

In search of DNA markers for mink tolerant to Aleutian disease

A preliminary report

DNA markers, also known as molecular markers, are one of the significant scientific advances of recent years, and are now widely used in a variety of applications in human medicine, aquaculture and agriculture. In essence, a DNA marker is an easily recognizable genetic flag that is used to identify individuals that carry particular forms of genes. The use of DNA markers greatly speeds up the rate of genetic improvement compared to traditional breeding methods.

Differences that are observed among mink for characteristics such as pelt quality, litter size and body weight, as well as tolerance to AD infection, are partly due to their genetic differences. Almost 100 years ago mink ranchers started with small wild mink with low fur quality and through selective breeding created today's larger mink, with high quality fur and good litter size. Indeed, you will be able to establish mink herds that can tolerate the AD virus by traditional breeding as well, but you likely do not want to wait many years to achieve your goal.

What makes tolerance to the AD virus different from body size, fur quality or litter size?

First, body size, fur quality and litter size can be accurately measured and recorded. By contrast, there is no way to measure the amount of viral exposure for different animals in the herd or the timing of exposure, making it difficult to accurately identify tolerant mink. One possibility is to uniformly infect all your mink, but this is not an easy task. In addition, there is currently no laboratory test to accurately identify tolerant mink. The iodine tests, ELISA and PCR have limitations (see my previous report).

Second, selection for body size, fur quality and litter size does not financially hurt you. You keep animals with large body size and good fur quality from large litters as replacements. Selection for tolerance, on the other hand, requires that you live with the virus and its consequences, such as increased mortality and reduced fertility and litter size. Intentionally infecting the herd could escalate these losses.

DNA markers are particularly important for traits that are difficult to measure, such as tolerance to the AD virus infection. Contrary to the iodine, ELISA or PCR tests, that are dependent on the stage of an animal's life and vary from time to time, an animal's genetic makeup remains unchanged throughout its lifetime. Only a single laboratory test is needed to predict an animal's degree of tolerance to the disease. DNA markers have the potential to even predict the level of tolerance of mink that have never been infected with the virus.

If DNA markers are developed, ranchers will be able to predict the level of tolerance of mink at birth using an affordable test, and retain only those mink that can tolerate the virus as replacements. When purchasing mink to broaden the bloodline of your mink herd, a DNA marker test could be used to identify tolerant mink prior to purchase. No other test has this potential. With the absence of a vaccine or a treatment, developing DNA markers for AD tolerance is a logical approach for dealing with the AD virus. This is why the focus of our research is on finding DNA markers for tolerance to the AD virus.

Is there any guarantee that a marker can be developed for tolerance in mink?

There are many success stories of the use of DNA markers in humans and livestock. DNA markers are used to identify human susceptibility to sickle cell anemia, Huntington's disease and some types of cancer. In animal industries, DNA markers have been used for selecting sheep for resistance to scrapie and footrot, in eradication of Porcine Stress Syndrome in swine populations and in identification of susceptible Atlantic salmon for pancreatic necrosis. Developing DNA markers is thus a real possibility. It is possible to find DNA markers for tolerant to AD but it may take several years of research because the genetic control of tolerance to AD is rather complex.

In order to develop DNA markers, three basic components should be in place:

- 1-A group of mink with accurate information on their level of tolerance to infection.
- 2-Complete pedigree of the animals (need to perform single-sire mating).

3-Screening the entire genomes of many tolerant and susceptible mink.

Our progress to date:

We established the AD Research Center, inoculated approximately 1750 mink from 2010 to 2013 with a local strain of the AD virus, and we have regularly monitored antibody production by CIEP and presence of the virus in blood and seven organs of these mink by PCR up to 1200 days after infection. Each female was bred with one male, thus we have complete pedigrees for all animals. We recorded reproductive performance and health conditions of all animals, as well as the level of antibodies and histopathological symptoms of AD in four organs of some of the animals that died or were pelleted to determine the extent of AD symptoms. This is the largest investigation of AD anywhere in the world.

The next step was to identify variability at the DNA level and identify those DNA variants that are associated with tolerance. We selected 285 mink differing in antibody production, viral replication, survival and disease symptoms and their entire genomes were analyzed using the novel method of next generation sequencing (Genotyping-By-Sequencing). Many thousands of DNA variants were identified in these animals, of which 1713 were of a suitable quality for further investigation into their associations with AD tolerance.

The preliminary results suggested that the presence of virus in blood at various times after inoculation, survival, and severity of AD symptoms in kidneys and liver, were strongly associated with genetic differences among mink. Associations between antibody level and severity of the disease symptoms in the lung were weakly associated with DNA variants. The results indicate that there are genetic differences that have the potential to be used as DNA markers to identify tolerant mink.

What is next?

First, we need to test many more animals to gain more confidence in the validity of the results. Second, it is necessary to validate the associations between potential DNA

variants and tolerance in different mink herds to ensure that the test would be applicable for mink of varying genetic backgrounds. Only confirmed DNA variants will be useful DNA markers.

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Hossain Farid and Sean Myles
Dalhousie University Faculty of Agriculture, Truro, Nova Scotia
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For more information, send an e-mail to ah.farid@dal.ca