Birds of a Feather, How Do They Get Sick Together?:

Exploring the Prevalence of Avian Influenza Virus (AIV) in Mallards and Blue-winged Teals

Group 5

Maria Kanellis (1223184), Abigail Luke (1190638), Haylee Rae (1183175), Aisha Shaikh (1191134)

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Dr. Joanna Bernhardt

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Introduction

Avian influenza virus (AIV) represents a significant threat to global avian health. Understanding the factors that affect avian influenza is crucial for predicting outbreaks, informing management strategies, and minimizing the risks of pandemics. Temperature, environmental factors, seasonality, and population demographics shape the prevalence and transmission of avian influenza viruses in waterfowl. Though much work has been done to observe these factors and how they interact with the virus, few have taken a closer look at how they interplay with each other, especially in a modern Canadian landscape.

Temperature has been identified as a critical factor in the persistence and spread of avian influenza. Research has shown that lower ambient temperatures extend virus survivability in fecal and water samples, enhancing the potential for environmental transmission (Zarkov & Urumova, 2013). In this study by Zarkov & Urumova (2013), the H6N2 virus persisted for up to 14 days at 4°C, compared to only two days at 22°C, indicating that cooler environments are more conducive to avian influenza stability.

Because of temperature's impact on avian influenza prevalence, seasonal variations also play a role in the prevalence of avian flu among wild waterfowl. The overwintering season (October—March) is characterized by higher avian influenza prevalence due to low temperatures, which favour virus persistence in the environment (Farnsworth et al., 2012). This seasonal peak influences infection rates in subsequent breeding seasons (April—September), suggesting a temporal carryover effect of avian influenza infection (Farnsworth et al., 2012).

Environmental variables, such as pH, salinity, and humidity, also affect avian influenza survivability. Avian influenza is the most stable in slightly basic (pH around 7.4–8.2) freshwater environments (Brown et al., 2009). High humidity also increases avian influenza's survival length in fecal samples, with higher moisture content prolonging infectivity (Zarkov &

Urumova, 2013; Lowen et al., 2007). These findings emphasize how different environmental conditions found in different regions affect avian influenza survival.

While there are studies that focus on temperature (Zarkov et al. 2013; Brown et al., 2009), they only collected data from a low number of sampled specimens (4 individuals) or focused on samples in isolation, respectively. Overall, there is a lack of focused studies that account for combined factors to understand their effects on AIV prevalence collectively.

Demographic factors, including age, sex, and population density also contribute to avian influenza dynamics. Younger, hatch-year birds exhibit higher susceptibility to infection, due to their underdeveloped immune systems and limited exposure to viruses (Farnsworth et al., 2012). In addition, male birds exhibit higher susceptibility to infection, which is likely due to their behavioural patterns of foraging and socializing more than female birds (Farnsworth et al., 2012). For population density, higher densities of waterfowl increase the infectability of avian influenza; dense populations will have more interactions between individuals and therefore the likeliness of infection is higher (Velkers et al., 2021).

Though these studies do provide some insight, they seem to be focused on specific regions. Farnsworth et al. (2012) only look at certain parts of the USA, and Velkers et al. (2021) only focus on areas near poultry farms. Due to restrictions in resources and time, it seems there is a general trend for demographic investigations to be spatially limited.

The question therefore arises: What environmental and demographic factors affect the prevalence of avian influenza in mallards and blue-winged teals in Canada? When conducting our review of the literature, we found that temperature, habitat type, and sex had the biggest impact on the prevalence of avian influenza within wild waterfowl populations, so we will be focusing on these three factors. Our temperature hypothesis states that variations in the prevalence of avian influenza in mallards and blue-winged teal are associated with temperature. Research has shown that lower ambient temperatures extend virus survivability in fecal and water samples, enhancing the potential for environmental transmission. We predict that a decrease in temperature will lead to an increase in avian influenza prevalence in these waterfowl species. We also hypothesize that habitat preference plays a critical role in avian influenza transmission as high humidity around water bodies increases avian influenza's spread. If our hypothesis is true, we predict that aquatic habitats will have a higher avian influenza prevalence. Finally, we hypothesize that sex will significantly affect the probability of an individual testing positive for avian influenza. Specifically, we predict that male birds will have a higher prevalence of avian influenza than females. Together, these hypotheses aim to shed light on the environmental and demographic determinants of avian influenza dynamics in wild waterfowl populations.

Methods

This study was done using the dataset publicly provided by Papp et al. (2017) on Dryad, in their study on factors affecting avian influenza in wild waterfowl populations in Canada. While the initial dataset was rich in raw data, we identified which variables we would test for in our analysis, then cleaned the dataset to include only the variables to be analyzed, specifically: Species (filtered for only mallards and blue-winged teal, listed as MALL and BWTE respectively), Sampling sites, AIV, Temperature, and Sex.

From there, we manually added habitat types, which were categorized as Bay, Creek, Farmland, Lake, Marsh, Mixed, Pond, River, Trails, or Urban. This was achieved with additional research using the dataset's Sampling Latitude and Sampling Longitude and inputting the coordinates into *www.gps-coordinates.net*. We also calculated total population density (total sampled per site), and proportion. To calculate the proportion for proper comparison between variables, we turned positive AIV cases into a percentage of infected specimens of the total number of samples from each location.

To identify any potential trends of AIV over time, we calculated the proportion of positive AIV for all specimens tested during each month of sampling throughout the study and plotted these proportions as a percentage represented in a stacked bar graph. Next, we ran a linear regression model in R to compare all the selected variables against AIV prevalence and plotted the results. To identify the best-fitting models based on the AIC (Akaike Information Criterion), we used the dredge function from the MuMIn package in R.

Results

The outcomes of this study focused on the environmental and demographic factors that affect AIV prevalence in waterfowl species (mallards and blue-winged teals) across Canada. In Figure 1, higher proportions of the sampled specimens were found to be infected with AIV in 2005; in addition, AIV periodically increased during the summer months. Next, the linear model was run using sex, temperature, and habitat type as predictor variables. When analyzing the effects different variables had on AIV prevalence, the linear model reported each factor (temperature, habitat type and sex) had a strong effect.

Contradictory to our first hypothesis, the linear regression model in Figure 2 showed that a higher temperature leads to a higher prevalence of AIV. In line with our second and third hypotheses, the linear regression model (Figure 3) showed that habitats with water bodies had a higher prevalence of AIV, and Figure 4 indicated that males were more likely to be infected with AIV than females. Models were evaluated using the Akaike Information Criterion (AIC) to predict the best fit of the proportion of the population infected with AIV, as seen in Table 1. Model 1, which included all factors, was determined to be the best fit with an AIC value of -20556.11.

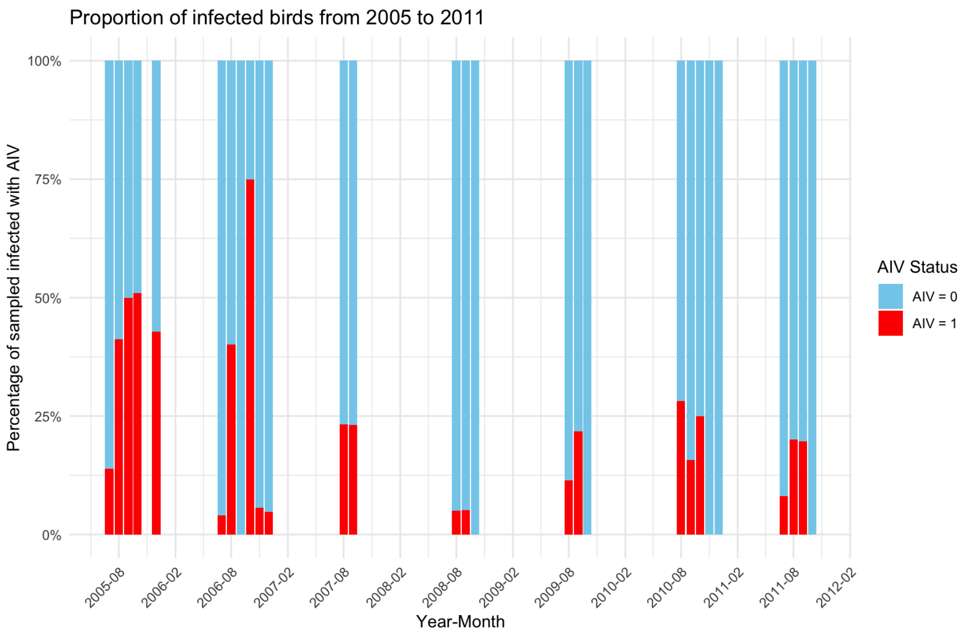

Figure 1. Proportion of infected mallards and blue-winged teals sampled across Canada from 2005-2011. Percentage of infected (red) is compared to the percentage of uninfected (blue) individuals during each month of sampling, n = 18,723.

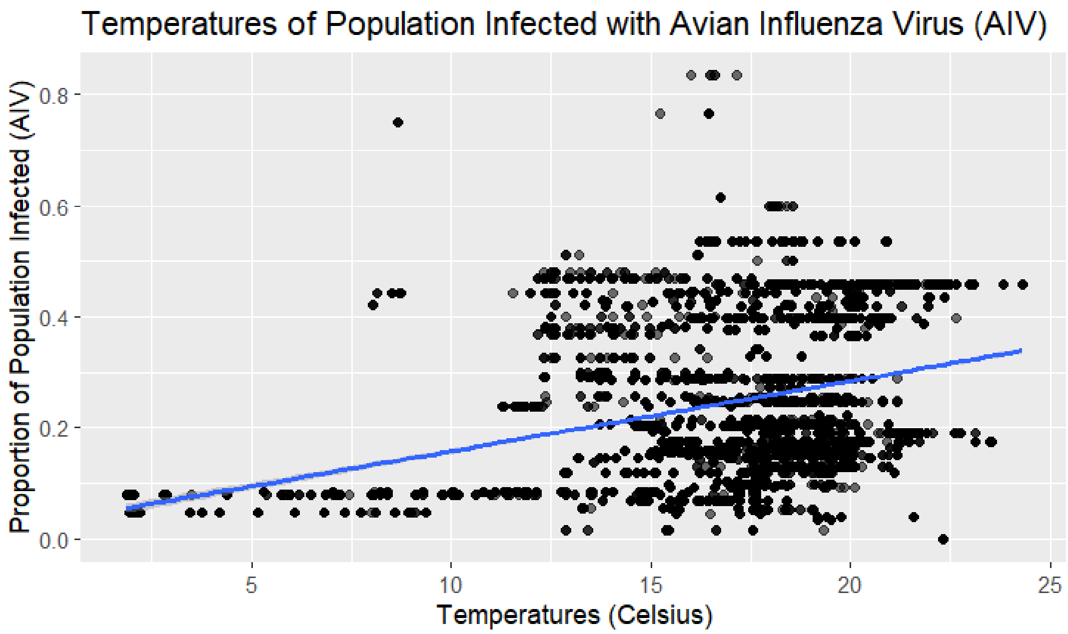

Figure 2. Linear regression of temperature vs. proportion of infected populations of mallards and blue-winged teals in 95 sites across Canada. A positive correlation between the prevalence of AIV and temperature was found, with the majority of infected individuals found around 15-20˚C (n = 95, p-value = < 2e-16, slope = 1.380e-02).

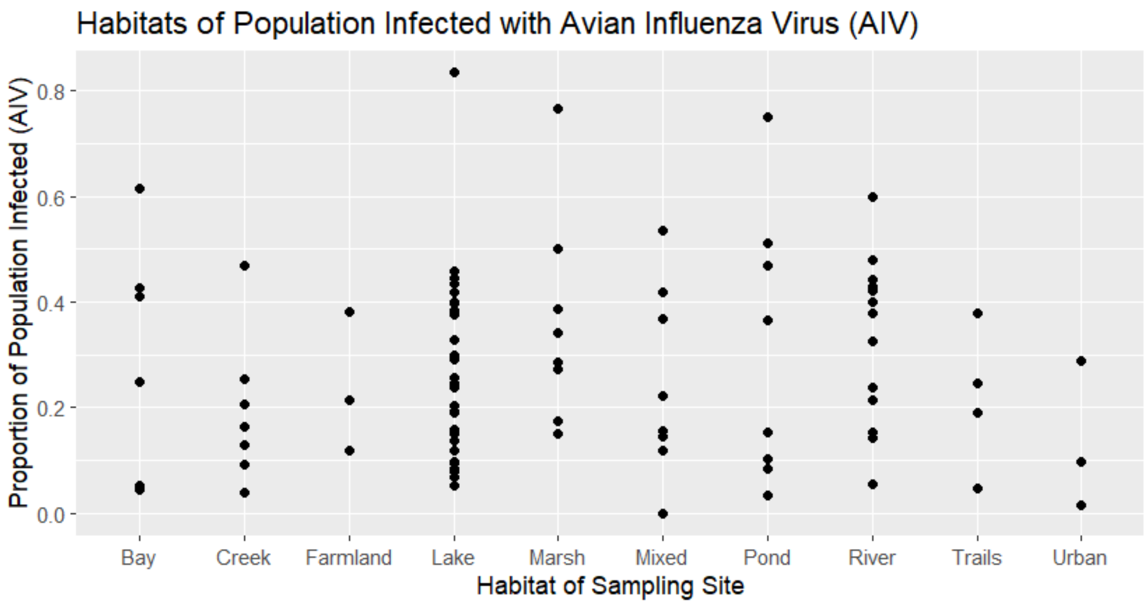

Figure 3. Proportion of Canadian mallard and blue-winged teal population infected with avian influenza virus and the type of environment in which they were found across 95 sites. All aquatic environments with the exception of creeks exceeded 60% infected. AIV was the least prevalent on farmland, trails, and urbanized areas ($n=95$, $p < 2.2e-16$).

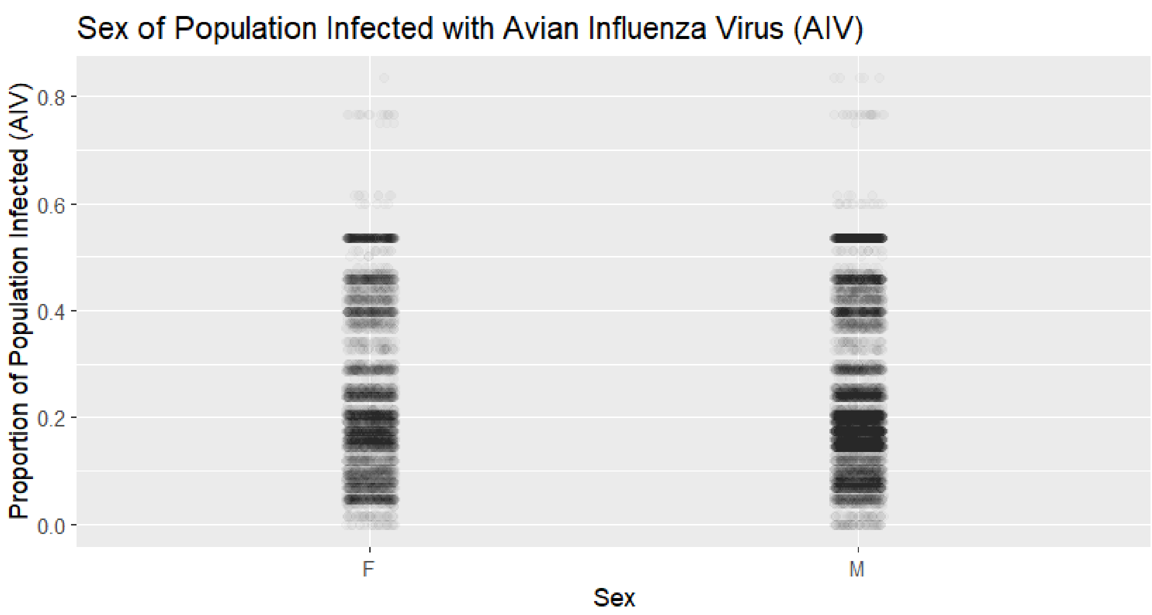

Figure 4. Proportion of male and female mallards and blue-winged teals infected with avian influenza virus over 95 sites across Canada. The darker bands indicate more populated replicates. A significant difference was found between the males and females ($n = 95$, P-value $= 3.18e-14$).

Table 1: AIC values for predicting the proportion of population infected with AIV.

This table displays the AIC values of various models. Proportion represents the percentage of the population in each site infected with AIV, sex represents male or females, temperature represents in degrees Celsius, type of site represents the habitat type (pond, lake, river, etc), and n represents the total number of mallards/ducks observed in each site. The model that includes n, sex, temperature, and the type of site has the lowest AIC value, indicating it is the best fit.

Model	AIC Value	ΔAIC
Model 1: Proportion ~ n + sex + temperature + type of site	-20556.11	
Model 2: Proportion ~ n + temperature + type of site	-20523.56	32.55
Model 3: Proportion ~ sex + temperature + type of site	-20522.18	34.03
Model 4: Proportion ~ temperature + type of site	-20493.03	63.08

Discussion

Studying the spread of avian influenza in wild waterfowl is crucial for wild animal and human health. By identifying hotspots and patterns of transmission, management efforts can be better directed toward specific regions, ensuring resources are used effectively to mitigate the spread. Monitoring wild waterfowl populations also plays a vital role in detecting species-to-species jumps, which could have significant ecological and public health implications, especially if the virus spreads to domestic poultry or humans. The findings have the potential to provide valuable guidance to farmers and food suppliers, helping them implement preventative measures to protect their livestock and maintain food security. Understanding how the virus evolves and transmits within and between species sheds light on its potential future impacts, enabling informed responses to outbreaks. Limited to only examining the spread of avian influenza between two different *Anas* species (mallards and blue-winged teals), our study nonetheless provides a good starting point for addressing the gaps in the literature on specific factors that affect avian influenza transmission. It demonstrates the significance of considering all factors influencing AIV prevalence.

While our hypothesis regarding temperature was disproved by our analyses–showing that higher temperatures lead to a higher prevalence of avian influenza–we were able to relate our results to a global issue many researchers are currently exploring. New research on the impacts of a warming climate has shown that warmer temperatures may be leading to higher prevalence and easier transmission of many diseases (Anwar et al., 2019). Increasing global temperatures would allow easier disease transmission by improving the survival and reproduction rates of pathogens and their vectors–carriers of diseases like mosquitoes or ticks (Anwar et al., 2019). Warmer conditions shorten the incubation period for pathogens, allowing them to become infectious more quickly (Anwar et al., 2019). Additionally, rising temperatures can expand the geographic range of these diseases into new areas, causing them to infect a greater portion of the population (Anwar et al., 2019). While most studies on this topic have focused on virus transmission in humans, many of these same principles can be applied to birds and the transmission of avian influenza. Climate change has been seen to affect the spread of avian influenza by changing bird migration patterns, increasing the survivability of the virus in certain environments, and speeding up the evolution of avian influenza in wild bird populations, making it more infectious (Gilbert et al., 2008).

This study, which relied on a publicly available dataset rather than collecting original data, has several limitations that could be addressed to strengthen its findings. Conducting a similar study with other *Anas* species or even other genera of waterfowl affected by avian influenza could provide valuable insights into interspecies variations and help determine whether the observed patterns are species-specific or broadly applicable across waterfowl.

Additionally, the study's geographic scope is limited, as the data used focused solely on Canada, which restricts its relevance to other regions. Expanding similar studies to different regions worldwide would help uncover regional differences and enhance the global applicability of the results. Furthermore, while we had initially hoped to investigate specific environmental factors, such as the depth, width, and temperature of the water—given the lack of literature regarding their role in avian influenza transmission—unfortunately, we were unable to include these variables due to a lack of consistent data across study sites. However, we were able to test the specific habitat types effect, and while this provided us with important information, we still experienced limitations as the distribution of habitat types was very skewed (i.e. many more lakes and rivers than farmlands or creeks). This could be better tested in future studies by having a larger number of study sites with a more even distribution of habitat type. Certain features of the water body like depth and width could also be recorded as part of a dataset. When probing deeper based on the water bodies, it brings to question if the type of water body plays an impact. Since AIV prevalence can be transmitted through fecal matter in water bodies, would smaller water bodies such as creeks and ponds have higher AIV than lakes and rivers? This is another possibility that could be further tested in the future. Addressing these gaps in future research would provide a more comprehensive understanding of the complex ecological dynamics involved in the transmission and development of avian influenza.

Dataset

Papp, Zsuzsanna; Clark, Robert G.; Parmley, E. Jane et al. (2018). Data from: The ecology of avian influenza viruses in wild dabbling ducks (Anas spp.) in Canada [Dataset]. Dryad. https://doi.org/10.5061/dryad.7gp59

GPS coordinates, latitude and longitude with interactive maps. GPS coordinates, latitude and longitude with interactive Maps. (n.d.). http://www.gps-coordinates.net/

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