SHORT COMMUNICATIONS

Journal of Wildlife Diseases, 48(2), 2012, pp. 444–448

Bait Trapping Linked to Higher Avian Influenza Virus Detection in Wild Ducks

Catherine Soos,[1,2,8] E. Jane Parmley,[3] Keith McAloney,[4] Bruce Pollard,[4] Emily Jenkins,[1,5] Fred Kibenge,[6] and Frederick A. Leighton[7,2] [1] Science and Technology Branch, Environment Canada, 115 Perimeter Rd., Saskatoon, Saskatchewan, S7N 0X4, Canada; [2] Department of Veterinary Pathology, Western College of Veterinary Medicine, University of Saskatchewan, 52 Campus Dr., Saskatoon, Saskatchewan S7N 5B4, Canada; [3] Canadian Cooperative Wildlife Health Centre, Ontario Veterinary College, University of Guelph, Guelph, Ontario, N1G 2W1, Canada; [4] Canadian Wildlife Service, Environment Canada, 17 Waterfowl Lane, Sackville, New Brunswick, E4L 1G6, Canada; [5] Department of Veterinary Microbiology, Western College of Veterinary Medicine, University of Saskatchewan, 52 Campus Dr., Saskatoon, Saskatchewan S7N 5B4, Canada; [6] Department of Pathology and Microbiology, Atlantic Veterinary College, University of Prince Edward Island, 550 University Avenue, Charlottetown, Prince Edward Island C1A 4P3, Canada; [7] Canadian Cooperative Wildlife Health Centre, Western College of Veterinary Medicine, University of Saskatchewan, 52 Campus Dr., Saskatoon, Saskatchewan S7N 5B4, Canada; [8] Corresponding author (email: catherine.soos@ec.gc.ca)

ABSTRACT: In 2007, we assessed whether trapping method influenced apparent prevalence of low pathogenic avian influenza viruses (AIV) in wild ducks sampled during Canada's Inter-agency Wild Bird Influenza Survey. Combined cloacal and oropharyngeal swabs were collected from 514 ducks captured by bait trapping (356) and netting from airboats (158), and tested by real-time reverse transcriptase polymerase chain reaction for influenza type A viruses. When controlling for species and capture site, ducks caught in bait traps were 2.6 times more likely to test positive for AIV compared with those netted from airboats (95% CI=1.2–6.0). If bait trapping increases AIV transmission among artificially aggregated ducks, this could have important implications for interpretation of disease surveillance results and waterfowl management programs.

Key words: Avian influenza virus, bait trap, Canada, detection, ducks, waterfowl.

Since the inception of Canada's Inter-agency Wild Bird Influenza Survey in 2005, over 19,000 live wild ducks have been caught and tested for avian influenza virus (AIV). No highly pathogenic avian influenza strains have been detected. In 2006, field biologists in Atlantic Canada observed that the proportion of AIV-positive ducks captured by bait traps appeared higher than for ducks netted from airboats. However, it was not possible to determine whether this reflected a true difference or whether it was due to other confounding factors such as capture date, duck species, or sampling site. Bait traps can result in focal areas of unusually high bird densities given that their main function is to attract and contain ducks. As such, bait traps have the potential to artificially increase local AIV prevalence by facilitating transmission among birds. Our objective was to evaluate whether ducks captured by bait traps are more likely to be infected with AIV compared to the general population, while controlling for species, time, and location of sampling. We used ducks netted from airboats as our control population, and assumed that the level of detection in birds trapped by this method was less prone to bias, and more reflective of the true AIV prevalence in the general population.

Between 30 July and 28 August 2007, wild dabbling ducks were captured, sampled, and banded at 17 sites within a 700-km^2 area spanning the New Brunswick–Nova Scotia border (Fig. 1). Ducks were netted from airboats at five sites and captured using bait traps at 12 sites (Fig. 1). Only one trapping method was applied at each site due to the small water surface area of wetlands (8–125 ha). At each bait trap site, one to three standard cloverleaf traps were set with top nets off and funnel entrances closed, and baited with corn for 5–7 days to allow ducks to acclimatize to traps. When banding and

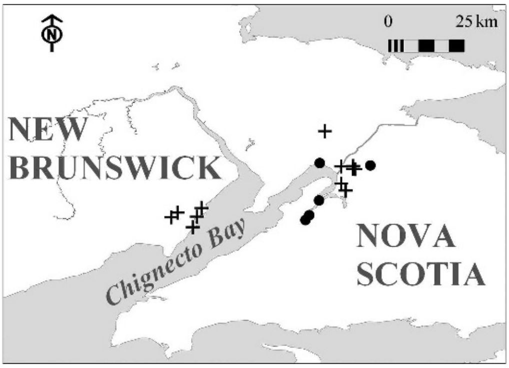

FIGURE 1. Location of duck capture sites between 30 July 2007 and 28 August 2007, indicating the use of bait trapping (plus signs) or airboat netting (circles). Ducks were captured at 17 sites clustered within two main gross complex sites: Shepody (45°42′N, 64°43′W, five sites) and Border Region (45°49′N, 64°16′W, 12 sites), separated by Chignecto Bay, an inlet of the Bay of Fundy. Maps were generated using ArcGIS® 9.0 – ArcMap™ version 9.1 (ESRI, Redlands, California, USA).

sampling commenced, top nets were placed on each trap and funnel entrances were opened. Once opened, traps were checked daily. Airboat captures were conducted at night, intermittently throughout the same time period. All ducks captured were banded, sampled for AIV testing, and released. Oropharyngeal and cloacal swabs were collected from each bird, placed together in a single vial, and stored as described by Parmley et al. (2011). Samples were tested for the presence of the M1 gene by real-time reverse transcriptase polymerase chain reaction (RRT-PCR; Spackman et al., 2002) at the University of Prince Edward Island, as described previously (Parmley et al., 2008).

Effect of trapping method on AIV status (RRT-PCR result) was assessed using binomial logistic regression employed with a random intercept (two-level) model, using capture site as a random effect to account for potential clustering at the site level. Two potential sources of geographic clustering were initially considered in models: capture site and gross complex. The 17 capture sites were clustered within two main gross complexes separated by Chignecto Bay (Fig. 1). When examined with gross complex in a three-level model, capture site accounted for all effects of clustering, thus two-level models were used with capture site as the only random effect. Potential confounding variables, including age, sex, species, and week of capture, were assessed individually using the same two-level model design. Variables showing an unconditional association ($P<0.20$) with AIV status were retained for model building. Variables remained in the model if significantly associated with the outcome ($P<0.05$) or if identified as confounding variables (change in odds ratios of other variables by 20% or more). All models were created in MLwiN® version 2.0 (Centre for Multilevel Modelling, University of Bristol, Bristol, UK).

We captured 514 dabbling ducks (subfamily Anatinae) of five species during the study period; 158 (31%) were netted from airboats and 356 (69%) were caught in bait traps (Table 1). Most species were captured using both trapping methods, except for American Wigeon (AMWI, *Anas americana*) and Mallard–Black Duck hybrids (MBDH, *Anas platyrhynchos* × *Anas rubripes*, Table 1). Despite being present throughout the study area, AMWI were not captured in bait traps, likely due to their preference for pondweeds, grasses, and algae (Mowbray, 1999), in contrast to other dabbling ducks, which have an affinity for seeds, grains, and corn used in bait traps. All but three ducks were hatch-year birds, of which 327 (64%) were flighted and 184 (36%) were not. Fifty-four percent (280/514) of ducks were male and 46% (234/514) were female.

Overall, 16% of ducks were positive for AIV; 21% of ducks caught in bait traps were positive, compared with only 6% of ducks netted from airboats (Table 1). Trapping method, species, and week of capture were unconditionally associated with AIV status in univariate, two-level models (models not shown), and hence were examined together in a single model as potentially important predictor vari-

TABLE 1. Sample sizes and overview of avian influenza infection status (as measured by real-time reverse transcriptase polymerase chain reaction of the M1 gene) in wild duck species captured using nets from airboats and bait traps in Atlantic Canada, 30 July 2007 to 28 August 2007.

	Netted from airboat		Caught in bait trap	
Species[a]	No. sampled	No. positive (%)	No. sampled	No. positive (%)
ABDU	52	3 (6)	186	34 (18)
AGWT	22	1 (5)	21	2 (10)
AMWI	55	2 (4)	0	0
MALL	29	4 (14)	141	32 (23)
MBDH	0	0	8	6 (75)
Total	158	10 (6)	356	74 (21)

[a] ABDU = American Black Duck (*Anas rubripes*); AGWT = American Green-winged Teal (*A. crecca*); AMWI = American Wigeon (*A. americana*); MALL = Mallard (*A. platyrynchos*); MBDH = Mallard–Black Duck hybrid (*A. platyrynchos* × *A. rubripes* hybrid).

ables. Age and sex were not associated with AIV status in univariate analyses and were not included in the final model (models not shown).

Only species and trapping method remained as fixed effects in the final model (Table 2), which explained 90.9% of the total null variance in AIV status. Variance among capture sites accounted for only 1.4% of total residual variation in AIV status. Nonetheless, it was important to control for effects of capture site by keeping it as a random effect in the model. After accounting for site and species, ducks caught in bait traps were 2.6 times more likely to test positive for AIV compared with netted ducks (Table 2). The MBDH were significantly more likely to test positive for AIV than mallards. This finding is most likely explained by the small sample of MBDH ($n=8$), all of which were caught in bait traps, rather than an increased susceptibility to being infected with AIV. To ensure that the inclusion of species caught by only one trap method did not confound the relationship between AIV infection status and trapping method, we ran the same models excluding MBDH and AMWI. The final model for this data subset included only trapping method as a significant variable, and the relationship between trapping method and AIV status was similar (OR=3.0, CI=1.3–7.3, $n=451$; model not shown). Species was no longer important since MBDH had accounted for most of the variation among species.

Our results suggest that ducks captured by bait traps have a higher probability of becoming exposed to and/or spreading AIV while concentrated around or within bait traps, compared with ducks netted from airboats, which are assumed to be more reflective of the general population. Ducks can shed AIVs for 1–10 days postinfection (e.g., Brown et al., 2006; Keawcharoen et al., 2008), and low pathogenic AIVs (LPAIVs) can survive in water for months (e.g., Stallknecht et al., 1990; Rohani et al., 2009). Thus it is plausible that many of the positive ducks were directly or indirectly exposed to AIVs within or around bait traps. Further studies are required to compare viruses recovered from birds within and among bait traps. If ducks are exposed to AIVs around or within bait traps, one would expect a high level of genetic similarity among viruses from birds captured within the same traps.

It might be argued that ducks infected with LPAIV have an increased tendency to feed around or enter bait traps compared to uninfected birds. If LPAIV infection compromised their health (e.g., Latorre-Margalef et al., 2008), infected ducks might seek bait traps as a relatively easy source of food. However, investigators

TABLE 2. Final (two-level) model[a] explaining the effect of trap method and species on avian influenza infection status (as determined by real-time reverse transcriptase polymerase chain reaction [RRT-PCR] for the M1 gene) in wild ducks sampled in Atlantic Canada, 30 July 2007 to 28 August 2007 (n=514).

Parameter	β	SE	Odds ratio	95% Confidence limits	P
Intercept	−2.166	0.424			
Bait trap method (reference = airboat)	0.973	0.421	2.6	1.2, 6.0	0.02
Species (reference = MALL)[b]					
ABDU	−0.348	0.276	0.7	0.4, 1.2	0.21
AGWT	−1.012	0.647	0.4	0.1, 1.3	0.12
AMWI	−1.162	0.836	0.3	0.1, 1.6	0.16
MBDH	2.258	0.845	9.6	1.8, 50.1	0.008
Capture site (random effect term)	0.048	0.098			0.62

[a] Model equation: RRT-PCR result$_{ij}$ ~ Binomial (n_{ij}, π_{ij}); Logit(π_{ij}) = log [$\pi_{ij}/(1-\pi_{ij})$] = β_{0j} constant + β_{ij} trap method + β_{ij} species + u_{0j}, where i indicates individual level, j indicates capture site level, and u_{0j} is the random effect term for capture site.

[b] MALL = Mallard (*Anas platyrynchos*); ABDU = American Black Duck (*A. rubripes*); AGWT = American Green-winged Teal (*A. crecca*); AMWI = American Wigeon (*A. americana*); MBDH = Mallard–Black Duck hybrid (*A. platyrynchos* × *A. rubripes* hybrid).

who have tested the condition bias hypothesis (Weatherhead and Ankney, 1984) in waterfowl species have shown no difference in body condition between ducks captured in baited traps compared with ducks captured by rocket nets at unbaited sites (e.g., Reinecke and Shaiffer, 1988).

In most areas of Canada, bait trapping is the method of choice to capture live, healthy ducks for waterfowl banding programs and concurrent sampling for AIV. If bait trapping increases AIV transmission and exposure among artificially aggregated ducks, this could have implications for interpretation of disease surveillance results and management practices. If the survey goal is to detect as many AIVs as possible, then bait traps are the best capture method, provided researchers recognize that the proportion of bait-trapped ducks positive for AIV (or other infectious diseases with short incubation periods) likely overestimates true prevalence. From a management perspective, given widespread use of bait traps for North American waterfowl banding programs, congregation of ducks at baited sites could have important impacts on the ecology and spread of AIV and other pathogens. This is particularly important should highly pathogenic AIV enter wild waterfowl populations.

We thank CCWHC staff for logistical and database assistance, numerous biologists of the Canadian Wildlife Service-Atlantic Region who collected samples, M. Kibenge, for technical assistance in the laboratory, L. Shirose for assistance with mapping, and K. Dufour and P.-Y. Daoust for helpful comments. This work was approved by Environment Canada's Animal Care Committee, and adhered to Canadian Council on Animal Care guidelines for humane animal use. Funding was provided by the Canadian Food Inspection Agency, Public Health Agency of Canada, and Environment Canada.

LITERATURE CITED

BROWN, J. D., D. E. STALLKNECHT, J. R. BECK, D. L. SUAREZ, AND D. E. SWAYNE. 2006. Susceptibility of North American ducks and gulls to H5N1 highly pathogenic avian influenza viruses. Emerging Infectious Diseases 12: 1663–1670.

KEAWCHAROEN, J., D. VAN RIEL, G. VAN AMERONGEN, T. BESTEBROER, W. E. BEYER, R. VAN LAVIEREN, A. D. M. E. OSTERHAUS, R. A. M. FOUCHIER, AND T. KUIKEN. 2008. Wild ducks as long distance vectors of highly pathogenic avian influenza virus (H5N1). Emerging Infectious Diseases 14: 600–607.

Latorre-Margalef, N., G. Gunnarsson, V. J. Munster, R. A. M. Fouchier, A. D. M. E. Osterhaus, J. Elmberg, B. Olsen, A. Wallensten, P. D. Haemig, T. Fransson, L. Brudin, and J. Waldenström. 2008. Effects of influenza A virus infection on migrating Mallard ducks. Proceedings of the Royal Society B 276: 1029–1036.

Mowbray, T. 1999. American Wigeon (*Anas americana*). *In* The birds of North America, No. 410, A. Poole and F. Gill (eds.). The Birds of North America, Inc., Philadelphia, Pennsylvania, pp. 1–32.

Parmley, E. J., N. Bastien, T. F. Booth, V. Bowes, P. A. Buck, A. Breault, D. Caswell, P.-Y. Daoust, J. C. Davies, S. M. Elahi, M. Fortin, F. Kibenge, R. King, Y. Li, N. North, D. Ojkic, J. Pasick, S. P. Pryor, J. Robinson, J. Rodrigue, H. Whitney, P. Zimmer, and F. A. Leighton. 2008. Wild bird influenza survey, Canada, 2005. Emerging Infectious Diseases 14: 84–87.

———, C. Soos, A. Breault, M. Fortin, E. Jenkins, F. Kibenge, R. King, K. McAloney, J. Pasick, S. P. Pryor, J. Robinson, J. Rodrigue, and F. A. Leighton. 2011. Detection of low pathogenic avian influenza viruses in wild ducks from Canada: Comparison of two sampling methods. Journal of Wildlife Diseases 47: 466–470.

Reinecke, K. J., and C. W. Shaiffer. 1988. A field test for differences in condition among trapped and shot Mallards. Journal of Wildlife Management 52: 227–232.

Rohani, P., R. Breban, D. E. Stallknecht, and J. M. Drake. 2009. Environmental transmission of low pathogenicity avian influenza viruses and its implications for pathogen invasion. Proceedings of the National Academy of Sciences of the United States of America 106: 10365–10369.

Spackman, E., D. A. Senne, T. J. Myers, L. L. Bulaga, L. P. Garber, M. L. Perdue, K. Lohman, L. T. Daum, and D. L. Suarez. 2002. Development of a real-time reverse transcriptase PCR assay for type A influenza and avian H5 and H7 hemagglutinin subtypes. Journal of Clinical Microbiology 40: 3256–3260.

Stallknecht, D. E., M. T. Kearney, S. M. Shane, and P. J. Zwank. 1990. Effects of pH, temperature, and salinity on persistence of avian influenza viruses in water. Avian Diseases 34: 412–418.

Weatherhead, P. J., and C. D. Ankney. 1984. A critical assumption of band-recovery models may often be violated. Wildlife Society Bulletin 12: 198–199.

Submitted for publication 4 May 2010.
Accepted 30 September 2011.