	90,198	
	Altosid® Pellets	4	Aug-17	Aug-27	62.88 kg	89,825	
	Vectolex® WSP	1	Jun-01	Jun-02	239 pouches	239	
	Vectolex® WSP	2	Jul-06	Jul-08	239 pouches	239	
	Vectolex® WSP	3	Aug-05	Aug-07	239 pouches	239	
	Vectolex® WSP	4	Sep-08	Sep-11	239 pouches	239	
	Subtotal						361,480
	Non-Roadside	Altosid® Pellets	1	Aug-26	Aug-26	0.70 g	1
Altosid® XR Briquets		1	Jun-01	Aug-25	2,314 briquets	2,314	
Total			Jun-01	Sep-11	----	363,795	

Source: Pestalto, 2009

Surface Water Treatment

Monitoring mosquito larval habitats to assess the presence and abundance of mosquito larvae was conducted using a standard plastic dipper following the sequential sampling method. On each surveillance visit, the standing water site was given a pool rating based on the total number of larvae observed. Larval samples were also collected and identified by Peel Public Health seasonal staff. In

Peel Region, if vectors were identified then the larval habitats were referred to Pestalto for treatment from June 1 to September 30.

In 2009, 405 surface water sites received a total of 656 treatments (Table 8). This was the second highest number of surface water treatments in the seven year history of Peel's larviciding program. This can, in part, be due to the abundant spring rainfall and cool temperatures which resulted in standing water sites remaining longer than normal.

Table 8 Standing Water Visit and Treatments, Region of Peel, 2009

Site Type	Sites	Surveillance Visits	Surface Area (m) Treated	Sites Treated	Treatments
Artificial Container	2	2	36	2	2
Ditch	374	652	8,951	291	502
Field Pool	80	117	4,719	61	87
Pond	2	8	7,652	2	7
Storm Water Management Pond	12	13	814	12	13
Wetland	4	4	266	3	3
Woodland Pool	50	65	3,061	34	42
Total	524	861	25,499	405	656

Table provided to Region of Peel by Pestalto

The pattern of sites treated with larvicide varied in each municipality (not shown). In Mississauga and Brampton, ditches and culverts accounted for a little over half of the total surface sites treated in each municipality. In Caledon, 76% of the sites treated were ditches and marshes. No storm water retention ponds were treated in Caledon or Brampton.

Risk Assessment Summary

Each year, from mid-June to October, Peel Public Health's West Nile Virus Working Group carries out a weekly risk assessment based on surveillance information collected during that week to identify the risk of human infection in the Peel Region. The working group consists of staff from various programs including environmental health, communications, epidemiology, and communicable disease. Various surveillance factors that influence the risk of WNV infection are evaluated. The factors included are:

- Seasonal temperatures
- Adult mosquito vector abundance
- Virus isolation rate in vector mosquito species
- Human cases of WNV
- Local WNV activity (equine, mosquito)

- Time of year
- WNV activity in proximal urban or suburban regions

Each surveillance factor is assigned a weighted score based on the observations of the previous week. The WNV Mosquito Adulticiding Risk Assessment form is completed weekly (Appendix C) and when the risk assessment level exceeds a value of three, a decision tree process is invoked whereby increased surveillance and the possibility of adulticiding are considered.

Public Education and Community Outreach

The prevention and reduction of WNV risk requires the involvement of many sectors. Engaging individual residents is integral in preventing human infections, particularly in advocating personal protective measures and the elimination of breeding sites on private property.

Peel Public Health has developed various educational resources about personal protective measures and to encourage individual and household activities that prevent or discourage the breeding of mosquitoes. The resources included flyers, fact sheets, posters, mailers and newspaper advertisements. All the materials were made available on the Region of Peel website at <http://www.peelregion.ca/health/westnile/resources/>.

As in previous years, Peel Public Health hand delivered WNV educational flyers to households in the vicinity of a positive mosquito batch or a human case. In 2009, 5167 households were in receipt of the flyer. Peel Public Health staff were also available to conduct stagnant water surveys on residential properties in areas where WNV was detected.

The Vector-Borne Disease team participated in six community displays in 2009. Health promotion items, which included fridge magnets, mosquito swatters and temporary tattoos, were distributed at these events.

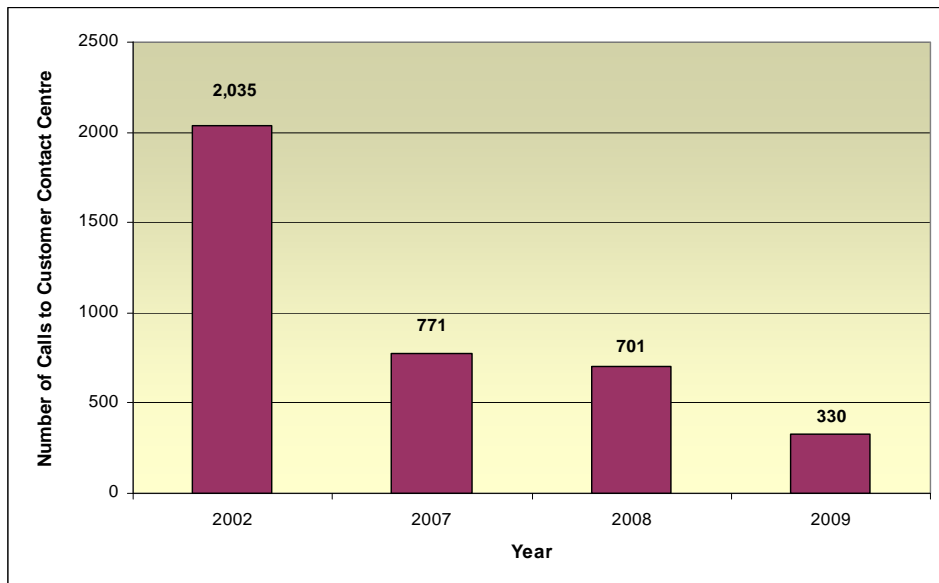
Unlike previous years, public perception (RRFSS) surveys were not conducted in 2009.

West Nile Virus Calls

The Region of Peel Customer Contact Centre is the first level of contact for WNV related inquiries, complaints and reports. WNV inquiries that were of a complex nature were forwarded to the Environmental Health Contact Centre. Peel residents were requested to call the Region with their stagnant water complaints and when there were any questions related to prevention and protection against WNV. The number of calls may be used as a crude indicator of public engagement and concern when compared over a number of years.

In 2009, a total of 330 calls were received (Figure 4). The number of calls received decreased every year with 2009 having the fewest number of calls since first starting to accept calls in 2002. This is in part due to the dead bird surveillance program ending in 2008 and the program starting one month later than previous years.

Figure 4 Number of WNV related calls to the Customer Contact Centre, Region of Peel May 1 - September 30, 2002, 2007-2009



Visits to the Website

The total number of visits to the WNV website in 2009 was 7,138. This is down slightly from 7,602 in 2008. Monthly visits to the website peaked at 911 in June. The website posts surveillance results, updates larviciding activities and provides the public with access to the VBD Prevention Plan as well as annual technical reports dating back to 2002.

Lyme Disease

Lyme disease was first recognized in the United States in 1975, following an outbreak of juvenile rheumatoid arthritis near the community of Lyme, Connecticut. The first reported case in Canada occurred in 1984.

Lyme disease is an illness caused by the bacterium, *Borrelia burgdorferi*, which can be spread through the bite of certain types of ticks. This bacterium is transmitted to ticks when they feed on infected animals and then to humans through the bites of the infected ticks. In Ontario, the disease is spread by the black-legged tick (*Ixodes scapularis*), also known as the deer tick. Lyme disease is the most common tick-borne disease in North America.

Lyme disease must be reported to the local health unit as it is both a reportable and communicable disease under the *Health Protection and Promotion Act*. The standard investigation includes confirming the diagnosis, collecting demographic data, determining location of exposure and investigating possible epidemiological links among cases. This is accomplished by completing the Ministry of Health and Long-Term Care Lyme Disease Human Case Investigation Report which is entered into the provincial Outbreak Module of the integrated Public Health Information System (iPHIS). To date, there has not been a confirmed human case that was locally acquired within Peel.

The first sign of infection is usually a circular rash called erythema migrans or EM. This rash occurs in approximately 70-80% of infected persons and begins at the site of a tick bite after a delay of 3-30 days.¹⁷ The rash gradually expands over a period of several days, reaching up to 30 cm across. The center of the rash may clear as it increases in size, resulting in a bull's-eye appearance. Patients may also experience symptoms of fatigue, chills, fever, headache, and muscle and joint aches, and swollen lymph nodes. In some cases, these may be the only symptoms of infection.

The infection may spread to other parts of the body within a few days to weeks, producing a variety of symptoms if it is not treated. The symptoms include loss of muscle tone on one or both sides of the face, severe headaches and neck stiffness due to meningitis, shooting pains that may interfere with sleep, heart palpitations and dizziness due to changes in heartbeat, and pain that moves from joint to joint. Many of these symptoms will resolve, even without treatment.¹⁷

After several months, approximately 60% of patients with an untreated infection will begin to have intermittent bouts of arthritis, with severe joint pain and swelling.¹⁷ Large joints are most often affected, particularly the knees.

Most cases of Lyme disease can be cured with antibiotics, especially if treatment is occurs early in the course of illness. However, a small percentage of patients with Lyme disease have symptoms that last months to years after treatment with

antibiotics. These symptoms can include muscle and joint pains, arthritis, cognitive defects, sleep disturbance, or fatigue. The cause of these symptoms is not fully understood.

Table 9 shows the number of Peel residents that have been diagnosed with Lyme disease since 2001.

Table 9 Lyme Disease Cases in the Region of Peel, 2001- 2009

Year	Cases
2001	1
2002	2
2003	2
2004	3
2005	3
2006	8
2007	5
2008	18
2009	1

There are areas in Ontario that are considered high risk for Lyme disease because the bacteria have consistently been found in black-legged ticks from these areas. These areas include the north shore of Lake Erie including the Long Point area, Rondeau Provincial Park, Turkey Point and the St. Lawrence Islands National Park.¹⁸ There are concerns that changes of climatic conditions such as warmer seasons could lead to conditions that are favourable for the establishment of black-legged tick populations in many parts of the province. It should be noted that ticks can be spread by birds, in particular songbirds that feed off the forest floor. Because these birds are migratory, there is the potential for new populations of the black-legged ticks to spread across the province. As a result, Peel Public Health undertook an active tick surveillance pilot project in 2009. Active surveillance involves collecting ticks in their natural habitat for identification and testing.

In August 2009, Peel Public Health's Vector-Borne Disease team received a Conservation Areas Access Permit from the Credit Valley Conservation Authority (CVCA) to conduct active black-legged tick surveillance study on CVCA lands. Access was granted for this work between August 27, 2009 and October 3, 2009 in the Rattray Marsh and Meadowvale Conservation Areas. The sites were chosen because of the suitability of habitat, host abundance and potential for dispersal by other hosts like migratory birds.

Between August 27th and September 30th, Peel Public Health's Vector-Borne Disease team staff conducted tick surveillance in and around hiking trails and forested floor areas in Meadowvale and Rattray Conservation Areas. During this pilot project Peel Health staff followed the MOHLTC's Active Tick Surveillance Guidelines.

Drag sampling was conducted to assess the potential distribution of black-legged ticks at the two sites. Drag sampling consists of dragging a flannel white cloth over and around vegetation and leaf litter where ticks may be waiting for a passing host. The drag cloths were made by attaching a one metre square flannel cloth to a 1.2 m wooden stick. A three metre nylon cord was attached to both ends of the stick so the drag can be pulled over vegetation and fallen leaves.

Figure 5 Drag Sampling For Black-Legged Ticks



A total of 270 person-hours of tick dragging were conducted by the six Vector-Borne Disease team students. One hundred hours of tick surveillance were undertaken in the Rattray Marsh and 170 hours at the Meadowvale Conservation Area.

Drag sampling of the two sites did not yield a single black-legged tick or any other tick species. At this time, it does not appear that a black-legged tick population is established in the Meadowvale Conservation Area or the Rattray Marsh. These two habitats can be presently classified as non-endemic for black-legged ticks. However, both sites have the combination of environmental factors to allow new populations of black-legged ticks to become established. Migrating birds or deer could introduce ticks harbouring *B. burgdorferi* to these two sites at some point in the future.

In 2010, Peel Health will continue to examine ticks submitted by Peel residents to identify sites where black-legged ticks are present. If an established tick population is identified or there is evidence of a confirmed case of Lyme disease that has been acquired locally then active surveillance including tick dragging will be undertaken.

Eastern Equine Encephalitis

Eastern equine encephalitis, commonly referred to as EEE, is a viral disease of wild birds that is transmitted to horses and humans by mosquitoes. Of the North American mosquito-borne diseases, EEE appears to be the most severe human pathogen; approximately 33% of people who develop EEE die of the disease, and many survivors have long-term health effects.¹⁹

In Ontario, outbreaks of EEE have occurred sporadically among horses, but no human cases have ever been confirmed. The lack of verified human cases of EEE in Ontario is not entirely understood, since human cases have repeatedly been reported in several states bordering the province. In 2009, two horse cases were reported in the province. The first incidence of the virus being found in Ontario mosquitoes occurred in September 2009, when mosquito pools in Wahta Mohawk Territory tested positive. This First Nations community is located in the Muskoka region near Bala.

Many species of mosquitoes can become infected with EEE virus. However, the most important mosquito species in maintaining the bird-mosquito transmission cycle is *Culiseta melanura*, whose preferred habitat is freshwater hardwood swamps. Adult mosquito surveillance conducted over the last several years has found this species present in Peel but in very low numbers. As noted in Table 10, this species was collected in Mississauga in 2003 and was found in Caledon in 2008.

In 2009, Peel Public Health continued to monitor the prevalence and distribution of *Culiseta melanura* using the region-wide WNV adult mosquito trapping network. No *Culiseta melanura* were collected in Peel traps in 2009; if any adults were found our service provider, Cosray Labs, was to conduct testing for the EEE virus.

Table 10 *Culiseta melanura* found in CDC light traps in the Region of Peel, 2003 and 2008

Year	Wk	Trap ID	Quantity	Location	Municipality
2008	40	T1	1	Bolton Deer Valley Rd & King St	Caledon
2003	38	E1	1	Sugar Bush Bristol Rd & McLaughlin Rd	Mississauga

As in previous years, seasonal field staff working in the Vector-Borne Disease program surveyed a wide range of aquatic habitats for the presence of mosquitoes in the larval stage. Larval surveillance is useful in determining the locations and time of year that mosquitoes use specific aquatic habitats, larval specimens were identified and counted in our in-house laboratory.

Culiseta melanura larvae were not found this past season. This may be due to the fact that *Culiseta melanura* larvae are difficult to collect using the standard WNV dipper methods. This species develop in dark or low light intensity areas such as holes beneath tree roots and stumps, and the underside of root systems of aquatic

plants in fresh-water swamps and marshes containing cool acid water. Thus, different methods of collection are being considered as part of the 2010 EEE surveillance program. Consistent routine testing and monitoring over a period of years will provide data upon which to revise and refine Peel's EEE surveillance.

Conclusion

In 2009, there was a decrease West Nile Virus activity in the Peel Region and across Ontario compared to 2008. In fact, the number of human cases and positive mosquitoes were the lowest recorded in the province since the first locally acquired human cases occurred in 2002.

This past summer was one of the wettest on record. The summer was also cooler than normal: Peel Region only experienced two 30°C days compared to normal 11. So, although the wet weather provided standing water sites for mosquitoes to breed and increase their populations, it was not hot enough to increase the replication rate of mosquitoes.

In 2009, Peel saw a significant drop in the number of positive mosquito batches from 21 in 2008 to four this past season. It is now evident that WNV levels will fluctuate widely from year to year based on precipitation, temperature, abundance of vector mosquito populations and many other factors.

Increased temperatures associated with climate change could increase the survival or replication rates of vector mosquitoes and may contribute to higher incidence of disease in the future. Climate changes could also expand the habitat and infectivity of other disease-carrying insects, increasing the potential for transmission of diseases such as Lyme disease and Eastern equine encephalitis in Peel Region. As a result, it is prudent to continue surveillance programs for these vector-borne diseases.

In 2010, Peel Public Health will continue surveillance, public education and larval mosquito reduction activities as these are essential WNV program components in a jurisdiction where WNV has been detected in a previous season.

In summary, Peel's 2010 VBD program will be similar to the 2009 program provided there are no further provincial funding cuts. However, increased *Culiseta melanura* surveillance may be considered based on the fact the EEE virus was found in Ontario mosquitoes in 2009.

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Appendices

Appendix A

Provincial Surveillance for West Nile Virus – Case Definitions

Provincial Surveillance for West Nile Virus (WNV)

Section A: Case Definitions

The current Case Definitions were drafted with available information at the time of writing. Case Definitions and Diagnostic Test Criteria are subject to change as new information becomes available.

1) West Nile Virus Neurological Syndrome (WNNS):

Clinical Criteria:

History of exposure in an area where WN virus (WNV) activity is occurring¹

OR

history of exposure to an alternative mode of transmission²

AND

onset of fever

AND NEW ONSET OF AT LEAST ONE of the following:

- encephalitis (acute signs of central or peripheral neurologic dysfunction), or
- viral meningitis (pleocytosis and signs of infection e.g. headache, nuchal rigidity), or
- acute flaccid paralysis (e.g. poliomyelitis-like syndrome or Guillain-Barré-like syndrome)³ or
- movement disorders (e.g., tremor, myoclonus) or
- Parkinsonism or Parkinsonia like conditions (e.g., cogwheel rigidity, bradykinesia, postural instability) or
- other neurological syndromes as defined in the note below

¹ History of exposure when and where West Nile Virus transmission is present, or could be present, or history of travel to an area with confirmed WNV activity in birds, horses, other mammals, sentinel chickens, mosquitoes, or humans.

² Alternative modes of transmission, identified to date, include: laboratory-acquired; in utero; receipt of blood components; organ/tissue transplant; and, possibly via breast milk.

³

A person with WNV-associated acute flaccid paralysis may present with or without fever or mental status changes. Altered mental status could range from confusion to coma with or without additional signs of brain dysfunction (e.g. paralysis, cranial nerve palsies, sensory deficits, abnormal reflexes, generalized convulsions and abnormal movements). Acute flaccid paralysis with respiratory failure is also a problem.

Note: A significant feature of West Nile viral neurologic illness may be marked muscle weakness that is more frequently unilateral, but could be bilateral. WNV should be considered in the differential diagnosis of all suspected cases of acute flaccid paralysis with or without sensory deficit. WNV-associated weakness typically affects one or more limbs (sometimes affecting one limb only). Muscle weakness may be the sole presenting feature of WNV illness (in the absence of other neurologic features) or may develop in the setting of fever, altered reflexes, meningitis or encephalitis. Weakness typically develops early in the course of clinical infection. Patients should be carefully monitored for evolving weakness and in particular for acute neuromuscular respiratory failure, which is a severe manifestation associated with high morbidity and mortality. **For the purpose of WNV Neurological Syndrome Classification, muscle weakness is characterized by severe (Polio-like), non-transient and prolonged symptoms.** Electromyography (EMG) and lumbar puncture should be performed to differentiate WNV paralysis from the acute demyelinating polyneuropathy (Guillain-Barré syndrome). Lymphocytic pleocytosis (an increase in WBC with a predominance of lymphocytes in the cerebrospinal fluid [CSF]) is commonly seen in acute flaccid paralysis due to WNV.

Other emerging clinical syndromes, identified during 2002 included, but were not limited to the following: myelopathy, rhabdomyolysis (acute destruction of skeletal muscle cells), peripheral neuropathy; polyradiculoneuropathy; optic neuritis; and acute demyelinating encephalomyelitis (ADEM). Ophthalmologic conditions including chorioretinitis and vitritis were also reported. Facial weakness was also reported. Myocarditis, pancreatitis and fulminant hepatitis have not been identified in North America, but were reported in outbreaks of WNV in South Africa. “Aseptic” meningitis without encephalitis or flaccid paralysis occurring in August and September when WNV is circulating may be due to non-polio enteroviruses circulating at the same time. This should be considered in the differential diagnosis.

[Sejvar J et al. JAMA (2003) Vol.290 (4) p. 511-515, Sejvar, J. et al. Emerg Infect Dis (2003) Vol 9 (7) p.788-93 and Burton, JM et al Can. J. Neurol. Sci. (2004) Vol.31 (2) p.185-193]

Suspect WN Neurological Syndrome Case:

Clinical criteria IN THE ABSENCE OF OR PENDING diagnostic test criteria (see below) AND IN THE ABSENCE of any other obvious cause.

Probable WN Neurological Syndrome Case:

Clinical criteria AND AT LEAST ONE of the probable case diagnostic test criteria (see below).

Confirmed WN Neurological Syndrome Case:

Clinical criteria AND AT LEAST ONE of the confirmed case diagnostic test criteria (see below).

2) West Nile Virus Non-Neurological Syndrome (WN Non-NS):

Clinical Criteria:

History of exposure in an area where WN virus (WNV) activity is occurring¹

OR

history of exposure to an alternative mode of transmission²

AND AT LEAST TWO of the following⁵ :

- fever,⁶
- myalgia ,
- arthralgia,
- headache,
- fatigue,
- lymphadenopathy,
- maculopapular rash

¹
History of exposure when and where West Nile Virus transmission is present, or could be present, or history of travel to an area with confirmed WNV activity in birds, horses, other mammals, sentinel chickens, mosquitoes, or humans.

²
Alternative modes of transmission, identified to date, include: laboratory-acquired; in utero; receipt of blood components; organ/tissue transplant; and, possibly via breast milk.

⁵
It is possible that other clinical signs and symptoms could be identified that have not been listed and may accompany probable case or confirmed case diagnostic test criteria. For example, gastrointestinal (GI) symptoms were seen in many WNV patients in Canada and the USA in 2003 and 2004.

⁶
Muscle weakness may be a presenting feature of WNV illness. **For the purpose of WNV Non-Neurological Syndrome classification, muscle weakness or myalgia (muscle aches and pains) is characterized by mild, transient, unlikely prolonged symptoms that are not caused by motor neuropathy.**

Suspect WN Non-Neurological Syndrome Case:

Clinical criteria IN THE ABSENCE OF OR PENDING diagnostic test criteria (see below) AND IN THE ABSENCE of any other obvious cause.

Probable WN Non-Neurological Syndrome Case:

Clinical criteria AND AT LEAST ONE of the probable case diagnostic test criteria (see below)

Confirmed WN Non-Neurological Syndrome Case:

Clinical criteria AND AT LEAST ONE of the confirmed case diagnostic test criteria (see below)

3) West Nile Virus Asymptomatic Infection (WNAI) ⁷ :

Probable WN Asymptomatic Infection Case:

Probable case diagnostic test criteria (see below) IN THE ABSENCE of clinical criteria

Confirmed WN Asymptomatic Infection Case:

Confirmed case diagnostic test criteria (see below) IN THE ABSENCE of clinical criteria

⁷
This category could include asymptomatic blood donors whose blood is screened using a Nucleic Acid Amplification Test (NAT), by Blood Operators (i.e. Canadian Blood Services or Hema-Quebec) and is subsequently brought to the attention of public health officials. The NAT that will be used by Blood Operators in Canada is designed to detect all viruses in the Japanese encephalitis (JE) serocomplex. The JE serocomplex includes WN virus and 9 other viruses, although from this group only WN virus and St Louis encephalitis virus are currently endemic to parts of North America. Blood Operators in Canada perform a supplementary WN virus-specific NAT following any positive donor screen test result.

Section B: West Nile Virus Diagnostic Test Criteria:

Probable Case Diagnostic Test Criteria:

AT LEAST ONE of the following:

<p>Detection of flavivirus antibodies in a single serum or CSF sample using a WN virus IgM ELISA ⁸ without confirmatory neutralization serology (e.g. Plaque Reduction Neutralization Test [PRNT]) OR</p>
<p>A 4-fold or greater change in flavivirus HI titres in paired acute and convalescent sera or demonstration of a seroconversion using a WN virus IgG ELISA ⁸ OR</p>
<p>A titre of $\geq 1:320$ in a single WN virus HI test, or an elevated titre in a WN virus IgG ELISA, with a confirmatory PRNT result OR [Note: A confirmatory PRNT or other kind of neutralization assay is not required in a health jurisdiction/authority where cases have already been confirmed in the current year]</p>
<p>Demonstration of Japanese encephalitis (JE) serocomplex-specific genomic sequences in blood by NAT screening on donor blood, by Blood Operators in Canada.</p>

⁸ Both CDC and commercial IgM / IgG ELISAs are now available for front line serological testing. Refer to appropriate assay procedures and kit inserts for the interpretation of test results.

Note: WNV IgM antibody may persist for more than a year and the demonstration of IgM antibodies in a patient's serum, particularly in residents of endemic areas, may not be diagnostic of an acute WN viral infection. Seroconversion (by HI, IgG ELISA or PRNT assays) demonstrates a current WNV infection. Therefore, the collection of acute and convalescent sera for serologic analysis is particularly important to rule out diagnostic misinterpretation early in the WNV season (e.g. May, June) and to identify initial cases in a specific jurisdiction. However, it should be noted that seroconversions may not always be documented due to timing of acute sample collection (i.e. titres in acute sera may have already peaked). If static titres are observed in acute and convalescent paired sera, it is still possible the case may represent a recent infection. To help resolve this, the use of IgG avidity testing⁹ may be considered to distinguish between current and past infection. The presence of both IgM antibody and low avidity IgG in a patient's convalescent serum sample are consistent with current cases of viral associated illness. However test results that show the presence of IgM and high avidity IgG are indicative of exposures that have occurred in the previous season. Immunocompromised individuals may not be able to mount an immune response necessary for a serological diagnosis. West Nile Virus diagnostic test criteria for these individuals should be discussed with a medical microbiologist.

⁹ Early in infection the immune system generates antibodies that bind relatively weakly to viral antigen (low avidity). As the infection proceeds, an increasing percentage of newly generated IgG antibody displays higher binding affinity to virus antigen and thus avidity also rises (Note: avidity is usually measured based upon the ability of IgG to dissociate from antigen preparations after incubation with a solution of urea). As long as high avidity IgG is not yet detected in the serum it can be assumed that the individual was exposed to the viral agent during a recent exposure. With respect to WNV infection it has not been precisely determined when (i.e. post-exposure) high avidity antibodies reach levels in serum that can be accurately detected by serological assays (there may be significant variation depending on the individual). However, it has been shown that greater than 95% of sera collected from individuals exposed to WNV 6-8 months previously will have IgG antibodies that bind strongly to viral antigen and will give high avidity scores using both IFA and ELISA testing formats. **Note: Avidity testing will not replace confirmatory neutralization testing, non-WNV flavivirus IgG antibody (Eg. dengue, SLE, etc.) may bind to the antigen preparations used in avidity assays.**

Confirmed Case Diagnostic Test Criteria:

It is currently recommended that health jurisdictions/authorities use the Confirmed Case Diagnostic Test Criteria to confirm index cases (locally acquired) in their area each year; for subsequent cases, health jurisdictions/authorities could use the Probable Case Diagnostic Test Criteria to classify cases in their area as "confirmed", **for the purposes of surveillance**. Throughout the remainder of the transmission season health jurisdictions/authorities may wish to document PRNT antibody titres to West Nile Virus in a proportion of cases, to be determined by that health jurisdiction/authority, in order to rule-out the possibility of concurrent activity by other flaviviruses. [For further information on diagnostic testing algorithms for

West Nile Virus, see the section entitled Laboratory Specimen Diagnostic Testing Algorithm in Appendix 4 of the National Guidelines for Response to West Nile Virus.]

AT LEAST ONE of the following:

A 4-fold or greater change in WN virus neutralizing antibody titres (using a PRNT or other kind of neutralization assay) in paired acute and convalescent sera, or CSF. OR
Isolation of WN virus from, or demonstration of WN virus antigen or WN virus-specific genomic sequences in tissue, blood, CSF or other body fluids OR
Demonstration of flavivirus antibodies in a single serum or CSF sample using a WN virus IgM ELISA ^{8,9} , confirmed by the detection of WN virus specific antibodies using a PRNT (acute or convalescent specimen). OR
A 4-fold or greater change in flavivirus HI titres in paired acute and convalescent sera or demonstration of a seroconversion using a WN virus IgG ELISA ^{8,9} AND the detection of WN specific antibodies using a PRNT (acute or convalescent serum sample).

⁸ Both CDC and commercial IgM / IgG ELISAs are now available for front line serological testing. Refer to appropriate assay procedures and kit inserts for the interpretation of test results.

Note: WNV IgM antibody may persist for more than a year and the demonstration of IgM antibodies in a patient’s serum, particularly in residents of endemic areas, may not be diagnostic of an acute WN viral infection. Seroconversion (by HI, IgG ELISA or PRNT assays) demonstrates a current WNV infection. Therefore, the collection of acute and convalescent sera for serologic analysis is particularly important to rule out diagnostic misinterpretation early in the WNV season (e.g. May, June) and to identify initial cases in a specific jurisdiction. However, it should be noted that seroconversions may not always be documented due to timing of acute sample collection (i.e. titres in acute sera may have already peaked). If static titres are observed in acute and convalescent paired sera, it is still possible the case may represent a recent infection. To help resolve this, the use of IgG avidity testing⁹ may be considered to distinguish between current and past infection. The presence of both IgM antibody and low avidity IgG in a patient’s convalescent serum sample are consistent with current cases of viral associated illness. However test results that show the presence of IgM and high avidity IgG are indicative of exposures that have occurred in the previous season. Immunocompromised individuals may not be able to mount an immune response necessary for a serological diagnosis. West Nile Virus diagnostic test criteria for these individuals should be discussed with a medical microbiologist.

⁹ Early in infection the immune system generates antibodies that bind relatively weakly to viral antigen (low avidity). As the infection proceeds, an increasing percentage of newly generated IgG antibody displays higher binding affinity to virus antigen and thus avidity also rises (Note: avidity is

usually measured based upon the ability of IgG to dissociate from antigen preparations after incubation with a solution of urea). As long as high avidity IgG is not yet detected in the serum it can be assumed that the individual was exposed to the viral agent during a recent exposure. With respect to WNV infection it has not been precisely determined when (i.e. post-exposure) high avidity antibodies reach levels in serum that can be accurately detected by serological assays (there may be significant variation depending on the individual). However, it has been shown that greater than 95% of sera collected from individuals exposed to WNV 6-8 months previously will have IgG antibodies that bind strongly to viral antigen and will give high avidity scores using both IFA and ELISA testing formats. **Note: Avidity testing will not replace confirmatory neutralization testing, non-WNV flavivirus IgG antibody (Eg. dengue, SLE, etc.) may bind to the antigen preparations used in avidity assays.**

Appendix B

Week Codes - 2009 - West Nile Virus

Week # (Sun to Sat)	2009
1	Jan 4 - Jan 10
2	Jan 11 - Jan 17
3	Jan 18 - Jan 24
4	Jan 25 - Jan 31
5	Feb 1 - Feb 7
6	Feb 8 - Feb 14
7	Feb 15 - Feb 21
8	Feb 22 - Feb 28
9	Mar 1 - Mar 7
10	Mar 8 - Mar 14
11	Mar 15 - Mar 21
12	Mar 22 - May 28
13	May 29 - Apr 4
14	Apr 5 - Apr 11
15	Apr 12 - Apr 18
16	Apr 19 - Apr 25
17	Apr 26 - May 2
18	May 3 - May 9
19	May 10 - May 16
20	May 17 – May 23
21	May 24 - May 30
22	May 31 - Jun 6
23	Jun 7 - Jun 13
24	Jun 14 - Jun 20
25	Jun 21 - Jun 27
26	Jun 28 - Jul 4

Week # (Sun to Sat)	2009
27	Jul 5 - Jul 11
28	Jul 12 - Jul 18
29	Jul 19 - Jul 25
30	Jul 26 - Aug 1
31	Aug 2 - Aug 8
32	Aug 9 - Aug 15
33	Aug 16 - Aug 22
34	Aug 23 - Aug 29
35	Aug 30 - Sep 5
36	Sep 6 - Sep 12
37	Sep 13 - Sep 19
38	Sep 20 - Sep 26
39	Sep 27 - Oct 3
40	Oct 4 - Oct 10
41	Oct 11 - Oct 17
42	Oct 18 - Oct 24
43	Oct 25 - Oct 31
44	Nov 1 - Nov 7
45	Nov 8 - Nov 14
46	Nov 15 - Nov 21
47	Nov 22 - Nov 28
48	Nov 29 - Dec 5
49	Dec 6 - Dec 12
50	Dec 13 - Dec 19
51	Dec 20 - Dec 26
52	Dec 27 - Jan 2

Appendix C - 2009 WNV Risk Assessment

Assessment week:

Date completed:

Completed by:

Surveillance Factor	Assessment	Benchmark	Assigned Value
1. Seasonal temperature	1	Two week mean daily temperature below normal (>2°)	
	3	Two week mean daily temperature at or near normal (±2°)	
	5	Two week mean daily temperature above normal (>2°)	
2. Adult mosquito vector abundance Determined by trapping adults, identifying them to species, and comparing numbers to those previously documented for an area	2	Vector abundance well below average (<50%) (or <25% of 2002 data)	
	4	Vector abundance below average (50%-90%) (or 25%-50% of 2002 data)	
	6	Vector abundance average (90%-150%) (or 50%-75% of 2002 data)	
	8	Vector abundance above average (150%-300%) (or 75%-150% of 2002 data)	
	10	Vector abundance well above average (>300%) (or >150% of 2002 data)	
3. Virus isolation rate in vector mosquito species MIR = $\frac{\text{\# of Positive Cx. Pools}}{\text{\# of Cx. Mosquitoes Tested}} \times 1000$ Tested in pools of 50. Expressed as minimum infection rate (MIR) per 1000 female mosquitoes tested (or 10 pools). A single positive pool with < 500 total <i>Culex</i> cannot score higher than 6.	2	MIR*1000 = 0	
	6	MIR*1000 = > 0 - 5	
	8	MIR*1000 = > 5 - 10	
	10	MIR*1000 = > 10	
4. Human Cases of WNV (Probable and Confirmed)	1	No human cases in province or neighbouring US states	
	2	≤ 10 human cases in neighbouring US states, and none in province	
	3	One human case acquired in province or 11-99 in neighbouring US states	
	4	Multiple human cases acquired in province, or ≥ 100 in neighbouring US states	
	5	One or more human cases acquired in region/area	
5. Local WNV activity (do not score if bird testing has stopped, unless benchmark factor is met for a score of 5)	1	No WNV in horses, or mosquitoes in the province	
	3	One or more positive mosquitoes or horses in the province	
	5	One or more positive mosquito batches or horses in Peel Region during week of assessment	
6. Time of Year (score only if virus activity detected in region/area)	1	Before June 15 or after September 15	
	3	Between June 15 and July 15, or between September 1 and September 15	
	5	Between July 15 and September 1	
7 Proximity to urban or suburban regions (score only if virus activity detected in region/area)	1	Virus activity in remote areas	
	2	Virus activity in rural areas	
	3	Virus activity in small towns	
	4	Virus activity in suburban/urban areas	
	5	Virus activity in suburban/urban areas with positive mosquito traps and previous infection rates >5 per 100,000 for a previous season	
Risk Assessment Level		Total	
		Divide total by 7 if summing surveillance factors 1-5 Divide total by 9 if summing surveillance factors 1-7 Divide total by 6 if summing surveillance factors 1-4 Divide total by 8 if summing surveillance factors 1-4 and 6,7 Average	