



**National Bee
Diagnostic Centre**
Technology Access Centre

Varrroa photo by Wendy Smith



NATIONAL HONEY BEE HEALTH SURVEY

2014 Alberta & Manitoba Data

2014 REPORT

The National Honey Bee Health Survey began in the summer of 2014 and continues until March 2018. The goal of the project is to establish a baseline of the overall health of honey bees in Canada over a four year period. All of the diagnostic tests in this report were performed at the National Bee Diagnostic Centre – Technology Access Centre in Beaverlodge, Alberta. As the project grows, samples from all Canadian provinces are collected.

2014 National Honey Bee Health Survey Results

Data represents samples collected in Alberta and Manitoba for year one of the study.

Report Date: April 30, 2015

In the following pages, you will find the visual inspection and diagnostic results for Alberta and Manitoba. 163 composite samples were collected from the apiaries of participating producers during the summer of 2014. The report includes the diagnostic results for Nosema, Viruses, EFB, AFB, Tracheal Mites, Varroa Mites & visual analysis for *Tropilaelaps spp.* mites.

We thank the associations, provincial apiculturists and the beekeepers for their participation and support for the National Honey Bee Health Survey project. The knowledge generated from this study will help to identify pathogen trends occurring within provincial regions, between regions, provinces as well as at a national level.

If you have any questions, please do not hesitate to contact us.

Sincerely,



Dr. Carlos Castillo

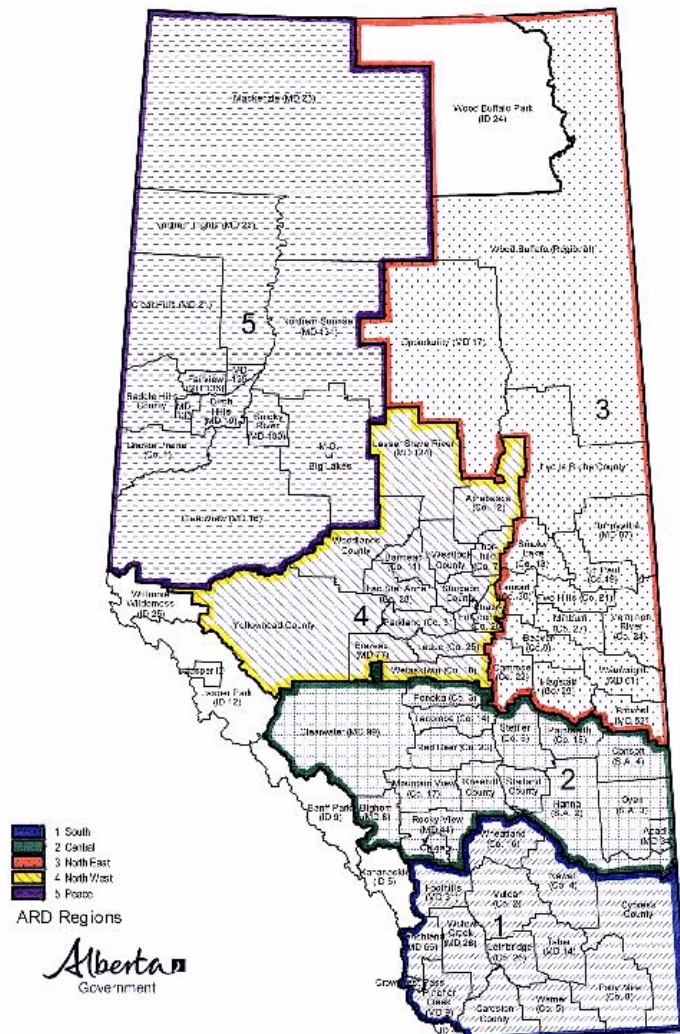
Applied Research Scientist

National Bee Diagnostic Centre – Technology Access Centre

Grande Prairie Regional College

National Honey Bee Health Survey 2014 Results for Alberta

In 2014, samples were taken from 123 apiaries in Alberta, representing 1,230 colonies. The following results were obtained from three types of composite samples that were collected from ten randomly-chosen colonies in each apiary of participating producers, during the summer of 2014. A live bee sample was collected and shipped directly to the NBDC-TAC. This sample was immediately put into an ultra-low temperature freezer to maintain the integrity of the RNA and DNA for further disease and pest analysis using molecular biology techniques. A second sample was collected, in which the bees were submerged in 70% ethyl alcohol to determine *Varroa* mite levels. The third sample consisted of material collected from the “knock test” of a brood frame, and was used to search for the presence of *Tropilaelaps spp.* mites.



Visual Inspection Results

For the visual inspection of each of the 10 colonies within the apiary, the three central brood frames were examined for potential disease symptoms. When examining colonies for the presence of queen cells or drone-laying queens, these conditions were scored as either being present or absent. The following chart reports the presence or absence of the diseases or conditions inspected for, as well as the regional and provincial averages.

Disease/Condition Averages	Peace Region	Northwest Region	Northeast Region	Central Region	South Region	Provincial Average
AFB	1.2%	0%	1.3%	3.1%	0%	0.45%
EFB	0%	0%	0%	0.8%	1.3%	0.17%
Sacbrood	0.6%	0%	0.7%	0%	0.5%	0.14%
Chalkbrood	10.0%	3%	12.7%	5.4%	7.1%	3.11%
Deformed Wing Bees	1.5%	0.4%	1.3%	1.5%	0%	0.39%
Black Shiny Bees	0%	0.4%	0%	0%	0%	0.04%
Small Hive Beetle (larvae or adult)	0%	0%	0%	0%	0%	0%
Wax Moth (larvae or adult)	0%	0%	0%	0%	0%	0%
Queen Cells Present	8.2%	5.7%	5.3%	14.6%	0.5%	2.79%
Drone Laying Queen	0.9%	1.3%	2%	3.1%	0.5%	0.63%

Nosema Results (Counting & Identification)

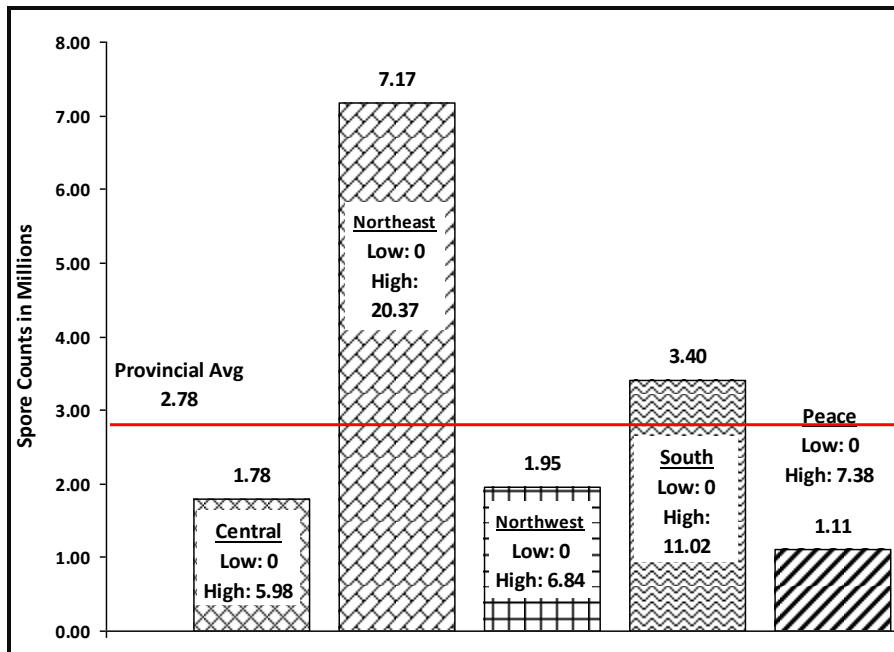
Sixty bees from the frozen live bee sample were macerated and analyzed for *Nosema* spp. infections. The macerated sample was examined using a Hemocytometer under a light microscope to obtain a *Nosema* spore count. The DNA was then extracted and a PCR (Polymerase Chain Reaction) amplification was carried out to identify the species of *Nosema* present in the sample (*N.apis*, *N.ceranae*, or both *N.apis* & *N.ceranae* or negatives).

AB Provincial *Nosema* Spore Count Average = 2.78 million spores/bee

Regional *Nosema* Spore Count Averages:

- Central Region = 1.78 million spores/bee
- Northeast Region = 7.17 million spores/bee
- Northwest Region = 1.95 million spores/bee
- South Region = 3.40 million spores/bee
- Peace Region = 1.11 million spores/bee

Average *Nosema* Spores / Bee per Alberta Region and Apiary



EFB Results (PCR Detection)

For the detection of EFB (*Melissococcus plutonius*), DNA was extracted from the frozen bee sample and a PCR amplification was carried out. PCR is a very sensitive detection technique which amplifies even the tiniest amount of DNA of the target organism. Please note that a positive result for EFB found through PCR does not necessarily mean that colonies within the tested apiary will exhibit clinical symptoms of an active EFB infection.

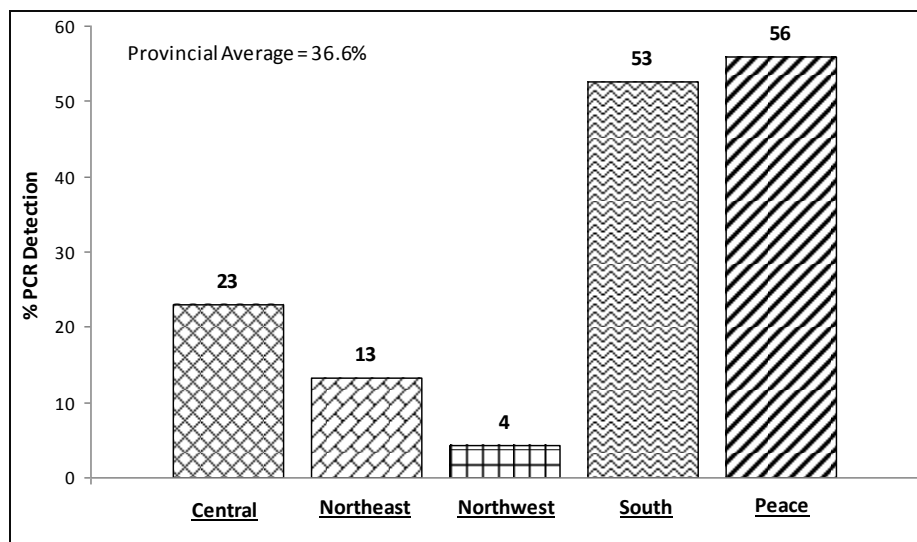
Across the Province of Alberta, only 6 colonies out of 123 samples were found to have visual clinical symptoms of EFB; all other positive results were detected through PCR methods. This suggests that many colonies in the province have low levels of the EFB pathogen present that can only be detected by molecular methods. Further research is necessary to understand positive detections of EFB in the lab with clinical symptoms in an apiary.

AB Provincial Average of Apiary-Level Positives for EFB by PCR Detection = 36.6%

Regional Average of Apiary-level EFB Positives by PCR Detection:

- Central Region = 23%
- Northeast Region = 13%
- Northwest Region = 4%
- South Region = 53%
- Peace Region = 56%

Detection of EFB in Alberta by PCR



Varroa Results

For the Varroa mite analysis, all bees (over 1,000) from the adult bee samples collected in ethanol, were processed in the laboratory to dislodge the mites. These “washes” provide the infestation level of the apiary, expressed in percentage (number of mites per 100 adult bees).

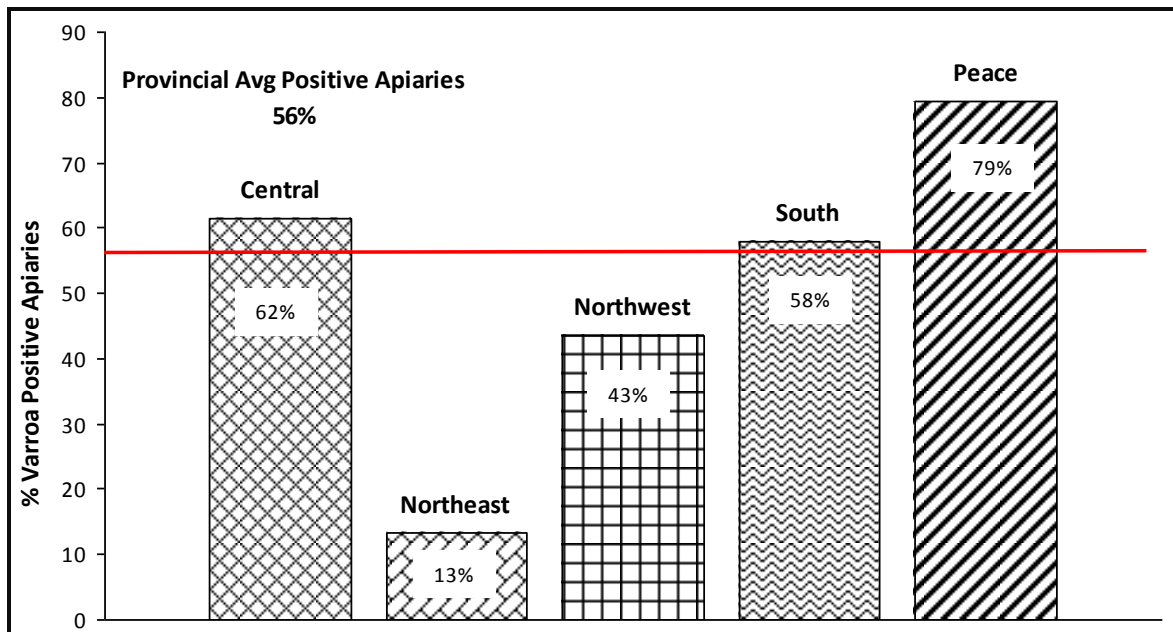
AB Provincial Average for Apiary-Level Detections for Varroa (any level of incidence) = 56% (Varroa was detected in 56% of apiaries we tested in Alberta).

AB Provincial Average for Varroa Infection Level per Apiary= 0.77%

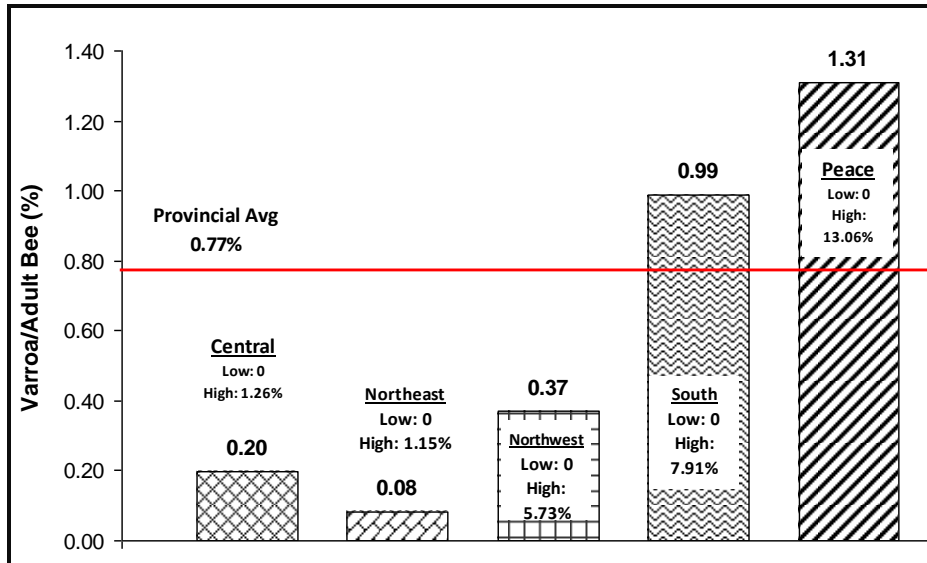
Regional Apiary-Level Averages of Varroa Infection Level:

- Central Region = 62% & 0.2%
- Northeast Region = 13% & 0.08%
- Northwest Region = 43% & 0.37%
- South Region = 58% & 0.99%
- Peace Region = 79% & 1.31%

% Varroa Positive Apiaries per Alberta Regions



% Varroa Infection Level per Region



Tracheal Mite Results (by PCR & Dissection)

DNA extracted from the frozen Live Bee sample was analyzed using PCR amplification to detect the DNA of Tracheal Mite (*Acarapis woodi*). If a positive detection occurred, 16 bees from the apiary sample were then dissected for tracheal mite identification.

NO Tracheal mites were found in the Alberta samples.

Tropilaelaps Results (Visual Analysis of “Knock” Sample)

The third sample that was taken in the field, by knocking an unsealed brood frame into a metal collection pan and analyzing the debris, was used to search for the presence of *Tropilaelaps* spp. mites. Samples were carefully examined under a dissecting microscope.

Tropilaelaps spp. mites are an exotic parasitic mite found in Asia is not known to occur in Canada. These mites have a quicker reproductive cycle, they can out produce Varroa mites.

No *Tropilaelaps* spp. were found in the Alberta samples.

AFB Results (by Bacterial Culture)

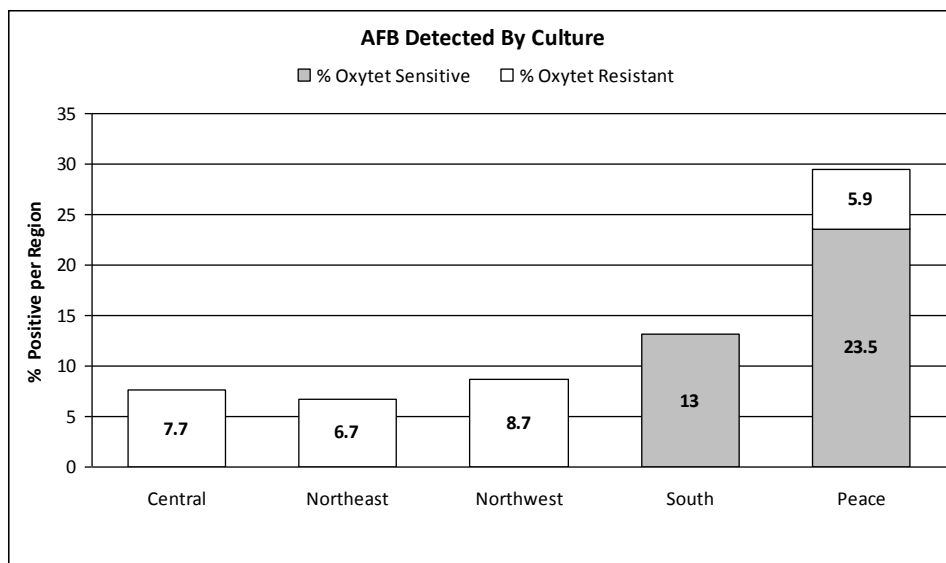
One hundred and twenty bees from each frozen bee sample per apiary were tested for the presence or absence of *Paenibacillus larvae*, the bacterium that causes AFB. Each sample was cultivated in triplicate on diagnostic media plates that supported the growth of the bacterium. For each test, the number of bacterial colonies that grew on the media were scored as the number of colony forming units (CFU). If AFB grew from a sample, bacterial colonies were then tested for resistance or sensitivity towards Oxytetracycline (oxytet) and Tylosin, which are registered antibiotics used for the control of AFB.

AB Provincial Average for Positive Detections of AFB by Culture = 15.4%

Regional Averages for Positive Detections of AFB by Culture:

- Central Region = 7.7% (1 of 13 samples tested positive for AFB)
- Northeast Region = 6.7% (1 of 15 samples tested positive for AFB)
- Northwest Region = 8.7% (2 of 23 samples tested positive for AFB)
- South Region = 13% (5 of 38 samples tested positive for AFB)
- Peace Region = 29.4% (10 of 34 samples tested positive for AFB)

The graph below shows the positive AFB samples that were sensitive or resistant to Oxytetracycline. Throughout Alberta, all samples in which AFB could be cultured were sensitive to Tylosin, meaning that the drug will control the disease.

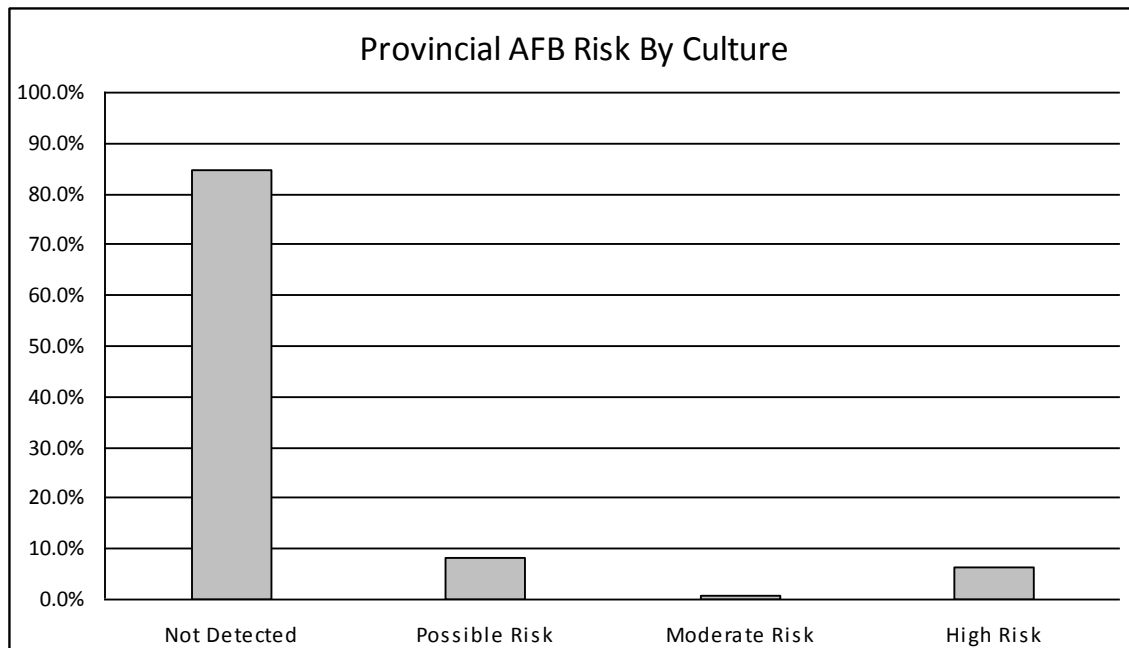


AFB Risk (by Bacterial Culture)

Based on previous research, apiaries were categorized into 4 nominal groups based on their propensity to exhibit clinical symptoms of the disease. These risk categories are designated based on the average number of bacterial colony forming units (CFUs) that were cultivated on diagnostic media plates: Not Detected, A Possible Risk is defined as any apiary with (1-99 CFUs), Moderate Risk (100-999 CFUs) and High Risk (>1,000 CFUs).

Provincial Breakdown of the AFB risk by culture:

- Not Detected = 84.6%
- Possible Risk = 8.1%
- Moderate Risk = 0.8%
- High Risk = 6.5%



Virus Detection

Fifty bees from the frozen live bee sample were macerated and analyzed for the detection of the following 7 viruses: ABPV (Acute Bee Paralysis Virus), BQCV (Black Queen Cell Virus), CBPV (Chronic Bee Paralysis Virus), DWV (Deformed Wing Virus), KBV (Kashmir Bee Virus), IAPV (Israeli Acute Paralysis Virus) & SBV (Sacbrood Virus). The apiaries were scored as “Positive” for detection of any level of the virus or “Negative” for the absence of the virus.

IAPV Israeli Acute Paralysis Virus, common in some regions, has been associated with colony losses

KBV Kashmir Bee Virus, uncommon, has been associated with colony losses

DWV Deformed wing virus, very common, associated with colony losses

ABPV Acute Bee Paralysis Virus, rare, has been associated with colony losses

CBPV Chronic Bee Paralysis Virus, rare

SBV Sac Brood Virus, very common in Canada

BQCV Black Queen Cell Virus, very common, may be associated with *Nosema* disease

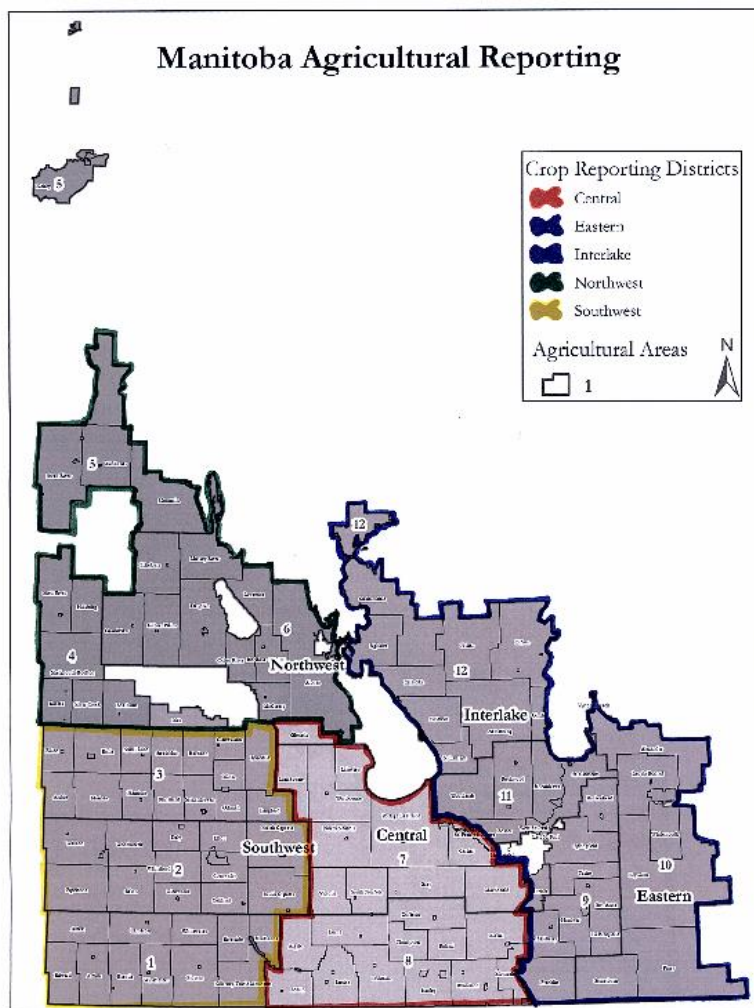
Virus	APIARY-LEVEL VIRAL DETECTIONS BY REGION					Provincial Average
	Central	South	Northeast	Northwest	Peace	
ABPV	0%	24%	13%	0%	12%	12%
BQCV	23%	66%	40%	30%	23%	40%
CBPV	8%	0%	7%	0%	0%	2%
DWV	23%	26%	33%	30%	26%	28%
KBV	0%	0%	0%	0%	0%	0%
IAPV	77%	66%	47%	30%	29%	48%
SBV	92%	100%	77%	96%	68%	86%

Detection of individual viruses may not be indicative of a problem. Multiple viruses could be a sign of colony health issues in the apiary, however the impact of multiple viruses on productivity and colony health is still being researched.

If you have any questions regarding the National Honey Bee Health Survey Alberta results, please feel free to contact Dr. Carlos Castillo at the National Bee Diagnostic Centre - TAC 780-357-7737 or ccastillo@gprc.ab.ca

National Honey Bee Health Survey 2014 Results for Manitoba

In 2014, 40 samples were taken from Manitoba apiaries, representing 400 colonies. The following results were obtained from three types of composite samples that were collected from ten randomly-chosen colonies in each apiary of participating producers, during the summer of 2014. A live bee sample was collected and shipped directly to the NBDC-TAC. This sample was immediately put into an ultra-low temperature freezer to maintain the integrity of the RNA and DNA for further disease and pest analysis using molecular biology techniques. A second sample was collected, in which the bees were submerged in 70% ethyl alcohol to determine Varroa mite levels. The third sample consisted of material collected from the “knock test” of a brood frame, and was used to search for the presence of *Tropilaelaps spp.* mites.



Visual Inspection Results

For the visual inspection of each of the 10 colonies within the apiary, the three central brood frames were examined for potential disease symptoms. When examining colonies for the presence of queen cells or drone-laying queens, these conditions were scored as either being present or absent. The following chart reports the presence or absence of the diseases or conditions inspected for, as well as the regional and provincial averages.

Disease/Condition Averages	Northwest Region	Southwest Region	Central Region	Eastern Region	Provincial Average
AFB	10%	0%	0%	0%	2.5%
EFB	0%	0%	0%	0%	0%
Sacbrood	1%	0%	0%	0%	0.25%
Chalkbrood	7%	1%	6%	5%	4.75%
Deformed Wing Bees	0%	0%	0%	0%	0%
Black Shiny Bees	0%	0%	0%	0%	0%
Small Hive Beetle (larvae or adult)	0%	0%	0%	0%	0%
Wax Moth (larvae or adult)	0%	0%	0%	0%	0%
Queen Cells Present	12%	9%	10%	28%	14.75%
Drone Laying Queen	1%	1%	2%	0%	1%

***Nosema* Results (Counting & Identification)**

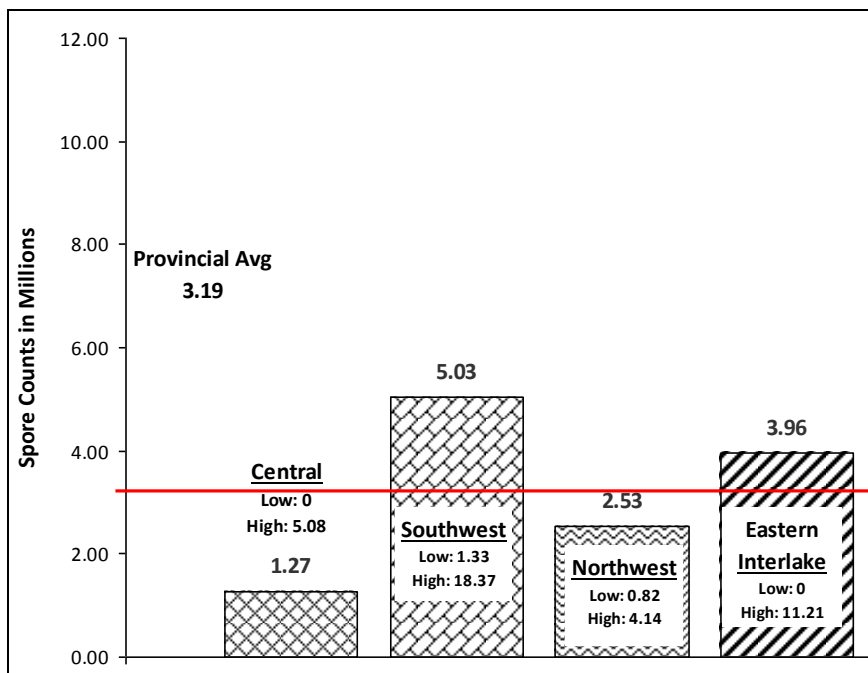
Sixty bees from the frozen live bee sample were macerated and analyzed for *Nosema* spp. infections. The macerated sample was examined using a Hemocytometer under a light microscope to obtain a *Nosema* spore count. The DNA was then extracted and a PCR (Polymerase Chain Reaction) amplification was carried out to identify the species of *Nosema* present in the sample (*N.apis*, *N.ceranae*, or both *N.apis* & *N.ceranae* or negatives).

MB Provincial *Nosema* Spore Count Average = 3.19 million spores/bee

Regional *Nosema* Spore Count Averages:

- Central Region = 1.27 million spores/bee
- Southwest Region = 5.03 million spores/bee
- Northwest Region = 2.53 million spores/bee
- Eastern-Interlake Region = 3.96 million spores/bee

Average *Nosema* Spore Count per Region



EFB Results (PCR Detection)

For the detection of EFB (*Melissococcus plutonius*), DNA was extracted from the frozen bee sample and a PCR protocol was carried out. PCR is a very sensitive detection technique which amplifies even the tiniest amount of DNA of the target organism. Please note that a positive result for EFB found through PCR does not necessarily mean that colonies within the apiary tested will exhibit clinical symptoms of an active EFB infection. Visually, no symptoms of EFB were seen in Manitoba; positive results were only detected through PCR methods.

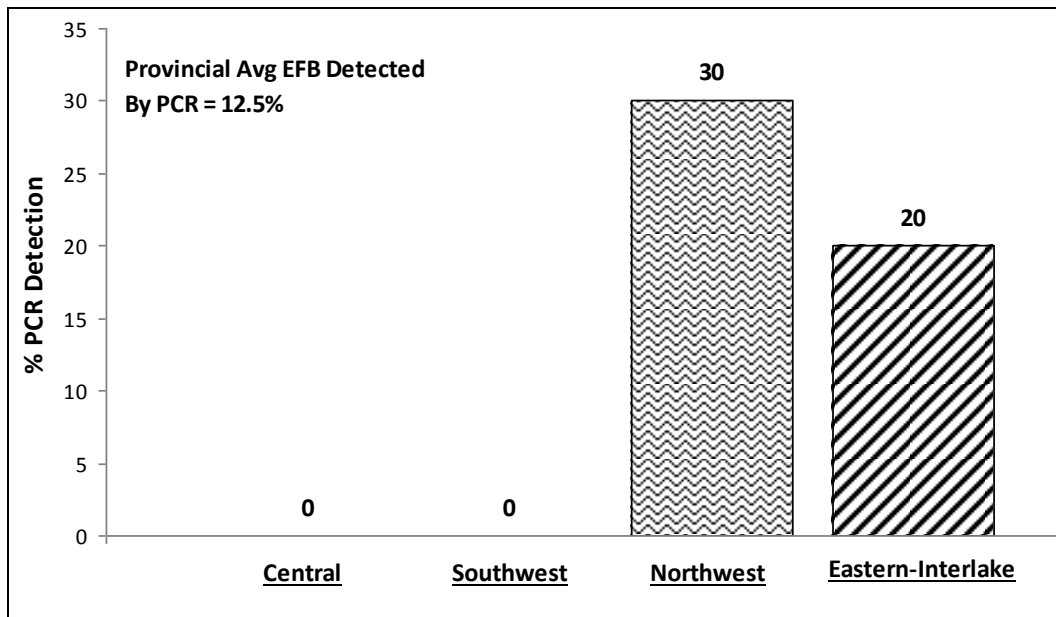
This suggests that colonies in the province have low levels of the EFB pathogen present that can only be detected by molecular methods. Further research is necessary to understand positive detections of EFB in the lab with clinical symptoms in an apiary.

MB Provincial EFB Positive PCR Detection = 12.5%

Regional EFB Positive PCR Detection Averages:

- Central Region = 0%
- Southwest Region = 0%
- Northwest Region = 30%
- Eastern-Interlake Region = 20%

Detection of EFB in Manitoba by PCR



Varroa Results

For the Varroa mite analysis, all bees (over 1,000) from the adult bee samples collected in ethanol, were processed in the laboratory to dislodge the mites. These “washes” provide the infestation level of the apiary, expressed in percentage (number of mites per 100 adult bees).

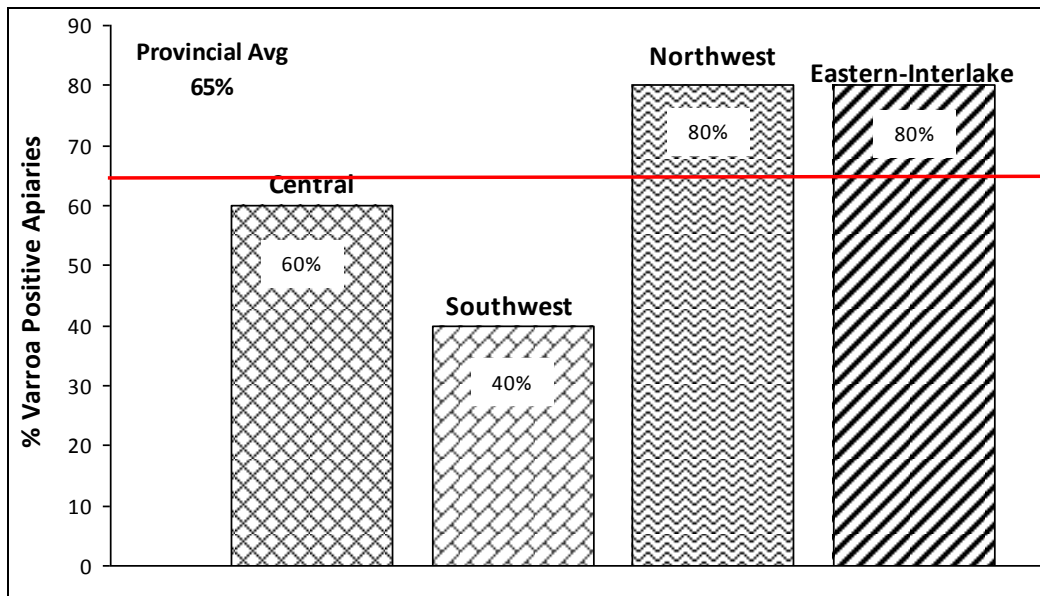
MB Provincial Varroa Positive Average Detection for Varroa (any level of incidence) = 65%
 (Varroa was detected in 65% of apiaries in Manitoba)

MB Provincial Varroa Average Infection Level = 0.67%

Regional Apiary-Level Averages of Varroa Detection and Infection Level:

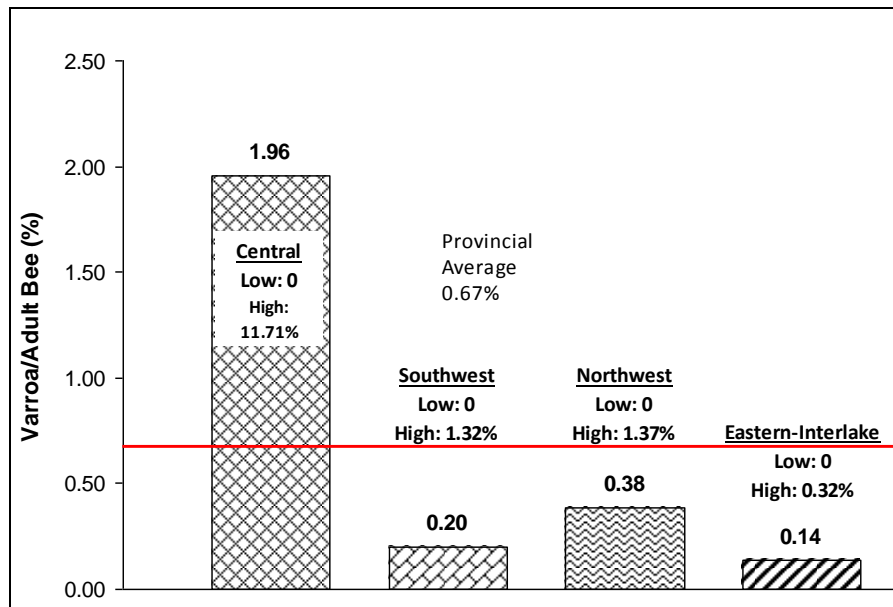
- Central Region = 60% & 1.96%
- Southwest Region = 40% & 0.2%
- Northwest Region = 80% & 0.38%
- Eastern-Interlake Region = 80% & 0.14%

% Varroa Positive Apiaries per Manitoba Regions



Varroa Results

% Varroa Infection Level per Manitoba Region



Tracheal Mite Results (by PCR & Dissection)

DNA extracted from the frozen Live Bee sample was analyzed using PCR amplification to detect the DNA of Tracheal Mite (*Acarapis woodi*). If a positive detection occurred, 16 bees from the apiary sample were then dissected for tracheal mite identification.

NO Tracheal mites were found in the Manitoba samples.

Tropilaelaps Results (Visual Analysis of “Knock” Sample)

The third sample that was taken in the field, by knocking an unsealed brood frame into a metal collection pan and analyzing the debris, was used to search for the presence of *Tropilaelaps* spp. mites. Samples were carefully examined under a dissecting microscope.

Tropilaelaps mites are an exotic parasitic mite found in Asia and are not known to occur in Canada. These mites have a quicker reproductive cycle, they can out produce Varroa mites.

No *Tropilaelaps* spp. specimens were found in the Manitoba samples.

AFB Results (by Bacterial Culture)

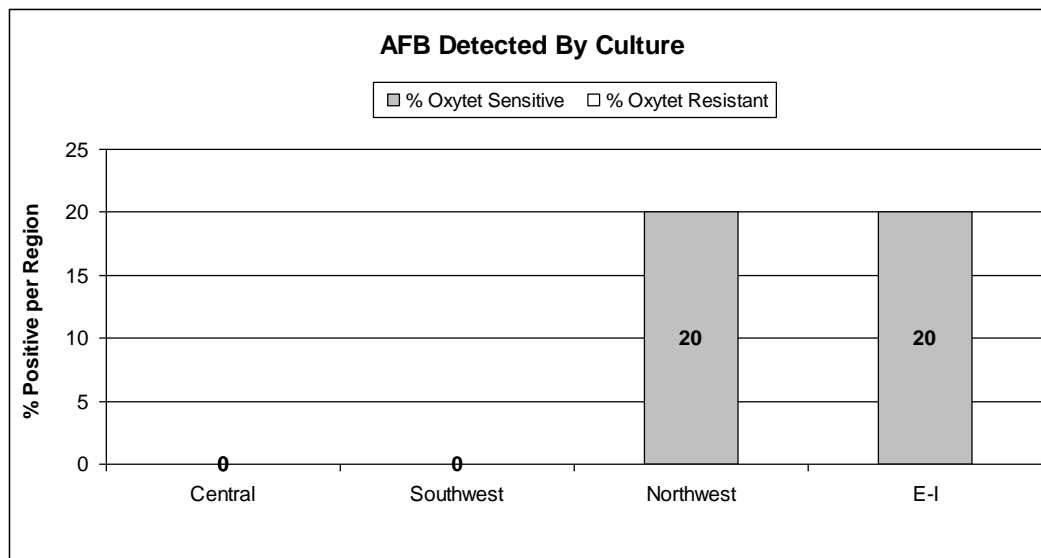
One hundred and twenty bees from each frozen bee sample per apiary were tested for the presence or absence of *Paenibacillus larvae*, the bacterium that causes AFB. Each sample was cultivated in triplicate on diagnostic media plates that supported the growth of the bacterium. For each test, the number of bacterial colonies that grew on the media were scored as the number of colony forming units (CFU). If grew from a sample, bacterial colonies were then tested for resistance or sensitivity towards Oxytetracycline (oxytet) and Tylosin, which are registered antibiotics used for the control of AFB.

MB Provincial AFB Positive Average Detection = 10%

Regional AFB Positive Detection Averages:

- Northwest Region = 20%
- Southwest Region = 0%
- Central Region = 0%
- Eastern-Interlake Region = 20%

The graph below shows the AFB positive samples that are sensitive or resistant to Oxytetracycline. For the province of Manitoba, all positives were sensitive to Oxytetracycline. All the AFB positive samples were sensitive to Tylosin, meaning that the drug will control the disease.

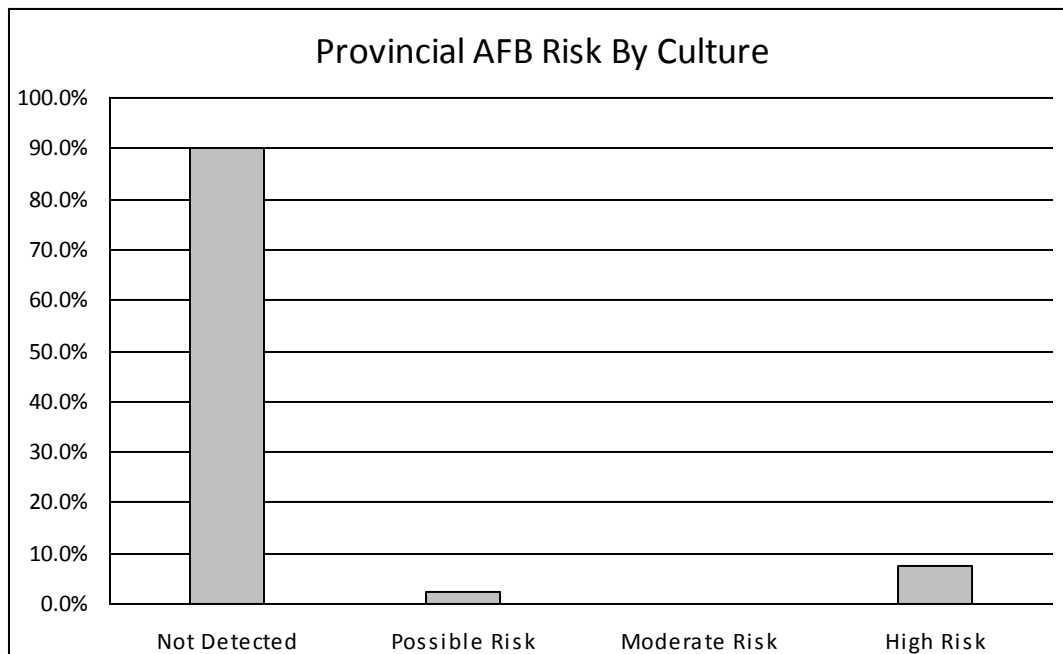


AFB Risk (by Bacterial Culture)

Based on previous research, apiaries were categorized into 4 nominal groups based on their propensity to exhibit clinical symptoms of the disease. These risk categories are designated based on the average number of bacterial colony forming units (CFUs) that were cultivated on diagnostic media plates: Not Detected, A Possible Risk is defined as any apiary with (1-99 CFUs), Moderate Risk (100-999 CFUs) and High Risk (>1,000 CFUs).

Provincial Breakdown of the AFB risk by culture:

- Not Detected = 90%
- Possible Risk = 2.5%
- Moderate Risk = 0%
- High Risk = 7.5%



Virus Detection

Fifty bees from the frozen live bee sample were macerated and analyzed for the detection of the following 7 viruses: ABPV (Acute Bee Paralysis Virus), BQCV (Black Queen Cell Virus), CBPV (Chronic Bee Paralysis Virus), DWV (Deformed Wing Virus), KBV (Kashmir Bee Virus), IAPV (Israeli Acute Paralysis Virus) & SBV (Sacbrood Virus). The apiaries were scored as “Positive” for detection of any level of the virus or “Negative” for the absence of the virus.

IAPV Israeli Acute Paralysis Virus, common in some regions, has been associated with colony losses

KBV Kashmir Bee Virus, uncommon, has been associated with colony losses

DWV Deformed wing virus, very common, associated with colony losses

ABPV Acute Bee Paralysis Virus, rare, has been associated with colony losses

CBPV Chronic Bee Paralysis Virus, rare

SBV Sac Brood Virus, very common in Canada

BQCV Black Queen Cell Virus, very common, may be associated with *Nosema* disease

Virus	POSITIVE REGIONAL AVERAGES				Provincial Average
	Central	Southwest	Northwest	Eastern Interlake	
ABPV	10%	20%	40%	30%	25.0%
BQCV	70%	70%	90%	90%	80.0%
CBPV	10%	20%	10%	30%	17.5%
DWV	60%	20%	60%	60%	50.0%
KBV	0%	0%	0%	0%	0.0%
IAPV	60%	60%	100%	80%	75.0%
SBV	90%	90%	100%	90%	92.5%

Detection of individual viruses may not be indicative of a problem. Multiple viruses could be a sign of colony health issues in the apiary, however the impact of multiple viruses on productivity and colony health is still being researched.

If you have any questions regarding the National Honey Bee Health Survey Manitoba results, please feel free to contact Dr. Carlos Castillo at the National Bee Diagnostic Centre-TAC 780-357-7737 or ccastillo@gprc.ab.ca