



## Prevalence of the Aleutian mink disease virus infection in Nova Scotia, Canada

A.H. Farid<sup>a,\*</sup>, M.L. Zillig<sup>a</sup>, G.G. Finley<sup>b</sup>, G.C. Smith<sup>b</sup>

<sup>a</sup> Department of Plant and Animal Sciences, Nova Scotia Agricultural College, Truro, Nova Scotia, B2N 5E3, Canada

<sup>b</sup> Veterinary Services, Nova Scotia Department of Agriculture, Truro, Nova Scotia, B2N 5E3, Canada

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### ABSTRACT

Despite many years of testing mink for serum antibodies against the Aleutian mink disease virus (AMDV) by counterimmunoelectrophoresis (CIEP) and elimination of reactors, this virus has remained the number one disease threat for the mink industry in Nova Scotia (NS). The objective of this study was to analyze CIEP test results to determine the success of the AMDV-control strategy in NS. A total of 2,964,920 CIEP test results from 82 ranches, spanning an eight-year period between 1998 and 2005, were analyzed. This survey included approximately 60% of the active ranchers in the province. The number of ranchers that tested their animals was 42 in 1998, gradually increased to 58 in 2003 and then showed some decline. The overall proportion of CIEP-positive mink was 3.34%, and varied between 5.22% in 1999 and 1.35% in 2005. The proportion of infected ranches ranged between 23.8% in 1998 and 70.7% in 2003. The overall trend was for a smaller proportion of infected animals but a larger proportion of infected ranches during this time period. Of the 82 ranches, 24 (29.3%) had negative CIEP in all tests, 15 (18.3%) had CIEP positive animals in every year tested, and 43 (52.4%) had positive and negative results in different years, indicating that AMDV infection was widespread in NS. There were 23 infected ranches with 8 years of uninterrupted testing. These ranchers performed 75.8% of the total samples tested (2,246,711), implying that they have diligently been trying to eradicate the virus. Infection persisted on three of these ranches for the entire 8 year period, and only two of the ranches remained CIEP negative for longer than four years. The average percentage of CIEP-positive mink on these ranches was 2.2, which was lower than 6.35% for the 33 infected ranches with occasional testing, and 73.6% and 82.4% for two ranches that had never used the CIEP test, showing that persistent test-and-removal strategy has been effective in reducing the prevalence of infected animals but has failed to eradicate the virus from most of the infected ranches.

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### 1. Introduction

Among Canadian provinces, Nova Scotia (NS) is the largest producer of mink pelts (Statistics Canada, 2009) with approximately 80% of the mink ranches located in Digby and Yarmouth Counties at the south-western end

of the province. The high concentration of mink ranches in this region presents a great opportunity for the spread of diseases. Infection with the Aleutian mink disease virus (AMDV) has been a serious problem for the mink industry in NS and Canada. Cho and Greenfield (1978) reported that 46.3% of 41 ranches from different Canadian provinces that each submitted 20 samples were CIEP-positive. Most recently, a survey of Canadian mink ranches showed 32% of 47 ranches who submitted samples from approximately 200 mink had CIEP reactors (Newman and Reed, 2006).

\* Corresponding author.

E-mail address: [hfarid@nsac.ca](mailto:hfarid@nsac.ca) (A.H. Farid).

Although the above surveys were voluntary, the number of participating ranchers was limited and the number of animals tested was small, they showed the presence of AMDV on mink ranchers across the country. AMDV infection is a global issue and is reported in many other mink producing countries, including Denmark (Chriël, 2000; Themudo et al., 2011), Finland (Knuuttila et al., 2009), Iceland (Gunnarsson, 2001), Ireland (Jahns et al., 2010), The Netherlands (Haagsma, 1968), Russia (Martynenko and Bogunov, 2007), Sweden (Olofsson et al., 1999) and the USA (Gohram et al., 1964; Oie et al., 1996). Severity of the disease caused by the AMDV depends on the virus strain and mink genetics (Bloom et al., 1975; Oie et al., 1996). The infection often causes economic losses by increasing adult and embryonic mortality, reducing reproductive performance (Padgett et al., 1967; Alexandersen, 1986; Hansen and Lund, 1988; Broll and Alexandersen, 1996) and causing development of undesirable white hair fibers on the pelt (Farid and Ferns, 2011). Since there is no vaccine or treatment for the disease, the common method of controlling AMDV is by the counterimmunoelectrophoresis (CIEP) test (Cho and Ingram, 1972) and elimination of seropositive animals. Specificity of the CIEP test is over 97%, implying that the test rarely shows false positive results, and its sensitivity could be as low as 75%, suggesting a small chance for false negative results (Bloom et al., 1975; Crawford et al., 1977; Aasted and Cohn, 1982; Alexandersen and Hau, 1985; Aasted et al., 1986; Uttenthal, 1992).

The first successful use of the CIEP for AMDV eradication from infected ranches was reported by Cho and Greenfield (1978), who were able to eliminate the virus from three commercial ranches in Canada in four years. CIEP testing and elimination of reactors in combination with disinfection and repopulation were used to reduce the prevalence of infection in Iceland from a high of 85–94% down to zero during 1982–1984 (Gunnarsson, 2001). Virus eradication by CIEP testing and removal of reactors in Denmark started in 1976 and was successful, although the infection persisted in a small district despite all the eradication efforts, and the virus sporadically appeared in subsequent years in other regions of the country (Chriël, 2000; Themudo et al., 2011). The use of CIEP for viral eradication was adopted by the mink industry in NS in the mid-1970s, but there has been no longitudinal survey of the prevalence of infection of mink herds in this province. The objective of this study was to analyze the results from CIEP testing by ranchers in NS, and to evaluate the success of the test-and-removal strategy in controlling AMDV infection.

## 2. Methods

### 2.1. Source of data

Results of testing of mink for AMDV infection by CIEP in NS, covering an eight-year period from 1998 to 2005, were compiled for this study. The CIEP test is based on a precipitation reaction between antibodies directed against the viral capsid proteins in serum samples and a commercial viral antigen (Cho and Ingram, 1972). The test is

performed in agarose gels, which are soaked overnight in sodium chloride to remove any non-specific proteins, and the results are scored under a stereoscope. The test has been performed by the ISO Accredited NS Department of Agriculture Laboratory since the mid-1970s. A second testing laboratory was established by the NS Mink Breeders Association in Weymouth, NS, in 2003. Although operation of a mink ranch requires a permit from the provincial Department of Agriculture, CIEP testing is not mandatory, except for new entrant ranchers who must start their operations with CIEP negative animals. The NS Mink Breeders Association, however, strongly encourages its members to follow the test-and-removal strategy to control the virus. CIEP test results are confidential and can only be released by the testing laboratories with the ranchers signed consent. For this study, all mink ranchers in NS were asked to sign a consent form, and data from all ranches whose owners responded favorably were used in this survey. Selection of the ranches was thus not at random. Two ranches, one in the eastern and one in the western part of the province, never used the CIEP as a means of controlling the AMDV infection. Arrangements were made to test a sample of 6066 mink from one ranch in 2003, and samples from the second ranch in 2000 ( $n = 59$ ), 2004 ( $n = 746$ ) and 2005 ( $n = 785$ ).

### 2.2. Statistical analysis

Data were analyzed by the Statistical Analysis System, V9.2 (SAS, 2008). Since data were not collected at random, it was necessary to take the effects of ranch and submissions into account to minimize biases due to differences among ranches for sample sizes and percentages of CIEP positive animals. For this reason, the number of reactors as a percentage of animals tested was calculated for each submission, and the results were analyzed for each year using the Proc Surveymeans of SAS with ranch as the strata and the numbers of animals in each submission as the weight. Inclusion of the weights eliminated biases due to the correlation between the sample size and percentage of CIEP positive animals in each submission. Means, standard errors and 95% confidence intervals were calculated. Two cases where one animal in an otherwise clean ranch tested positive were discarded.

Ranches were further classified into one of the four groups for a more accurate description of their status, namely (a) those that were CIEP-negative in all tests during this period, (b) never used CIEP as a selection tool, (c) had 8 years of uninterrupted testing, and (d) infected ranches with partial testing. Data of the ranches with 8 years of uninterrupted testing were further analyzed by PROC MIXED of SAS. The percentage of reactors in each submission was analyzed using a model which included the fixed effects of year and season and the random effect of ranch. The numbers of animals in each submission was used as the weight. Differences between average sample prevalence were assessed in a post hoc pairwise testing procedure with Bonferroni adjustment for multiple comparisons.

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