

Avian Cholera Causes Marine Bird Mortality in the Bering Sea of Alaska

Barbara Bodenstern,^{1,6} Kimberlee Beckmen,² Gay Sheffield,³ Kathy Kuletz,⁴ Caroline Van Hemert,⁵ Brenda Berlowski,¹ and Valerie Shearn-Bochsler¹ ¹US Geological Survey, National Wildlife Health Center, 6006 Schroeder Rd., Madison, Wisconsin 53711, USA; ²Alaska Department of Fish and Game, Division of Wildlife Conservation, 1300 College Road, Fairbanks, Alaska 99701, USA; ³University of Alaska Fairbanks, Marine Advisory Program, Pouch 400, Nome, Alaska 99762, USA; ⁴US Fish and Wildlife Service, Migratory Bird Management, 1011 E Tudor Rd., Anchorage, Alaska 99503, USA; ⁵US Geological Survey, Alaska Science Center, 4210 University Drive, Anchorage, Alaska 99508, USA; ⁶Corresponding author (email: bbodenstern@usgs.gov)

ABSTRACT: The first known avian cholera outbreak among wild birds in Alaska occurred during November 2013. Liver, intestinal, and splenic necrosis consistent with avian cholera was noted, and *Pasteurella multocida* serotype 1 was isolated from liver and lung or spleen in Crested Auklets (*Aethia cristatella*), Thick-billed Murres (*Uria lomvia*), Common Eider (*Somateria mollissima*), Northern Fulmars (*Fulmarus glacialis*), and gulls (*Larus* spp.).

Avian cholera, caused by the highly contagious bacterium *Pasteurella multocida*, rapidly causes death in wild birds, particularly in waterfowl (Samuel et al. 2007). The disease has been reported in >180 bird species (Samuel et al. 2007). Transmission can occur by bird-to-bird contact, aerosolized bacteria, and ingestion of bacteria in contaminated environments. Outbreaks involving wild birds occur annually in North America, and evidence suggests that the disease tends to follow the migration routes of some birds, most notably Lesser Snow Geese (*Chen caerulescens caerulescens*; Samuel et al. 2005b). Outbreaks are explosive and can kill thousands of birds in a short time (Samuel et al. 2007).

Saint Lawrence Island (SLI) lies in the Bering Strait region of Alaska and is home to two Alaska Native communities, Gambell (63°46'46.98"N, 171°44'27.92"W) and Savoonga (63°41'42.44"N, 170°28'44.51"W). Reliance on marine resources, including birds, for subsistence remains essential for the nutritional, cultural, and economic needs of these island communities. Over

85% of subsistence-harvested resources in the region are marine derived (Ahmasuk 2008). The island's abundance of marine resources is due largely to the influence of the Anadyr and Alaska currents, which bring cold, nutrient-rich water from the deep to the shallow shoals of the northern Bering Sea (Piatt and Springer 2003). Mixing of these currents occurs north of SLI and facilitates prey availability for piscivores (murre), planktivores (auklets), and omnivores (fulmars; Piatt and Springer 2003). All cholera-impacted species from this event occupy coastal waters surrounding SLI, which have been designated Globally Important Bird Areas of the United States (Smith et al. 2014).

In November 2013, unusual marine bird mortality off the northern coast of SLI was reported by citizens in Gambell and Savoonga to the University of Alaska Fairbanks Marine Advisory Program and the Alaska Department of Fish and Game. Observers reported hundreds of bird carcasses washing ashore near the two villages. Clinical signs in live birds included swimming in circles with heads laying over their backs and occasionally holding wings in the air. Food safety and public health were immediate local concerns.

To document the scope of the die-off, the US Fish and Wildlife Service and the US Geological Survey Alaska Science Center (USGS-ASC) contracted with island residents to survey accessible coast-

line and collect specimens for diagnostics and research. Twenty-one kilometers of coastline along the north side of the island were surveyed on 23–25 November 2013, during which 912 bird carcasses were recorded. Species found were Crested Auklets (*Aethia cristatella*) and murre (*Uria* spp.), with much smaller numbers of gulls (*Larus* spp.), Northern Fulmars (*Fulmarus glacialis*), eiders (*Somateria* spp.), a Black-legged Kittiwake (*Rissa tridactyla*), and an unidentified duck (*Anatidae* spp.). Responders were unable to conduct beach surveys on other sections of the island due to logistics, limited daylight, and deteriorating weather; therefore, the full extent of the die-off is unknown.

Specimens submitted to USGS National Wildlife Health Center, Madison, Wisconsin, consisted of three Crested Auklets, three Northern Fulmars, three Thick-billed Murres (*Uria lomvia*), four gulls (Glaucous [*Larus hyperboreus*] or Glaucous-winged [*Larus glaucescens*] or hybrids; species could not be determined), and one Common Eider (*Somateria mollissima*). Routine aerobic bacterial isolation was performed on the indicated tissues (Table 1) by using tryptic soy agar with 5% sheep blood (BD Diagnostics, Sparks, Maryland, USA) incubated for 18–24 h at 37 C, and bacterial isolates were identified by using biochemical test (API strips, BioMérieux, Marcy l'Étoile, France). *Pasteurella multocida* was isolated in liver from three Crested Auklets, three Thick-billed Murres, two Northern Fulmars, one Common Eider, and one gull by using the agrose gel precipitin test (Heddleston et al. 1972) and characterized as serotype 1, the most common serotype found in wild birds (Samuel et al. 2007). The bacterium was detected in liver from one gull by next-generation sequencing. Bacterial 16S rRNA gene amplicons were prepared for Illumina sequencing and combined into one multiplexed library by using established primers and protocols (Caporaso et al. 2012) at USGS-ASC.

Sequencing was performed by using two 2×150 PE cycles runs on Illumina MiSeq, each including a 15% PhiX spike. With QIIME (Caporaso et al. 2010), raw Illumina sequence data were quality filtered, joined and demultiplexed, and assigned to operational taxonomic units (OTUs; representing 97% DNA sequence similarity) picked by using the open-reference workflow (Caporaso et al. 2010). Representative OTU sequences were aligned to the GreenGenes 16S rRNA gene reference database, and taxonomy was assigned by using the RDP classifier (Cole et al. 2014). No other gross abnormalities, pathogenic bacteria, viruses, or parasites were detected in birds infected with *P. multocida*.

This unusual mortality event was the first confirmed report of avian cholera in wild birds in Alaska (Philo 1981). It was also unusual in its impact on species not commonly associated with avian cholera mortality. Samuel et al. (2005a) found that Greater White-fronted Geese (*Anser albifrons frontalis*) breeding in Alaska have antibodies to *P. multocida*. The closest geographic outbreaks were reported on Banks Island, Northwest Territories, Canada, and involved Lesser Snow Geese (Samuel et al. 1999). Outbreaks have been reported in Common Eiders on East Bay, Nunavut, Canada (Descamps et al. 2009). Christensen et al. (1997) reported mortality of Common Eiders at wintering areas within the sea ice in Denmark due to avian cholera in 1996. The disease was reported in Common Murre (*Uria aalge*) in the Baltic Sea (Samuel et al. 2007).

Changes in arctic and subarctic ecosystems due to global climate change may provide an increased opportunity for novel disease outbreaks. The discovery of avian cholera in marine bird populations in Alaska suggests that *P. multocida* may emerge as a significant pathogen in species previously not considered high risk. The potential impacts of avian cholera in this region are of special conservation concern for the threatened

TABLE 1. Diagnostic summary of marine birds found dead on Saint Lawrence Island, Alaska, USA, November 2013.

Species	Age	Sex	Body condition	Gross lesions observed	Organ	Bacterial isolates	Diagnosis
Thick-billed Murre (<i>Uria lomvia</i>)	After hatch year	Male	Excellent	Enteritis, enlarged liver with miliary white foci	Liver	<i>Pasteurella multocida</i>	Avian cholera
Northern Fulmar (<i>Fulmarus glacialis</i>)	After hatch year	Female	Fair	Enteritis, enlarged liver with miliary white foci	Lung	<i>P. multocida</i>	Avian cholera
Crested Auklet (<i>Aethia cristatella</i>)	After hatch year	Female	Excellent	Enteritis, enlarged liver with miliary white foci	Lung	<i>P. multocida</i>	Avian cholera
Crested Auklet (<i>A. cristatella</i>)	Unknown	Male	Good	Liver pale, intestine scavenged	Liver	<i>P. multocida</i>	Avian cholera
Crested Auklet (<i>A. cristatella</i>)	Unknown	Male	Good	Enteritis, liver pale	Lung	<i>P. multocida</i>	Avian cholera
Thick-billed Murre (<i>U. lomvia</i>)	Unknown	Male	Good	Thick yellow material in intestine, liver pale and mottled	Liver	<i>P. multocida</i>	Avian cholera
Thick-billed Murre (<i>U. lomvia</i>)	Unknown	Female	Good	Enlarged spleen	Lung	<i>P. multocida</i>	Avian cholera
Northern Fulmar (<i>F. glacialis</i>)	Unknown	Male	Good	Thick yellow material in intestine, liver pale and mottled	Lung	<i>P. multocida</i>	Avian cholera
Common Eider (<i>Somateria mollissima</i>)	After hatch year	Male	Excellent	Splenic necrosis, liver and intestine scavenged	Liver	<i>P. multocida</i>	Avian cholera
Northern Fulmar (<i>F. glacialis</i>)	Hatch year	Female	Good	Thick yellow material in intestine, miliary white foci in liver	Spleen Liver	<i>P. multocida</i> No bacterial growth	Suspect avian cholera
Unidentified gull (<i>Larus</i> spp.) ^a	After hatch year	Unknown	Good	Fibrinous pericarditis	Liver	<i>P. multocida</i>	Avian cholera
Unidentified gull (<i>Larus</i> spp.) ^a	Hatch year	Male	Good	Hemorrhagic enteritis, liver necrosis	Lung Liver	No bacterial growth <i>Plesiomonas shigelloides</i> , <i>Escherichia coli</i>	Gunshot; potential oil ingestion
Unidentified gull (<i>Larus</i> spp.) ^a	After hatch year	Female	Emaciated	Aspiration pneumonia, hemorrhagic enteritis	Liver	No bacterial growth	Aspiration pneumonia
Unidentified gull (<i>Larus</i> spp.) ^a	Hatch year	Male	Emaciated	Hemorrhagic enteritis, peritonitis, liver necrosis	Liver	<i>Salmonella</i> Typhimurium	Salmonellosis

^a Gulls submitted for testing could not be conclusively identified to species and may have been Glaucous-winged (*L. glaucescens*), Glaucous (*L. hyperboreus*), or hybrids.

Spectacled Eider (*Somateria fischeri*), and the world population overwinters in these waters (Petersen et al. 1999). Although *P. multocida* serotype 1 is not zoonotic, investigations and response to future mortality events in this region must consider public health and food security concerns, because many marine species are used as food by local communities.

We greatly appreciate the efforts of many individuals and organizations, including P. Rookok, P. Pungowiyi, D. Pungowiyi, D. Akeya, F. Kava, M. Kiyuthlook, and T. Noongwook, Native Village of Savoonga; E. Ungott, C. Irrigoo, H. Koonooka, A. Konahok, and R. Bushu, Native Village of Gambell; M. Koonooka and G. Noongwook, Alaska Eskimo Whaling Commission; R. Gerlach, Alaska Department of Environmental Conservation; L. Castrodale, Alaska Department of Health and Social Services; P. Bente and L. Hughes, Alaska Department of Fish and Game; V. Metcalf, Eskimo Walrus Commission; B. Ahmasuk, Kawerak Subsistence Program; R. Stimmelmayer, North Slope Borough Department of Wildlife Management; E. Taylor and D. Irons, US Fish and Wildlife Service; J. Pearce, L. Zeglin, and A. Reeves, US Geological Survey, Alaska Science Center. Use of trade or product names does not imply endorsement by the US government.

LITERATURE CITED

- Ahmasuk A, Trigg E. 2008. *Bering Strait region local and traditional knowledge pilot project: A comprehensive subsistence use study of the Bering Strait region. North Pacific Research Board Final Report, Project No. 643*. Anchorage, Alaska, 342 pp. http://doc.nprb.org/web/06_prjs/643_NPRB%20final%20report%201-10-2008.doc. Accessed April 2015.
- Caporaso JG, Kuczynski J, Stombaugh J, Bittinger K, Bushman FD, Costello EK, Fierer N, Gonzalez Peña A, Goodrich JK, Gordon JI, et al. 2010. QIIME allows analysis of high-throughput community sequencing data. *Nat. Methods* 7:335–336.
- Caporaso JG, Lauber CL, Walters WA, Berg-Lyons D, Huntley J, Fierer N, Owens SM, Betley J, Fraser L, Bauer M, et al. 2012. Ultra-high-throughput microbial community analysis on the Illumina HiSeq and MiSeq platforms. *ISME J* 6:1621–1624.
- Christensen TK, Bregnballe T, Andersen TH, Dietz HH. 1997. Outbreak of pasteurellosis among wintering and breeding Common Eiders *Somateria mollissima* in Denmark. *Wildl Biol* 3:125–128.
- Cole JR, Wang Q, Fish JA, Chai B, McCarrell DM, Sun Y, Brown CT, Porras-Alfaro A, Kuske CR, Tiedje JM. 2014. Ribosomal Database Project: Data and tools for high throughput rRNA analysis. *Nucleic Acids Res* 42:D633–D642.
- Descamps S, Gilchrist HG, Bety J, Buttler EI, Forbes MR. 2009. Cost of reproduction in a long-lived bird: Large clutch size is associated with low survival in the presence of a highly virulent disease. *Biol Lett* 5:278–281.
- Heddleston KL, Gallagher JE, Rebers PA. 1972. Fowl cholera: Gel diffusion precipitin test for serotyping *Pasteurella multocida* from avian species. *Avian Dis* 16:925–936.
- Petersen MR, Larned WW, Douglas DC. 1999. At-sea distribution of Spectacled Eiders: A 120-year-old mystery resolved. *Auk* 116:1009–1020.
- Philo LM. 1981. Avian cholera. In: *Alaskan wildlife diseases*, Dieterich RA, editor. Institute of Arctic Biology, University of Alaska Fairbanks, Fairbanks, Alaska, pp. 295–300.
- Piatt JF, Spinger AM. 2003. Advection, pelagic food webs and the biogeography of seabirds in Beringia. *Mar Ornithol* 31:141–154.
- Samuel MD, Botzler RG, Wobeser GA. 2007. Avian cholera. In: *Infectious diseases of wild birds*, Thomas NJ, Hunter DB, Atkinson CT, editors. Blackwell Publishing, Ames, Iowa, pp. 239–269.
- Samuel MD, Shadduck DJ, Goldberg DR. 2005a. Avian cholera exposure and carriers in greater white-fronted geese breeding in Alaska, USA. *J Wildl Dis* 41:498–502.
- Samuel MD, Shadduck DJ, Goldberg DR, Johnson WP. 2005b. Avian cholera in waterfowl: The role of lesser snow geese and Ross's geese as disease carriers in the Playa Lakes Region. *J Wildl Dis* 41:48–57.
- Samuel MD, Takekawa JY, Samelius G, Goldberg DR. 1999. Avian cholera mortality in lesser snow geese nesting on Banks Island, Northwest Territories. *Wildl Soc Bull* 27:780–787.
- Smith MA, Walker NJ, Free CM, Kirchoff MJ, Drew GS, Warnock N, Stenhouse IJ. 2014. Identifying marine Important Bird Areas using at-sea survey data. *Biol Conserv* 172:180–189.

Submitted for publication 2 December 2014.

Accepted 4 April 2015.