Nosemosis (*Nosema* disease) - Peter Armitage[1]

Editor's note: this is the first in a five part series discussing diseases that may affect honey bees in Newfoundland and Labrador. In future newsletters, we'll cover American Foulbrood, Chalkbrood, European Foulbrood, and Sacbrood Virus.

The germ theory of disease wasn't even an idea back in 1617 when honey bees were first brought to the Island of Newfoundland (Crane, 1999). We owe it to pioneering 19th century scientists like Robert Koch and Louis Pasteur to prove that infectious diseases are caused by germs, micro-organisms — bacteria and viruses. Not surprisingly, then, our knowledge of honey bee pathogens is a product of the bioscientific revolution. Gershom Franklin White is credited with groundbreaking pathogen research related to honey bees having demonstrated conclusively in 1907 that the bacterium *Bacillus larvae* is the cause of American Foulbrood (AFB) disease (White, 1907). *Nosema apis* was described by Enoch Zander in 1909, and Sacbrood Virus in 1913, again by White (Huang, 2011; Pernal and Clay, 2013, p.23).

The domestic Newfoundland and Labrador (NL) honey bee stock(s) has several apparently endemic pathogens, many if not all of which may have arrived with our beekeeping pioneers going back to the 1970s. Prior to the Canadian National Honey Bee Health (CNHBH) Survey in 2016 (NBDC, 2016), the domestic stock of honey bees in Newfoundland and Labrador (NL) was surveyed for various pathogens, pests and disease in 2004, 2007, 2009 (Williams, et al., 2010, p.585), and 2010 (Shutler, et al., 2014). Rogers' informal surveys in 2004 detected AFB, Chalkbrood, and European Foulbrood, but not Trachael mites or *Varroa destructor* mites (Williams, et al., 2010, p.585). *Nosema apis* was detected for the first time in 2007 (Williams, 2010, p.3). Additional testing in 2009 confirmed the presence of *Nosema apis* in the NL honey bee stock and the absence of Trachael and *Varroa* mites (Williams, et al., 2010, p.586).

The belief, therefore, that the domestic NL honey bee stock is "clean" is wrong. What sets this stock apart from the rest of North America, however, is the current absence of *Varroa destructor*, Wax Moth, Small Hive Beetle, and Tracheal Mite. The domestic NL stock appears also to be free of Acute Bee Paralysis Virus, Chronic Bee Paralysis Virus, Israeli Acute Paralysis Virus, and Kashmir Bee Virus, as well as AFB, although there have been outbreaks of the latter as recently as 15 years ago.

This article focuses on two common, closely related and potentially serious honey bee pathogens – *Nosema apis* and *Nosema ceranae*. Much has been written about these pathogens, and readers are referred to the beekeeping and scientific literature for more information (e.g., Dyce Lab for Honey Bee Studies, nd.; Fries, 2009, 1997; Huang, 2011; Pernal and Clay, 2013; Pettis, et al., 2015; Ritter, 2015; Sammataro and Avitabile, 2011).

Nosema apis and *Nosema ceranae*

Nosema spp. are a spore-forming fungi that invade the digestive epithelium (cell lining) of the honey bee midgut. The pathogen may remain as a "covert" or "inapparent" infection with no

[1] **Cautionary note.** Like many if not most matters apicultural, this topic is very complex. The author is a beekeeper who reads a lot, not an expert on honey bee pathogens or diseases. Please consult the relevant beekeeping and scientific literature for more detailed information. The references for this article are a good starting point. Thanks to George Carayanniotis, Catherine Dempsey and Dan Price for helpful comments on an earlier draft of this article.

clinical symptoms for years, and for that reason it is often overlooked by beekeepers. However, it may erupt into a severe "overt" infection (Nosemosis, *Nosema* disease) depending on a combination of factors, and may threaten colony survival. *Nosema* is transmitted horizontally among, and vertically within, colonies by way of trophallaxis (exchange of regurgitated liquids), common water sources and packaged or caged queens, in comb, honey and pollen, and by drifting and robbing foragers contacting fecal material on frames and combs in neighbouring hives.[2] Spores are viable in fecal material for more than a year, and it takes only a few of them to infect bees (Pernal and Clay, 2013, p.17; Sammataro and Avitabile, 2011, pp.190-191).

Nosema apis affects colonies mostly in later winter and spring when bees have been confined to the hive for lengthy periods of time, not able to leave on cleansing flights. Poor nutrition and cold, wet weather appear to aggravate the disease. *Nosema ceranae* infection peaks in the spring and early summer but is a risk to colonies throughout the year. The major difference between the two species is that heavy *Nosema ceranae* infections do not cause the fecal staining associated with *Nosema apis,* which is why it is sometimes referred to as the "dry *Nosema*" (Penn State Cooperative Extension, nd.). Furthermore, *Nosema ceranae* "can cause a bee colony to die within eight days after exposure, much faster than with *N. apis.* It appears to affect foraging bees the most, killing them while they are outside and leaving the home colony weak" (ibid., p.193). *Nosema apis* proliferates only in older bees, but *Nosema ceranae* will also proliferate in "shorter-living summer bees, and thus causes typical symptoms, especially crawling bees, throughout the year" (Ritter, 2015). For reasons that remain unclear, it appears that *Nosema ceranae* quickly becomes the dominant, more prevalent species wherever it becomes established (Pettis, et al., 2015, p.843; Ritter, 2015).

Testing of NL bees sampled in 2010 was negative for this species (Shutler, et al., 2014). However, the researchers who conducted the testing could not "state with absolute certainty that *N. ceranae* is absent in Newfoundland honey bees due to our initial positive detection in two colonies. Constant vigilance is therefore required" (ibid., p.7). Testing in 2016 as part of the CNHBH Survey confirmed the presence of *Nosema ceranae*.[3]

Symptoms and Diagnosis

The classic symptom of a full-blown, severe *Nosema apis* infection is fecal matter splattered all over the front of hives, landing boards and frames. However, there are a large number of additional symptoms that beekeepers should consider when compiling evidence for an outbreak of either of the two *Nosema* species. They are summarized in Table 1.

[2] In a somewhat outdated article about Nosemosis (ca. 2009), Randy Oliver noted that no one had yet discovered the means of transmission of *Nosema ceranae*, given that dysentery is not associated with infection by this species (Oliver, nd).

[3] *Nosema ceranae* was detected in samples from four of five apiaries in the survey (NBDC, 2016).

Table 1. Symptoms of *Nosema apis* and *Nosema ceranae* infection (from Dyce Lab for Honey Bee Studies, nd.; Penn State Cooperative Extension, nd.; Ritter, 2015; and Sammataro and Avitabile, 2011).

Nosema apis	*Nosema ceranae*
• Feces on combs, top bars, bottom boards, inside and outside walls of hives • Bee crawling aimlessly on bottom board, near entrance or on ground; some drag along as if their legs are paralyzed • Bees unable to fly or able to fly only short distances • K-wing • Swollen and greasy abdomens • Bees not eating when fed syrup • Reduced brood production • Queen supersedure • Reduced worker lifespan • Reduced honey production • Increased winter mortality • Reduced spring build up • Dwindling colony strength • Heavy winter losses	• Usually asymptomatic (i.e., no dysentery). • Crawling bees seen in front of the hive • Swollen and greasy abdomens • Queen supersedure • Reduced brood production • Reduced honey production • Decreases in colony strength can be slow or sudden • Bees often die while foraging • Early death of foragers • Bees not eating when fed syrup

Nosemosis can be hard to diagnose given that other diseases or colony conditions may have similar symptoms (e.g., dysentery), and the fact that colonies may have infections of both *Nosema* species simultaneously.[4] The standard method of testing colonies for *Nosema* infection, whether symptomatic or not, has been to examine and count spores using a microscope and hemocytometer. A spore count >1 million has been the threshold at which beekeepers would treat the disease.[5] However, symptoms of disease have been observed with far lower spore counts, and conversely no disease symptoms have been observed with far higher counts.[6] In the absence of diagnostic equipment, beekeepers sometimes attempt diagnosis by examining honey bee midguts, although this approach is unreliable. "If the midgut (ventriculus) is swollen and a dull grayish white, and the circular constrictions of the gut (similar to constrictions on an earthworm's body) are no longer evident, then nosema is the culprit….The normal gut is brownish red or yellowish, with many circular constrictions" (Sammataro and Avitabile, 2011, p.192).

[4] One of five apiaries tested positive for both species in the CNHBH Survey in 2016 (NBDC, 2017:20).
[5] The 2007 survey to investigate *Nosema* sampled 21 colonies of which 14 were positive for *Nosema* spp. Eleven colonies had bees with spore counts >1 million, with the bees in one colony having 7.5 million spores (Geoff Williams, personal communication, 28 Oct. 2016).
[6] Paul Kozak, Provincial Apiarist, Government of Ontario, personal communication, Sept. 8 Sept. 2017.

Heavy dysentery stains on the walls of a hive point to Nosemosis but are not a sure indicator of this disease (photo from Huang, 2011, p.3; see also Webster, 2010).

Nosema ceranae cannot be distinguished reliably from its cousin species without the application of molecular biology methods, namely, polymerase chain reaction (PCR) (OIE, 2013). To-date PCR has been able to determine reliably the presence (+) or absence (-) of the species. However, quantitative PCR (qPCR) that quantifies the amount of pathogen RNA in samples (e.g., 2.21E+09 copies/bee) is not supported by data allowing us to determine a threshold at which the infection is covert and therefore a serious health problem for a colony.

Nosemosis management

The scientific community and beekeepers have much to learn about *Nosema apis* and *Nosema ceranae* given their complex modes of infection, epidemiology and pathology. Nonetheless, beekeepers have developed a number of Integrated Pest Management practices to minimize disease outbreaks and control them when the need arises. These include the following (from Fries, 2009; Perennia, 2016; Pettis, et al., 2005, p.843; Pernal and Clay, 2013; Ritter, 2015; Sammataro and Avitabile, 2011, pp.193-194):

- Maintain strong colonies, and in the fall provide them with young, prolific queens, and a large population of young bees;
- Do not place hives on the ground to prevent ill bees, getting back into the hives (especially crawlers);
- Offer good forage to colonies to the greatest extent possible thereby preventing summer bees getting too old;
- Select good apiary locations with good sun exposure, protection from prevailing cold winds, and good ventilation (i.e., not damp spots that trap moisture);
- Insulate hives for winter and position them facing south to promote cleansing flights when weather permits;
- Provide good hive ventilation during winter (e.g., minimally an upper entrance);
- Supply colonies adequately with carbohydrates (honey, sugar syrup) and pollen or pollen substitute;
- Feed heavy sugar syrup (2:1) in the fall;

- Provide fresh, clean water to colonies;
- Dispose of comb that is soiled with fecal material, and/or treat combs of dead colonies with 60% acetic acid over a sponge cloth or some other absorbent material;[7]
- Given that *Nosema ceranae* is vulnerable to freezing, freeze comb from infected hives in a deep-freezer (~-18 deg. C) for at least one week.[8]

It is common practice across North America for beekeepers to monitor their colonies routinely for *Nosema* and use chemotherapy in the form of fumagillin to treat their bees when they get spore counts >1 million. Many beekeepers treat with fumagillin prophylactically, whether they have high spore counts and disease symptoms or not.[9]

In conclusion, it is reasonable to expect that news of pathogens and diseases in our domestic NL honey bees comes as a surprise to many novice beekeepers. However, our test results for *Nosema* should not be cause for alarm. They should be cause for vigilant monitoring and education so that we can learn how to prevent, identify and manage *Nosema* disease in the future. Let's work together on these matters for the benefit of our honey bees as well as ourselves as members of a supportive beekeeping community.

References

Crane, E. 1999. *The World History of Beekeeping and Honey Hunting*. London: Duckworth.

Dyce Lab for Honey Bee Studies. nd. Nosema Disease: Information for Identification and Control in New York. Cornell University. https://pollinator.cals.cornell.edu/sites/pollinator.cals.cornell.edu/files/shared/documents/Nosema%20Infosheet.compressed.pdf, accessed 26 Sept. 2017.

Fries, Ingemar. 2009. "Nosema ceranae in European honey bees (Apis mellifera)." *Journal of Invertebrate Pathology*. September. 103. DOI: 10.1016/j.jip.2009.06.017. https://www.researchgate.net/publication/38086103_Nosema_ceranae_in_European_honey_bees_Apis_mellifera, accessed 27 Sept. 2017.

Fries, Ingemar. 1997. "Protozoa." In Roger A. Morse and Kim Flottum (ed.). *Honey Bee Pests, Predators and Diseases*. Medina, Ohio: A.I. Root Company. 3rd Edition. pp.59-73.

Huang, Zachary. 2011. "Effects of Nosema on Honey Bee Behavior and Physiology." *American Bee Journal* and *Bee Culture*. September. http://articles.extension.org/pages/60674/effects-of-nosema-on-honey-bee-behavior-and-physiology, accessed 26 Sept. 2017.

Morse, Roger A. and Kim Flottum (eds.). 1997. *Honey Bee Pests, Predators and Diseases*.

[7] 120 ml of 60% acetic acid are required for 10 frames of comb (Ritter, 2015).

[8] See Fries (2009) and Ritter (2015). It appears that freezing comb does not kill 100% of the *Nosema ceranae* spores (Randy Oliver, BEE-L post, 26 Sept. 2017). Nonetheless, freezing appears to terminate significant numbers of the spores, and therefore, appears to be a useful management tool to control the pathogen. Note that freezing, at least at household deep-freezer temperatures, does not kill *Nosema apis* spores, and hence this management option does not apply to that species.

[9] Fumagillin (Fumagilin-B) is available through many beekeeping supply companies. Consult the relevant beekeeping literature for information about how to administer this drug should you ever need to use it.

Medina, Ohio: A.I. Root Company. 3rd Edition.

National Bee Diagnostic Centre (NBDC). 2016. Canadian National Honey Bee Health Survey. 2016 Report. Beaverlodge, Alberta. https://www.gprc.ab.ca/doc.php?d=2016NHBHS, accessed 21 Sept. 2017.

Newfoundland and Labrador Beekeeping Association (NLBKA). 2017. Research Priorities. http://nlbeekeeping.ca/data/documents/NLBKA_research_priorities29mar2017.pdf, accessed 26 Sept. 2017.

Oliver, Randy. nd. "Nosema ceranae: Kiss of Death or Much Ado about Nothing?" ScientificBeeKeeping.com. http://scientificbeekeeping.com/nosema-ceranae-kiss-of-death-or-much-ado-about-nothing/, accessed 26 Sept. 2017.

Penn State Cooperative Extension. nd. *A Field Guide to Honey Bees and their Maladies.* Penn State University, College of Agricultural Sciences. http://pcbeekeepers.org/wp-content/uploads/2015/02/Penn-State-Extension-A-Field-Guide-to-Honey-Bees-and-Their-Maladies.pdf

Pernal, S.F. and H. Clay (eds.). 2013. *Honey Bee Diseases and Pests*. Beaverlodge, Alberta: Canadian Association of Professional Apiculturists. Third Edition.

OIE (World Organization for Animal Health). 2013. "Nosemosis of Honey Bees." *Manual of Diagnostic Tests and Vaccines for Terrestrial Animals*. http://www.oie.int/fileadmin/Home/eng/Health_standards/tahm/2.02.04_NOSEMOSIS_FINAL.pdf, accessed 25 Sept. 2017.

Perennia. 2016. *Fall Honey Bee Management Guide*. Atlantic Tech Transfer Team for Apiculture. http://www.perennia.ca/wp-content/uploads/2016/04/Fall-Management-Guide.pdf, accessed 27 Sept. 2017.

Pettis, J.S., Y.P. Chen, J. Ellis, J.D. Evans, K.D. Rennich, D. vanEngelsdorp, H. Shimanuki. 2015. "Diseases and Pests of Honey Beeys." In Joe M. Graham (ed.). *The Hive and the Honey Bee*. Hamilton, Illinois: Dadant & Sons. pp.833-879.

Ritter, Wolfgang. 2015. "Practical Beekeeping: Beekeeping with the 'New' Parasite." *Bee Culture*. 25 May. http://www.beeculture.com/practical-beekeeping-beekeeping-with-the-new-parasite/, accessed 26 Sept. 2017.

Shutler, Dave, Krista Head, Karen L. Burgher-MacLellan, Megan J. Colwell, Abby L. Levitt, Nancy Ostiguy, Geoffrey R. Williams. 2014. "Honey Bee *Apis mellifera* Parasites in the Absence of *Nosema ceranae* Fungi and *Varroa destructor* Mites." *PLOS One*. 9(6): E98599. Doi:10.1371/journal.pone.0098599. https://www.researchgate.net/publication/263320994_Honey_Bee_Apis_mellifera_Parasites_in_the_Absence_of_Nosema_ceranae_Fungi_and_Varroa_destructor_Mites, accessed 27 Sept. 2017.

Sammataro, Diana and Alphonse Avitabile. 2011. *The Beekeeper's Handbook*. Ithaca: Cornell University Press. Fourth edition.

Webster, Tom. 2010. "*Nosema ceranae*: the Inside Story." *American Bee Journal* and *Bee Culture*. April. http://articles.extension.org/pages/27064/nosema-ceranae-the-inside-story, accessed 26 Sept. 2017.

White, Gershom Franklin White. 1907. The cause of American foul brood. U.S. Department of Agriculture, Bureau of Entomology Circular 94.

Williams, Geoffrey R. 2010. 2009 Newfoundland and Labrador Honey Bee Disease Survey. Report to Forestry and Agrifoods Agency, Government of Newfoundland and Labrador. February. http://nlbeekeeping.ca/data/documents/2009_honey_bee_disease_report.pdf, accessed 26 Sept. 2017.

Williams, Geoffrey R., Krista Head, Karen L. Burgher-MacLellan, Richard E.L. Rogers, Dave Shutler. 2010. "Parasitic Mites and Microsporidians in Managed Western Honey Bee Colonies in Newfoundland and Labrador, Canada." *Can. Entomol.* 142:584-588.