2021 WEST NILE VIRUS YEAR END REPORT

Grey Bruce Public Health

March 3, 2022

CONTENTS

1.0 OVERVIEW

The adult mosquito surveillance program was a critical component of the overall West Nile Virus (WNV) program for Grey Bruce Public Health in the 2021 season.

No WNV-positive mosquito pools or human cases were reported in Grey Bruce Public Health in 2021; however, in Ontario, a total of twenty-three (23) human cases were reported (Public Health Ontario, 2021).

There were six (6) Eastern Equine Encephalitis (EEEV) positive mosquito pools, and seven (7) equine cases were reported in Ontario in 2021. No human cases of EEEV were reported (PHO, 2021).

2.0 WEST NILE VIRUS TRANSMISSION DYNAMICS

West Nile Virus (WNV) is a member of the viral family Flaviviridae and is considered to be a classic arbovirus (arthropod-borne virus). Arboviruses are a large group of viruses transmitted by blood-feeding insects. WNV is transmitted by mosquitoes, primarily to birds, but it can sometimes spread to mammalian populations as well (Figure 1). There are two types of mosquito vectors involved in the WNV transmission cycle: 1) Enzootic vectors – which feed primarily on birds (and are referred to as bird-biting vectors) and 2) Bridge vectors – which feed on both birds and mammals, but primarily on mammals.

WNV was first isolated in the West Nile district of Uganda in 1937. WNV was initially endemic only in the eastern hemisphere, but spread to the western hemisphere in 1999, where it was first discovered in the greater New York City area. The first positive dead bird was reported in 2001 in Southern Ontario and the virus has since spread throughout Canada and become endemic. In 2021, in Ontario alone, there were 23 WNV-positive humans, 11 WNV-positive dead birds, and 107 WNV-positive mosquito pools. There were no WNV-positive equine cases reported in 2021. (PHO, 2021).

Mammals are considered dead-end hosts of WNV because they do not produce significant viremia to be able to infect any mosquitoes that feed upon them. Mosquitoes from the genus *Culex* are the main enzootic vectors responsible for amplifying WNV in bird populations. Thus, most control programs emphasize the reduction of *Culex* species populations. Without a significant *Culex* population there is inadequate amplification of WNV and therefore limited risk of human infection.

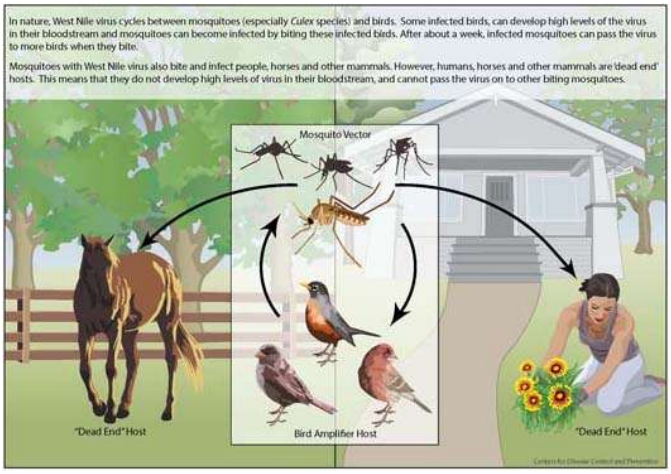

Figure 1. West Nile Virus Transmission Cycle (Centers for Disease Control and Prevention, 2021).

2.1 THE ROLE OF CULEX SPECIES IN WNV TRANSMISSION

According to most researchers, the major WNV enzootic vectors in Ontario are *Culex pipiens* and *Culex restuans*, which are both very competent vectors. *Cx. restuans* is an early season species and is replaced by *Cx. pipiens* as the season progresses. Research by Dr. Curtis Russell indicates that in certain instances, *Cx. pipiens* may be attracted to humans as well as to birds (Russell, 2008). Thus, *Cx. pipiens* may also serve as a bridge vector of WNV to humans. Other studies have shown that *Cx. pipiens* can transmit WNV to humans, potentially being responsible for up to 80% of human cases (Kilpatrick et al., 2005).

It has been shown that the risk of human disease increases in areas with large numbers of *Culex* mosquitoes throughout the season, whereas areas lacking high numbers of *Culex* mosquitoes have a much lower incidence of human cases. According to Dr. Henry Cuevas (pers. comm.) average daily temperatures must be at least 16.3˚C for amplification of the virus to occur within the mosquito.

Mosquitoes have a complex life cycle, with four discrete stages: egg, larva, pupa and adult (Figure 2). The first three life stages are aquatic and *Culex* mosquitoes thrive in organically enriched standing water.

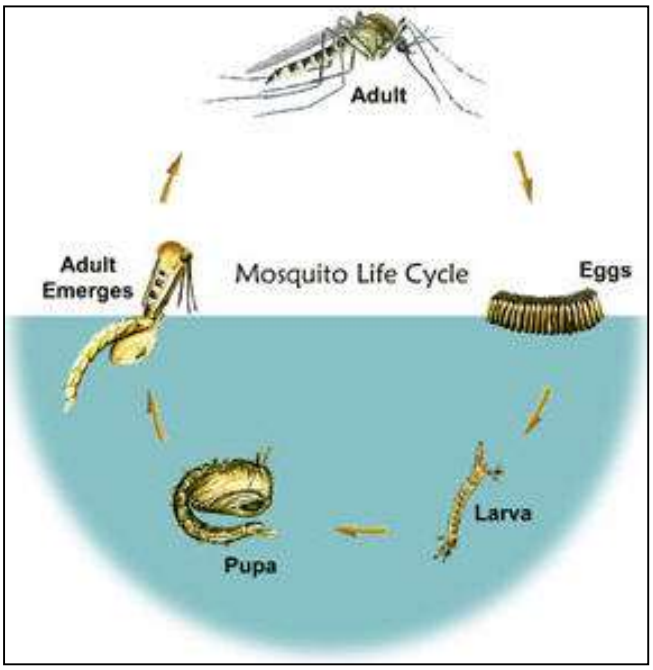

Figure 2. Mosquito Life Cycle (United States Environmental Protection Agency, 2021, recreated from a publication by D.M. Wood).

3.0 EASTERN EQUINE ENCEPHALITIS VIRUS TRANSMISSION DYNAMICS

Eastern Equine Encephalitis virus (EEEV) is a member of the viral family Togaviridae and is considered to be a classic arbovirus (arthropod-borne virus). EEEV is transmitted by mosquitoes, primarily to birds, but it can sometimes spread to mammalian populations as well (Figure 3). There are two types of mosquito vectors involved in the EEEV transmission cycle: 1) Enzootic vectors – which feed primarily on birds (and are referred to as bird-biting vectors) and 2) Bridge vectors – which feed on both birds and mammals, but primarily on mammals.

EEEV was first discovered in Massachusetts, USA in 1831. There are four lineages of EEEV of which Group I is endemic to North America and the Caribbean and is the main cause of human related cases. Groups IIA, IIB and III are primarily responsible for equine illness in Central and South America. The first positive horse was reported in 1938 in Southern Ontario.

Mammals are thought to be dead-end hosts of EEEV because they do not produce significant viremia to be able to infect any mosquitoes that feed upon them. The mosquito *Culiseta melanura* is the main enzootic vector responsible for amplifying EEEV in bird populations. Without a significant *Culiseta melanura* population there is inadequate amplification of EEEV and therefore limited risk of human infection.

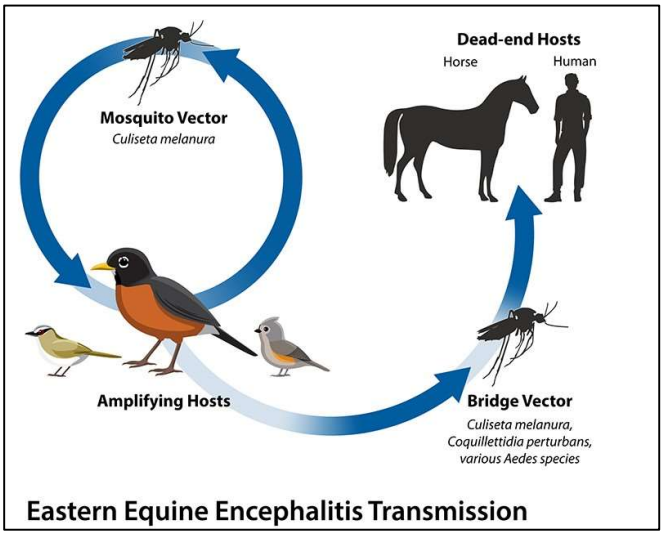

Figure 3. Eastern Equine Encephalitis Virus Transmission Cycle (CDC, 2021).

3.1 THE ROLE OF CULISETA MELANURA IN EEEV TRANSMISSION

Culiseta melanura is the main enzootic vector of EEEV in Ontario. The preferred habitat of this species is freshwater, hardwood swamps where they lay their eggs in the underground crypts in the root mats of trees. While this mosquito will occasionally bite humans, their preference is for an avian host. As a result, contracting EEEV from a bite of *Culiseta melanura* is not considered a significant risk to humans. Transmission to humans is more commonly associated with bridge vectors, such as *Aedes vexans*, *Coquillettidia perturbans*, and some species of the genus *Culex*. In 2021, there were 6 EEEV-positive mosquito pools reported (PHO, 2021).

Reports of human infection are very rare, with the United States reporting an average of 11 human cases per year. (CDC, 2021). No human EEEV cases were reported in Canada in 2021.

Horses are susceptible to EEEV infection, and some cases can be fatal. However, infected horses are not considered to be of significant risk to humans because, like humans, they are thought to be dead-end hosts (CDC, 2021). In 2021, seven (7) EEEV-positive horses were reported in Canada, all seven cases were reported in Ontario (CAHSS, 2021).

4.0 WEST NILE VIRUS ACTIVITY SUMMARY FOR CANADA, 2021

4.1 WEST NILE VIRUS HUMAN CASES IN CANADA, 2021

A total of thirty-four (34) West Nile virus (WNV) cases have been reported in Canada in 2021 (Figure 4). Twenty-nine (29) were clinical cases of which six (6) were classified West Nile Virus Neurological syndrome, eight (8) were classified as West Nile Virus Non-Neurological syndrome and thirty (30) were unclassified. There were five (5) asymptomatic infections reported. There have been no deaths associated with WNV reported to PHAC in 2021.

Figure 4. Total Human West Nile Virus cases in Canada, 2021. Red areas indicate WNV-positive cases. Number of reported cases marked within the province. (Reported by PHAC as of November 18, 2021. Ontario numbers provided by PHO).

4.2 WEST NILE VIRUS POSITIVE BIRD CASES IN CANADA, 2021

Dead birds were collected and submitted to Canadian Wildlife Health Cooperative (CWHC) as part of the 2021 surveillance season. In Canada, samples were submitted from the eastern, western and central parts of the country. In total, 29 birds tested positive for WNV. The birds that tested positive were submitted from Quebec (17), Ontario (11), and Manitoba (3). (Figure 5).

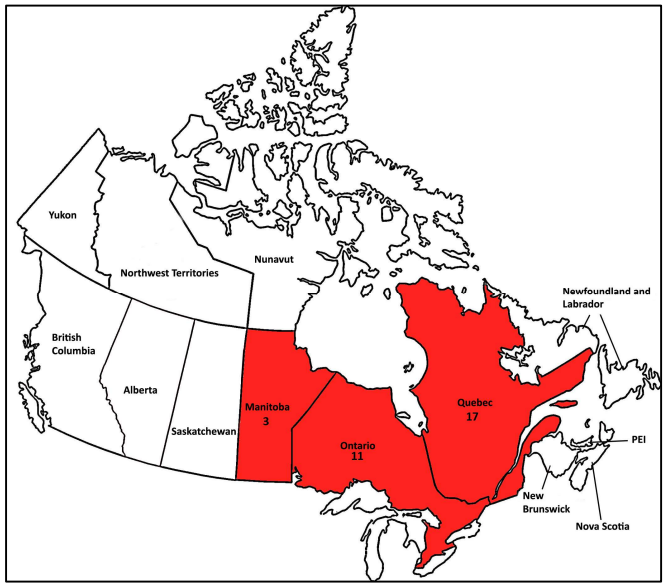

Figure 5. West Nile Virus positive bird cases in Canada, 2021. (Numbers confirmed from PHAC as of November 18, 2021. Ontario numbers provided by CWHC).

4.3 WEST NILE VIRUS POSITIVE EQUINE CASES IN CANADA, 2021

According to the Canadian Animal Health Surveillance System (CAHSS), eighteen (18) horses tested positive for West Nile Virus in 2021 (Figure 6). The cases were reported in Saskatchewan (8), Manitoba (6), Ontario (3) and Alberta (1).

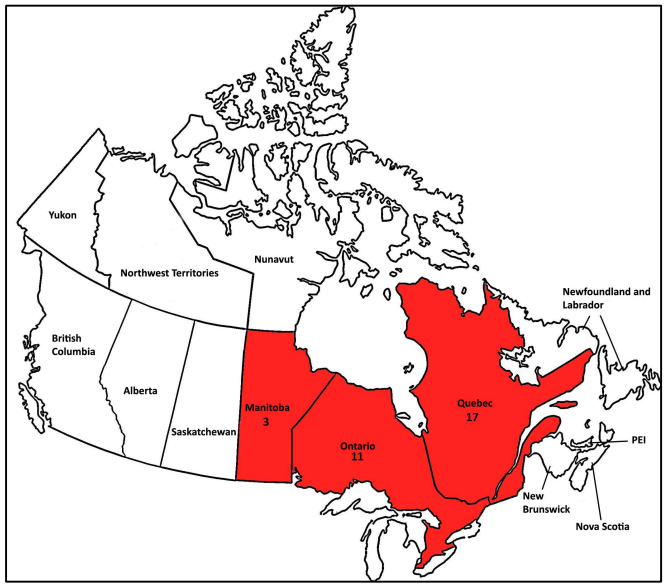

Figure 6. West Nile Virus positive equine cases in Canada, 2021. (Numbers confirmed from CAHSS as of November 18, 2021).

4.4 WEST NILE VIRUS POSITIVE MOSQUITO CASES IN CANADA, 2021

A total of 229 mosquito pools tested positive for West Nile virus in Canada in 2021 (Figure 7). The majority of positive mosquito pools (n=116) were reported from Manitoba, followed by Ontario (n=105), Quebec (n=10) and Saskatchewan (n=9).

Figure 7. West Nile Virus positive mosquito pools in Canada, 2021. Red areas indicate WNV-positive cases with numbers indicating the total number of positive pools. (Reported by PHAC as of November 18, 2021. Ontario numbers provided by PHO).

5.0 WEST NILE VIRUS ACTIVITY IN THE UNITED STATES, 2021

As of November 16, 2021, a total of 1,974 cases of West Nile virus disease in people have been reported to CDC. Of these, 1362 (69%) were classified as neuroinvasive (neurological) and 612 (31%) were classified as non-neuroinvasive (non-neurological). Exact numbers of mosquito, bird and equine cases was not easily accessible.

6.0 WEST NILE VIRUS ACTIVITY SUMMARY FOR ONTARIO, 2021

6.1 WEST NILE VIRUS HUMAN CASES IN ONTARIO, 2021

As of November 18, 2021, 23 human WNV cases have been reported in 10 different health units (Figure 8). The positive cases were from Toronto (5), Chatham-Kent (4), Windsor-Essex (3), Durham (2), Niagara (2), Peel (2), Waterloo (2), and one each from Hamilton, Eastern Ontario, and Wellington-Dufferin-Guelph.

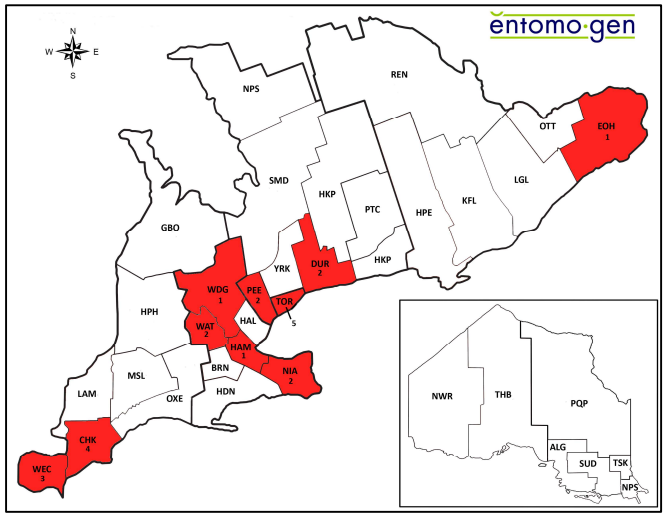

Figure 8. West Nile Virus human cases in Ontario, 2021. Red areas indicate WNV-positive cases. Number of reported cases recorded within health unit boundary. (Reported by Public Health Ontario as of November 22, 2021).

6.2 WEST NILE VIRUS POSITIVE BIRD CASES IN ONTARIO, 2021

Twelve (12) WNV-positive birds were reported from six Health Units in Ontario in 2021 (Figure 9). In total six different species of bird tested positive for WNV. Four (4) American Crows (*Corvus brachyrhynchos*), three (3) Red Tailed Hawks (*Buteo jamaicensis*), two (2) Blue Jays (*Cyanocitta cristata*) one (1) Coopers Hawks (*Accipiter cooperii*), one (1) Common Raven (*Corvus corax*) and one (1) Loggerhead Shrike (*Lanius ludovicianus*).

The WNV-positive birds were reported from Toronto (3), Hamilton (3), Peel (2), Wellington-Dufferin-Guelph (2), Halton (1), Niagara (1).

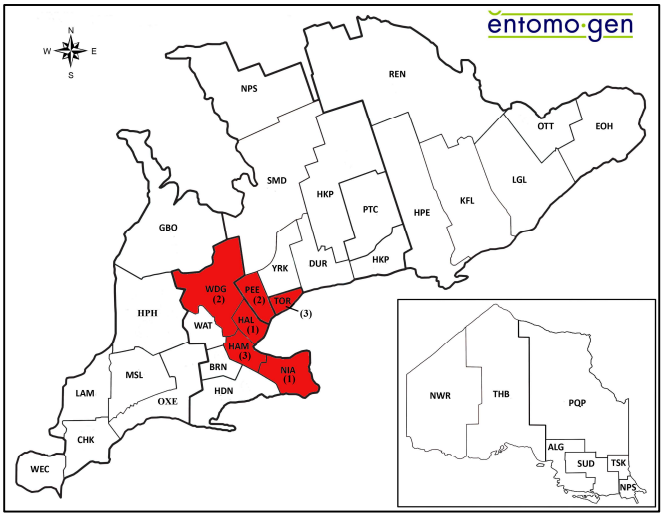

Figure 9. West Nile Virus bird cases in Ontario, 2021. Red areas indicate WNV-positive cases. Number of reported cases recorded within health unit boundary. (Reported by CWHC as of November 26, 2021).

6.3 WEST NILE VIRUS POSITIVE EQUINE CASES IN ONTARIO, 2021

According to the Canadian Animal Health Surveillance System (CAHSS), three horses tested positive for West Nile Virus in 2021 (Figure 10). The positive cases were reported from Chatham-Kent, Haliburton, Kawartha Pine Ridge and Leeds, Grenville and Lanark Health units.

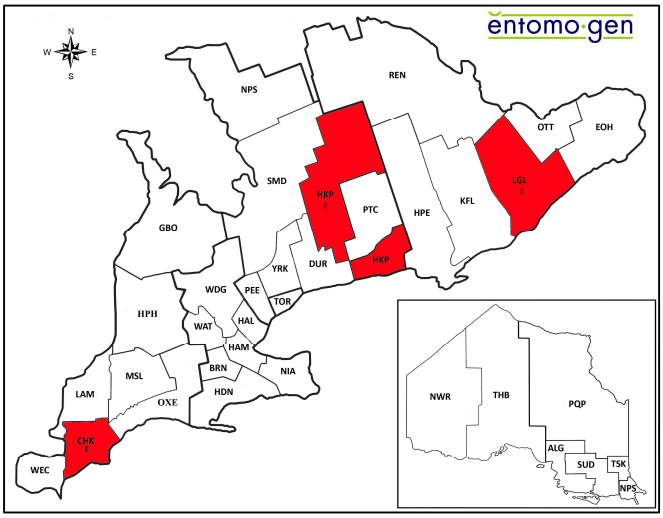

Figure 10. West Nile Virus positive horses in Ontario, 2021. Red areas indicate WNV-positive cases. Number of reported cases recorded within health unit boundary. (Reported by Public Health Ontario as of November 18, 2021).

6.4 WEST NILE VIRUS POSITIVE MOSQUITO CASES IN ONTARIO, 2021

From mosquito surveillance conducted by provincial health units, 107 WNV-positive mosquito pools were confirmed from 17 separate Ontario Health Units in 2021 (Figure 11). The two WNV-positive pools from Lambton County were reported by First Nations community Walpole Island as part of their own mosquito surveillance program.

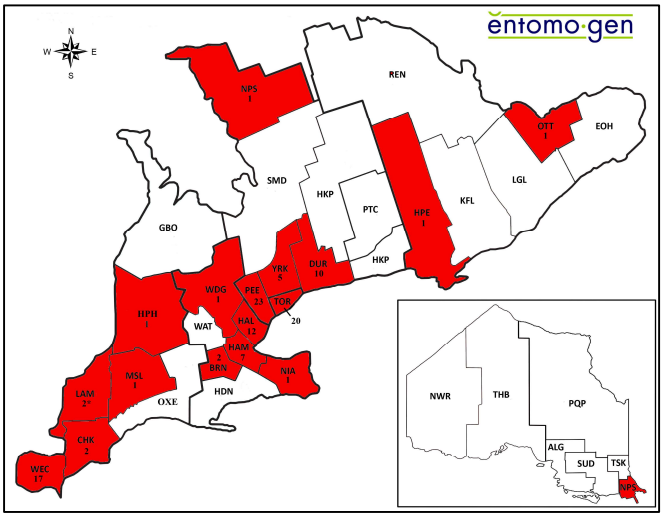

Figure 11. West Nile Virus positive mosquitoes in Ontario, 2021. Red areas indicate WNV-positive cases. Number of reported cases recorded within health unit boundary. (Reported by Public Health Ontario as of November 18, 2021).

The positive pools were from Peel (23), Toronto (20), Windsor-Essex (17), Halton (12), Durham (10), Hamilton (7), Ottawa (7), York (5), Brant (1), Chatham-Kent (2), Lambton (2). Hastings Prince Edward, Huron Perth, Middlesex London, Niagara, North Bay Parry Sound, Ottawa and Wellington-Dufferin-Guelph each reported one WNV-positive pool.

7.0 EASTERN EQUINE ENCEPHALITIS VIRUS ACTIVITY SUMMARY FOR ONTARIO, 2021

7.1 EASTERN EQUINE ENCEPHALITIS VIRUS EQUINE CASES IN ONTARIO, 2021

Seven (7) EEEV-positive horses were reported from 3 Health Units in Ontario in 2020 (Figure 12). The cases were reported in North Bay Parry Sound (5), Eastern Ontario (1) and Leeds, Grenville and Lanark (1).

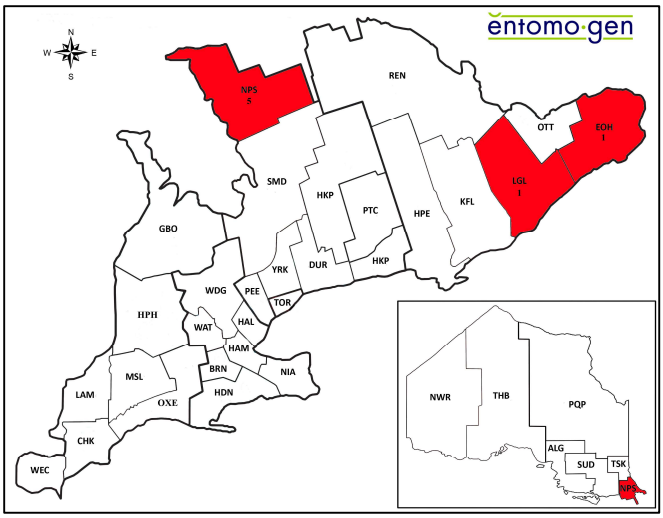

Figure 12. Eastern Equine Encephalitis Virus positive horses in Ontario, 2021. Red areas indicate WNV-positive cases. Number of reported cases recorded within health unit boundary. (Reported by CAHSS as of October 22, 2021).

7.2 EASTERN EQUINE ENCEPHALITIS HUMAN AND MOSQUITO CASES IN ONTARIO, 2021

There were no EEEV-positive human cases reported in however, 6 mosquito pools tested positive for EEEV in 2021 (Figure 13). Three (3) of the EEEV-positive mosquito pools were reported from North Bay Parry Sound District Health Unit while the other three were reported by First Nations Community Wahta Mohawk Territory located within the boundaries of the Simcoe Muskoka District Health Unit.

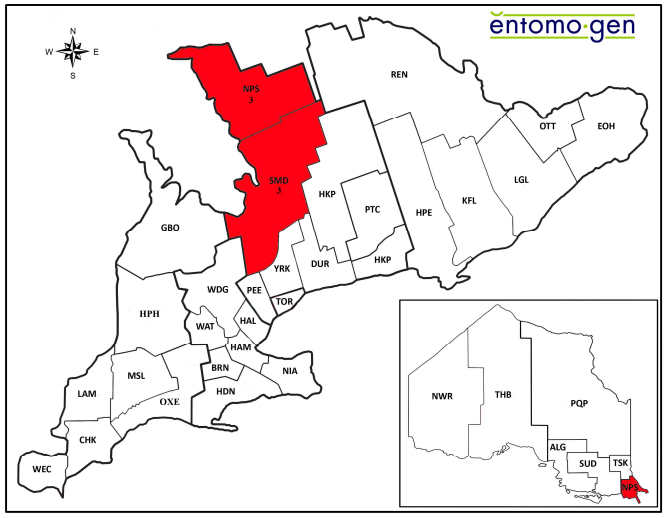

Figure 13. Eastern Equine Encephalitis Virus positive horses in Ontario, 2021. Red areas indicate WNV-positive cases. Number of reported cases recorded within health unit boundary. (Reported by CAHSS as of October 22, 2021).

8.0 TIMING OF WNV-POSITIVE CULEX PIPIENS/RESTUANS IN GREY BRUCE PUBLIC HEALTH BASED ON 2021 TEMPERATURES

Based on an accumulated degree-day model used by Public Health Ontario, the rate at which WNV replicates within the adult females of *Culex pipiens/restuans* depends on ambient temperatures. Below 18.3˚C (average daily field temperature) there is no extrinsic incubation of WNV but above this temperature threshold the virus will replicate in the mosquito. 380 accumulated degree-days were required for 50% of infected *Culex pipiens/restuans* mosquitoes to test positive for WNV. Figure 13a shows the total accumulated degree-days that occurred during the 2021 season in Ontario, highlighting Grey Bruce Public Health in red. According to this model, there appeared to be insufficient heat units in 2021 for amplification of the virus in *Culex* spp. mosquitoes.

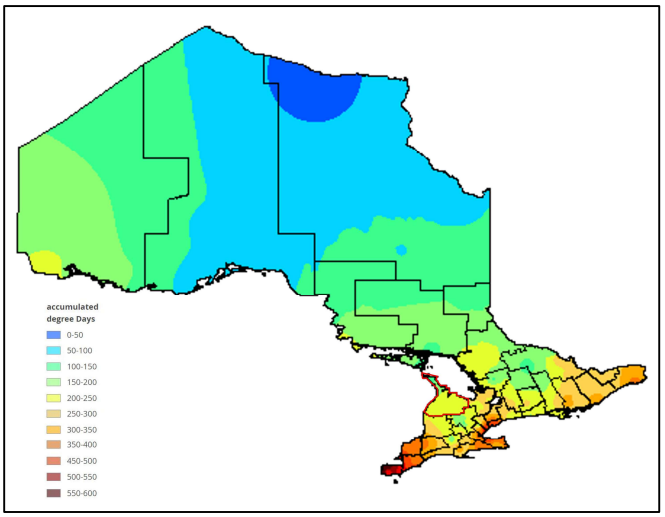

Figure 13a. Accumulated Degree-Day Graph for Ontario, highlighting Grey Bruce Public Health, 2021.

Figure 13b shows the gradual increase in Accumulated degree-days (grey shaded area) that occurred during the 2021 season in Grey Bruce Public Health. In total, there were 172.3 accumulated degree days, based on temperature readings taken from the Wiarton A Station.

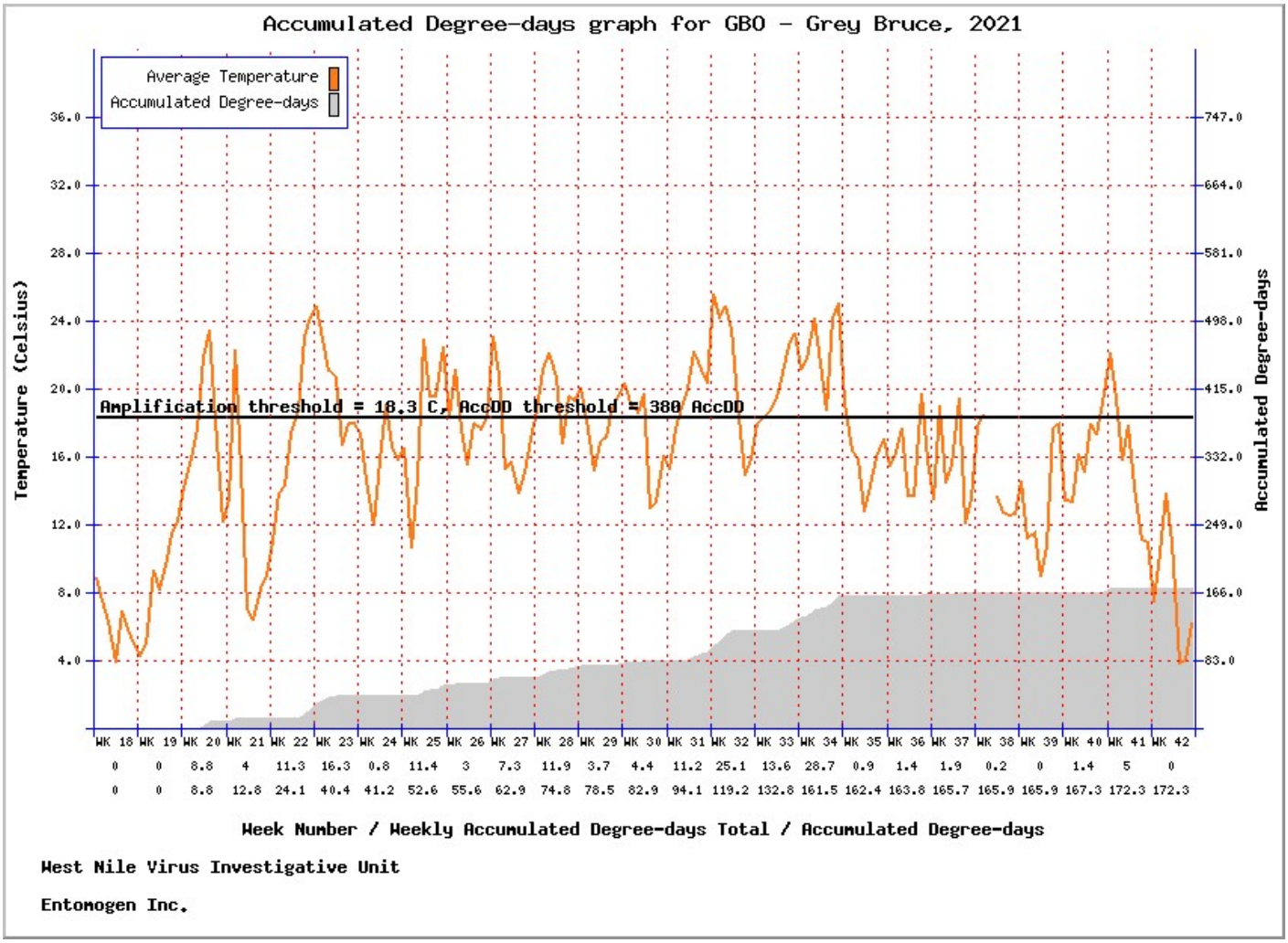

Figure 13b. Accumulated Degree-Day Graph for Grey Bruce Public Health, 2021.

9.0 GREY BRUCE PUBLIC HEALTH MOSQUITO SURVEILLANCE DATA, 2021

A total of eighty-five (85) traps were submitted from 18 different sites during the 2021 trapping season. Figure 14 shows the location of all 18 sites within Grey Bruce Public Health.

17 traps per month were submitted to Entomogen Inc. processing, between May 11 and September 28, 2021. Traps were sorted to a maximum subsample of up to 150 mosquitoes. Any additional mosquitoes were labeled and stored as extras. Sorted mosquitoes were identified to the species level and individuals of the same species were pooled for subsequent viral testing. Appendix A shows a summary of the number of mosquitoes trapped in each of the 18 sites within Grey Bruce Public Health.

In total, there were 6,948 mosquitoes collected, of which 10 were unidentifiable (i.e., damaged females) and 316 were unidentified males. A subsample of 3,117 mosquitoes was examined under a dissecting microscope.

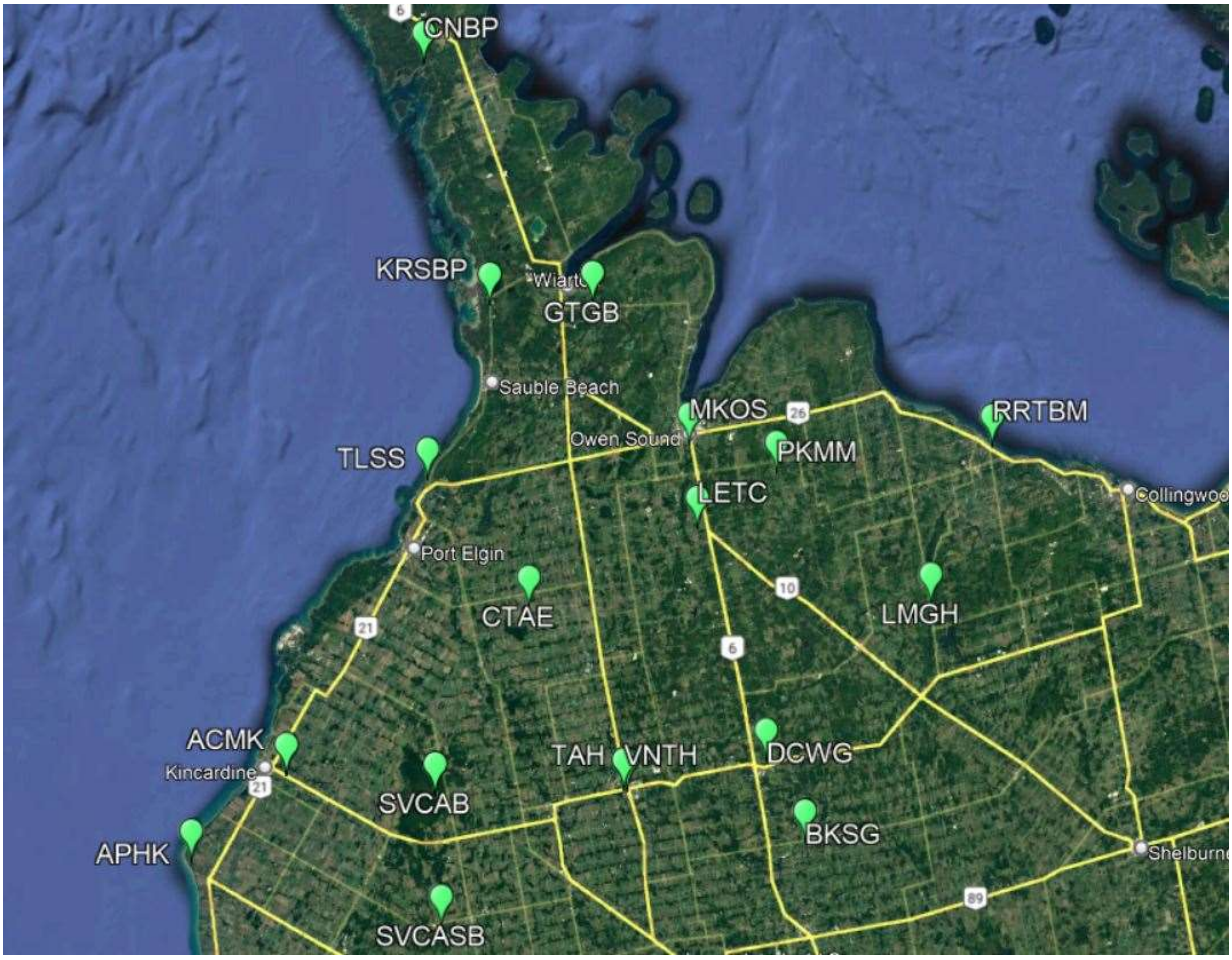

Figure 14. Adult mosquito trap sites for Grey Bruce Public Health, 2021

9.1 MOSQUITO SPECIES COLLECTED IN GREY BRUCE PUBLIC HEALTH, 2021

Figure 15 shows the species found in Grey Bruce Public Health throughout the season. The majority of mosquitoes were non vector species (58%), which are of no significant concern with regards to WNV. Potential bridge vectors species, capable of biting an infected bird and transmitting the virus from the infected bird to a human, horse, or other mammal are highlighted in pink in Figure 15. These species made up approximately 32% of the species identified from traps collected in 2021; thus, humans living within Grey Bruce Public Health may have come in contact with blood feeding *Aedes/Ochlerotatus* mosquitoes.

Enzootic vectors, or bird-biting mosquitoes, composed primarily of *Culex pipiens/restuans*, made up approximately 5% of the species collected. Research indicates that *Cx. pipiens* may be attracted to humans as well as to birds (Russell, 2008). Therefore, humans may have come in contact blood feeding *Culex* as well.

Table 1 lists the mosquito species identified from sites in Grey Bruce Public Health. The primary WNV enzootic vector was *Culex pipiens/restuans* (4.94% of the population) and the primary WNV bridge vector was *Ochlerotatus canadensis* (9.24% of the population). There were a significant number (n=142, 5% of the population) of *Culiseta melanura* – the primary EEEV vector –collected from traps submitted in 2021.

Table 1. Mosquitoes identified from Sites in Grey Bruce Public Health in 2021

	WNV Enzootic Vectors	
154	*Culex pipiens/restuans*	4.94%
1	*Culex species*	0.03%
	WNV Bridge Vectors	
288	*Ochlerotatus canadensis*	9.24%
230	*Aedes vexans vexans*	7.38%
154	*Ochlerotatus stimulans*	4.94%
109	*Ochlerotatus trivittatus*	3.50%
107	*Anopheles punctipennis*	3.43%
40	*Ochlerotatus triseriatus*	1.28%
37	*Anopheles quadrimaculatus*	1.19%
29	*Ochlerotatus japonicus*	0.93%
16	*Anopheles walkeri*	0.51%
	EEEV Vector	
142	*Culiseta melanura*	4.56%

	Non Vectors	
949	*Coquillettidia perturbans*	30.45%
508	*Ochlerotatus broad-banded*	16.30%
210	*Ochlerotatus black-legged*	6.74%
71	*Aedes/Ochlerotatus species*	2.28%
45	*Aedes cinereus*	1.44%
13	*Uranotaenia sapphirina*	0.42%
5	*Ochlerotatus excrucians*	0.16%
3	*Anopheles earlei*	0.10%
2	*Culex territans*	0.06%
2	*Ochlerotatus grossbecki*	0.06%
1	*Culiseta morsitans*	0.03%
1	*Psorophora ferox*	0.03%

9.2 TRAP INDEX OF GROUPS BY WEEK IN GREY BRUCE PUBLIC HEALTH, 2021

The **trap index (TI)** is a useful tool for summarizing trap data and for comparing different time periods and locations (Figure 16). We use TI to show population fluctuations of a particular group of mosquitoes (enzootic vectors vs. bridge vectors). TI is the average number of females per taxon per trap night.

There was a significant WNV bridge vector population throughout the season, which peaked with a TI value of 81 in week 33. The low WNV enzootic vector populations (consisting almost entirely of *Cx. pipiens/restuans*) and EEEV vector population (*Culiseta melanura)*, also peaked at week 33 TI values of 19.2. and 31.4, respectively.

9.3 MOSQUITO SPECIES DISTRIBUTION AMONG SITES IN GREY BRUCE PUBLIC HEALTH, 2021

The pie charts in Figure 17 show WNV enzootic species (yellow), WNV bridge species (pink), and non-vectors (green). Site APHK produced the highest percentage of WNV enzootic vectors (59.1%) while site RRTBM produced the highest percentage of WNV bridge vectors (64.2%). Overall, site SVCAB had the highest number of identified mosquitoes while site TAH had the lowest number. Total numbers and percentages for all sites can be found in Appendix B.

10.0 WEST NILE VIRUS AND EASTERN EQUINE ENCEPHALITIS VIRUS ANALYSIS, 2021

Appendix C outlines the order of preference for viral testing. A maximum of three pools per trap were sent for WNV and/or EEEV viral testing.

10.1 WEST NILE VIRUS VIRAL TESTING RESULTS

From the 155 traps submitted a total of 145 were pools sent for WNV-testing. There were no WNV-positive pools reported in Grey Bruce Public Health in 2021.

10.2 EASTERN EQUINE ENCEPHALITIS VIRUS ANALYSIS, 2021

There was a total of eleven (11) pools of *Culiseta melanura* – the primary vector for Eastern Equine Encephalitis – tested from Grey Bruce Public Health in 2021. All eleven pools tested negative for EEEV.

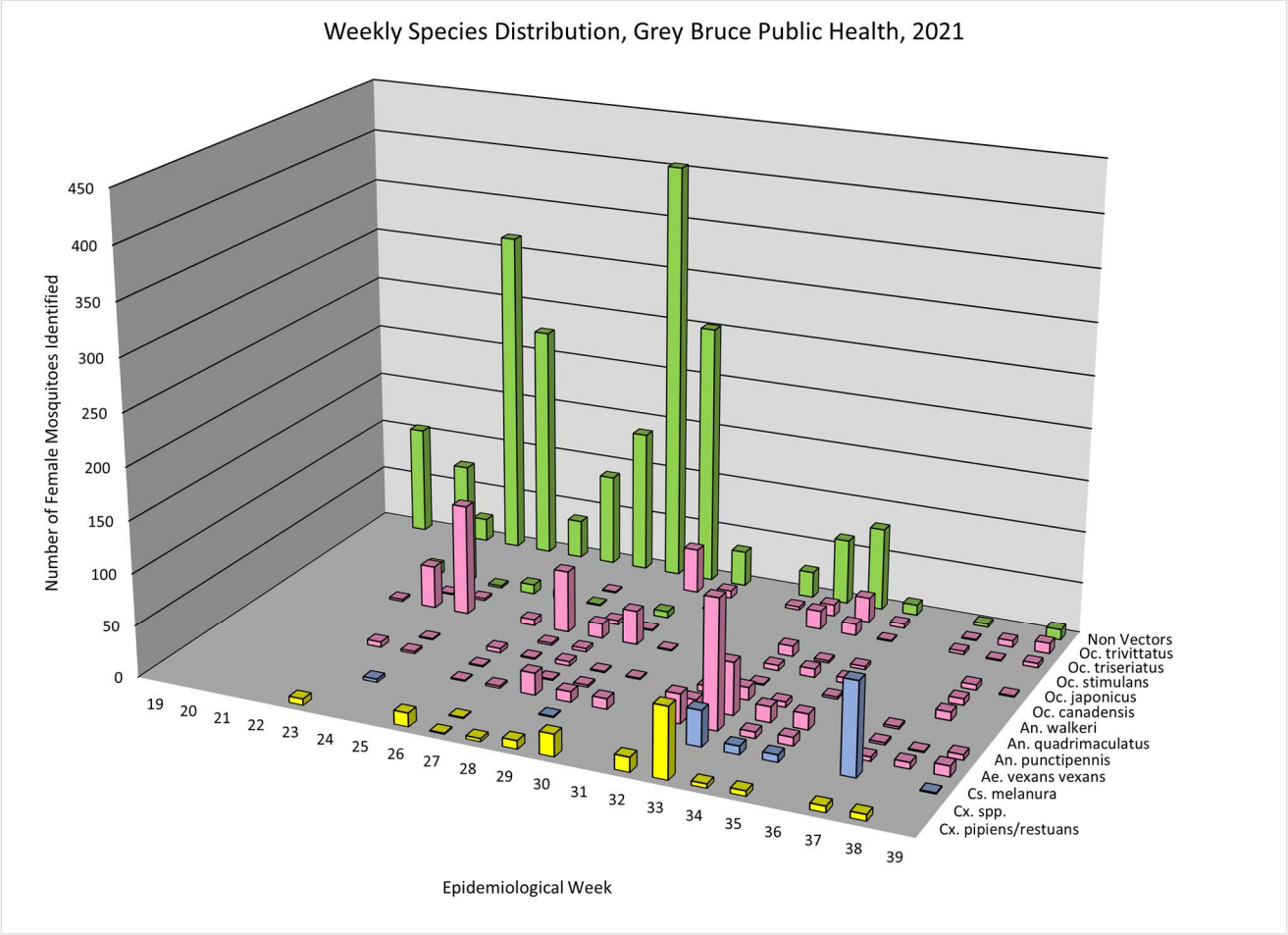

Figure 15. Species Distribution for Grey Bruce Public Health, 2021. Yellow bars represent bird-biting (WNV enzootic vector) species, pink bars represent WNV bridge vectors, and green bars represent non-vector species.

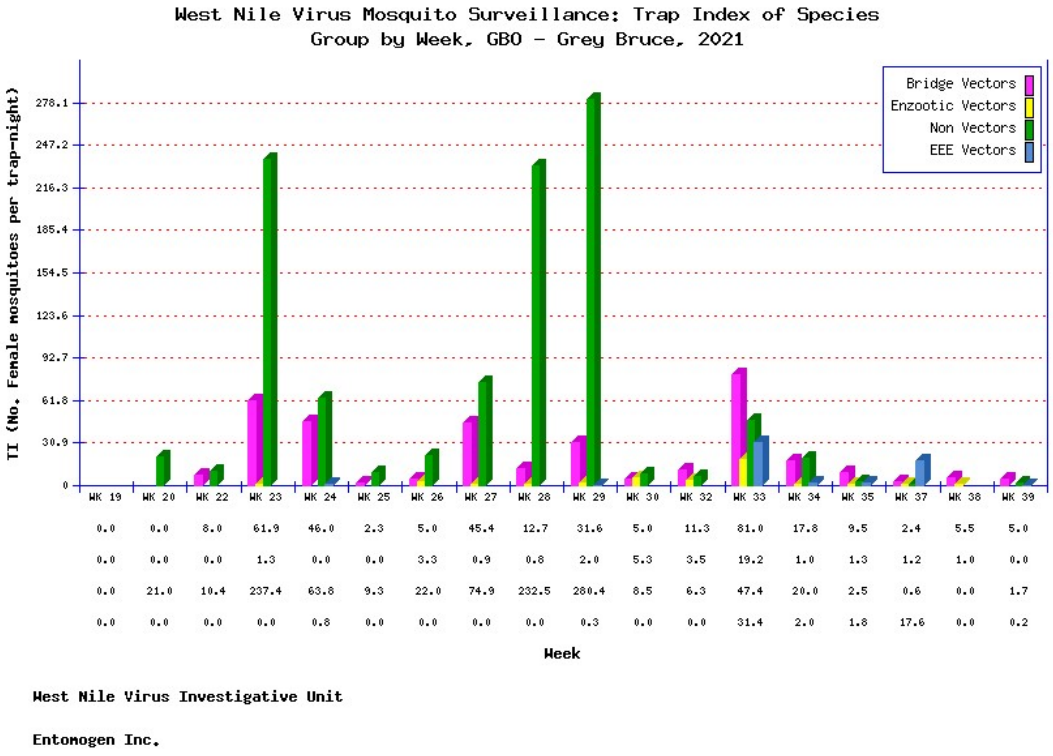

Figure 16. Trap Indices of WNV Enzootic Vectors (yellow), WNV Bridge Vectors (pink) and Non Vectors (green), Grey Bruce Public Health, 2021

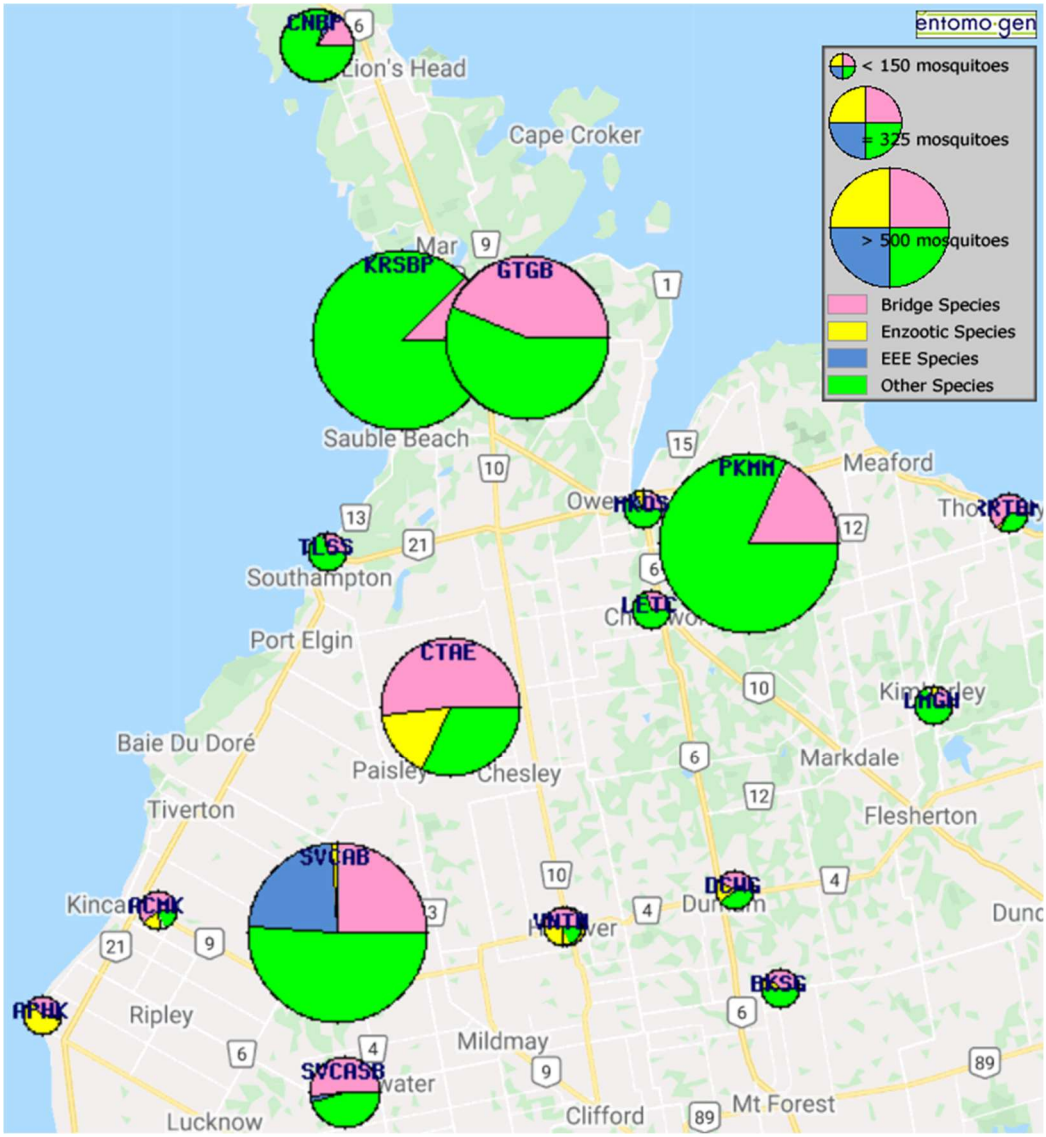

Figure 17. Grey Bruce Public Health Key Species Distribution Map, 2021

11.0 SUMMARY

The percentage of *Culex pipiens/restuans*, the main enzootic mosquito complex, was determined to be 4.94% of the total mosquito population in the 2021 season.

The threshold value of 380 accumulated degree-days was not crossed and there were no WNV human, avian or mosquito cases reported in Grey Bruce Public Health. Temperatures were slightly warmer in the province of Ontario in 2021; however, an absence of WNV activity within its borders would put Grey Bruce Public Health at **low** risk for endemic WNV transmission to humans.

In 2021, there were seven (7) EEEV-positive horses and six (6) EEEV-positive mosquito pools reported in Ontario. None of them were reported within Grey Bruce Public Health; however, a significant number (n=142) of the main EEEV enzootic vector (*Culiseta melanura*) were collected from traps set in 2021. Although there was a significant enzootic vector population the lack EEEV activity locally would put Grey Bruce Public Health at a **low** risk for endemic EEEV transmission to humans.

12.0 RECOMMENDATIONS

Temperatures in 2021 were only slightly warmer than 2020, however yearly global average temperatures continue to increase which puts more northern health units at risk for potential WNV infections.

In 2016, *Ae. albopictus* and *Ae. aegypti* – two exotic species that are vectors of many diseases including dengue, zika and chikungunya - were discovered during routine surveillance in Windsor-Essex County. These findings demonstrate the importance for continued mosquito surveillance activities, not only for virus detection, but also to monitor range expansion and potential introduction of invasive species.

WNV positive humans, bird and mosquitoes were confirmed in Ontario in 2021, therefore the trapping and viral testing program should be continued at the same intensity in 2022.

The main EEEV enzootic vector *Culiseta melanura* was identified in Grey Bruce Public Health in 2021. In addition, EEEV bridge vector species - *Ochlerotatus canadensis*, *Aedes vexans vexans* and *Coquillettidia perturbans* – made up approximately 47% of all mosquito species collected in Grey Bruce Public Health in 2021. Although no EEEV activity was reported within the health unit, there were positive mosquito pools and equine cases reported in the province of Ontario in 2021. Grey Bruce Public Health may want to consider more targeted trapping, and/or expanding EEEV testing to include bridge vector species in 2022.

13.0 ACKNOWLEDGMENTS

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14.0 APPENDICES

APPENDIX A – TRAP NUMBERS FOR THE EIGHTEEN SITES IN GREY BRUCE PUBLIC HEALTH

Site Code	Total Mosquitoes Identified	Number of Extras	Number of Traps	Latitude	Longitude	City
ACMK	34	0	5	44.165398	-81.59069	Kincardine
APHK	22	0	5	44.062466	-81.746269	Ripley
BKSG	92	0	5	44.088457	-80.7407	Holstein
CNBP	235	0	5	45.00355	-81.37013	Stoke's Bay
CTAE	328	75	5	44.363441	-81.192861	Arran-Elderslie
DCWG	108	0	5	44.183871	-80.8038	Durham
GTGB	330	130	5	44.721105	-81.088459	Wiarton
KRSBP	314	500	5	44.719218	-81.258703	Wiarton
LETC	61	0	5	44.45699	-80.915767	Chatsworth
LMGH	135	0	5	44.365889	-80.5326	Flesherton
MKOS	106	0	5	44.5548	-80.929142	Owen Sound
PKMM	297	900	5	44.521345	-80.785348	Bognor
RRTBM	123	0	5	44.55129	-80.429848	Thornbury
SVCAB	522	1900	5	44.143721	-81.3461	Brockton
SVCASB	232	0	5	43.987853	-81.3354	Teesewater
TAH	2	0	1	44.148964	-81.0316	Hanover
TLSS	127	0	5	44.513351	-81.36029	Southhampton
VNTH	49	0	4	44.14907	-81.037	Hanover

APPENDIX B – NUMBER AND PERCENTAGES OF IDENTIFIED MOSQUITOES SEPARATED INTO RESPECTIVE GROUPS IN GREY BRUCE PUBLIC HEALTH

Site Code	WNV Enzootic Vectors	WNV Bridge Vectors	EEEV Vectors	Non Vectors	Total Mosquitoes Identified
ACMK	5 (14.7%)	21 (61.8%)	0 (0.0%)	8 (23.5%)	34
APHK	13 (59.1%)	8 (36.4%)	0 (0.0%)	1 (4.5%)	22
BKSG	9 (9.8%)	36 (39.1%)	0 (0.0%)	47 (51.1%)	92
CNBP	1 (0.4%)	36 (15.3%)	9 (3.8%)	189 (80.4%)	235
CTAE	53 (16.2%)	169 (51.5%)	1 (0.3%)	105 (32.0%)	328
DCWG	21 (19.4%)	45 (41.7%)	0 (0.0%)	42 (38.9%)	108
GTGB	2 (0.6%)	142 (43.0%)	3 (0.9%)	183 (55.5%)	330
KRSBP	1 (0.3%)	39 (12.4%)	0 (0.0%)	274 (87.3%)	314
LETC	0 (0.0%)	19 (31.1%)	0 (0.0%)	42 (68.9%)	61
LMGH	8 (5.9%)	29 (21.5%)	0 (0.0%)	98 (72.6%)	135
MKOS	11 (10.4%)	26 (24.5%)	8 (7.5%)	61 (57.5%)	106
PKMM	1 (0.3%)	53 (17.8%)	0 (0.0%)	243 (81.8%)	297
RRTBM	3 (2.4%)	79 (64.2%)	0 (0.0%)	41 (33.3%)	123
SVCAB	9 (1.7%)	128 (24.5%)	116 (22.2%)	269 (51.5%)	522
SVCASB	1 (0.4%)	121 (52.2%)	5 (2.2%)	105 (45.3%)	232
TAH	1 (50.0%)	1 (50.0%)	0 (0.0%)	0 (0.0%)	2
TLSS	3 (2.4%)	34 (26.8%)	0 (0.0%)	90 (70.9%)	127
VNTH	13 (26.5%)	24 (49.0%)	0 (0.0%)	12 (24.5%)	49

APPENDIX C – WEST NILE VIRUS AND EASTERN EQUINE ENCEPHALITIS VIRUS VIRAL TESTING ORDER OF PREFERENCE

Ontario Ministry of Health and Long-Term Care Mosquito Species for Viral Testing	
1	*Culex pipiens/restuans (WNV)*
2	*Culex salinarius (WNV)*
3	*Ochlerotatus japonicus (WNV)*
4	Culex tarsalis (WNV)
5	*Aedes vexans vexans/ Aedes vexans nipponi (WNV)*
6	*Ochlerotatus triseriatus (WNV)*
7	*Anopheles punctipennis(WNV)*
8	*Ochlerotatus trivittatus (WNV)*
9	*Anopheles walkeri (WNV)*
10	*Ochlerotatus stimulans (WNV)*
11	*Anopheles quadrimaculatus (WNV)*
12	*Ochlerotatus canadensis (WNV)*
*	*Culiseta melanura (EEEV)*
**	*Aedes albopictus/Stegomyia albopicta (WNV)*
* Since this species is found in low numbers and is the main enzootic vector for EEEV, it is to be tested for EEEV as part of the three pool limit	
** Since this species is found in very low numbers and is a highly competent vector, it is suggestd that it be tested for WNV as part of the three pool limit	

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