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Southward expansion of high pathogenicity avian influenza H5 in wildlife in South America: estimated impact on wildlife populations, and risk of incursion into Antarctica

OFFLU ad-hoc group on HPAI H5 in wildlife of South America and Antarctica

Andrew Breed: Epidemiology and One Health Section, Department of Agriculture, Fisheries and Forestry, Canberra, Australia. <u>andrew.breed@aff.gov.au</u>

Meagan Dewar: Future Regions Research Centre, Federation University Australia, Berwick Australia. <u>m.dewar@federation.edu.au</u>

Laura Dodyk: Aves Argentinas, Caba, Buenos Aires, Argentina. dodyk@avesargentinas.org.ar

Thijs Kuiken: Department of Viroscience, Erasmus University Medical Centre, Rotterdam, The Netherlands. <u>t.kuiken@erasmusmc.nl</u>

Ricardo Matus: Centro Bahía Lomas, Facultad de Ciencias, Universidad Santo Tomás, Chile. <u>rmatusn@gmail.com</u>

Patricia Pereira Serafini: Laboratório de Biomarcadores de Contaminação Aquática e Imunoquímica, Universidade Federal de Santa Catarina, Florianópolis, Brazil. <u>patricia.serafini@icmbio.gov.br</u>

Marcela Uhart: Karen C. Drayer Wildlife Health Center, School of Veterinary Medicine, University of California, USA. <u>muhart@ucdavis.edu</u>

Ralph Eric Thijl Vanstreels: Karen C. Drayer Wildlife Health Center, School of Veterinary Medicine, University of California, USA. <u>ralph_vanstreels@yahoo.com.br</u>

Michelle Wille: Centre for Pathogen Genomics, Department of Microbiology and Immunology, The University of Melbourne, at the Peter Doherty Institute for Infection and Immunology, Melbourne, Victoria, Australia. <u>Michelle.wille@unimelb.edu.au</u>

Disclaimer: This statement provides the point of view of independent OFFLU experts, and does not necessarily reflect the position of the parent organisations FAO and WOAH.

Abstract

This report summarizes the spread of high pathogenicity avian influenza of the subtype H5 (HPAI H5) clade 2.3.4.4b in South America since it was first detected in the region in October 2022, and evaluates the risk for incursion into Antarctica in the near future. The focus of the report is on HPAI H5 in wildlife, and not spread in poultry or rare spillover to humans, as these are the subject of other reports. We do recognize and fully support a One Health approach to HPAI H5: an integrated, unifying approach that aims to sustainably balance and optimize the health of people, animals (both wild and domestic) and ecosystems.

Since HPAI H5 was first reported in Colombia in October 2022 (WAHIS, 2022a), it has spread across South America with devastating consequences for its wild bird and mammal populations (Gamarra-Toledo et al, 2023b; Leguia et al, 2023). In Peru and Chile, more than 500,000 wild birds of at least 65 species and more than 20,000 wild mammals of at least 15 species were reported dead, with actual mortality likely many times larger. This mortality is a potential threat to the conservation of several wild animal species. For example, HPAI-H5-associated losses included about 36% of the endangered Peruvian pelican (*Pelecanus thagus*) population in Peru, about 13% of the vulnerable Humboldt penguin (*Spheniscus humboldti*) population in Chile, and about 9% of the South American sea lion (*Otaria flavescens*) population in Peru and Chile.

After its first detection in South America, HPAI H5 virus has spread rapidly to the southern tip of South America (south of 40°S latitude), a distance of more than 6000 km in just three months. The recently reported detection of this virus in a South American sea lion at Puerto Williams, Chile (54.9°S), is the southernmost detection of the virus to date. There is a substantial risk it will continue southwards and in the near future reach Antarctica and its offshore islands, which are home to more than 100 million breeding birds, 6 species of pinnipeds and 17 species of cetaceans. This risk may be increased in coming months due to the spring migration of wild birds from South America to breeding sites in the Antarctic. The negative impact of HPAI H5 on Antarctic wild birds and mammal populations could be immense, both because of their likely susceptibility to mortality from this virus, and their occurrence in dense colonies of up to thousands of pinnipeds and hundreds of thousands of birds, allowing efficient virus transmission.

Although little can be done to stop HPAI H5 from spreading to Antarctica and causing mortality in Antarctic wild birds and mammals, there are several detection and response options possible. These include surveillance and accurate documentation of HPAI-H5-associated mortality events, and following guidelines to reduce risk of human-mediated virus spread.

1. Emergence and spread of HPAI H5 from 1996 until 2022

The currently circulating HPAI H5N1 is derived from viruses of the goose/Guangdong/96- (gsGD)lineage that were first detected in commercially-farmed geese in China in 1996 and have circulated and evolved in poultry. Multiple strains of virus within the gsGD lineage spread across east and southeast Asia in 2003-04, some of which was the result of spread by wild birds (e.g. to Japan and Republic of Korea). In 2005 the first intercontinental wave of transmission of gsGD viruses occurred across Eurasia and into Africa and has been followed by multiple waves of intercontinental spread. Viruses in the gsGD-lineage viruses have evolved into 5th order clades and have formed multiple genotypes (Pohlmann et al, 2022). The A(H5N1) virus that emerged in 2020 belongs to clade 2.3.4.4b and has caused numerous outbreaks in wild birds in Asia and Europe, typically during autumn and winter, as well as in Africa, but has persisted year-round in wild birds in Europe since 2021. That same year, HPAI H5 spread across the Atlantic Ocean to North America (Caliendo et al, 2022), where it spread rapidly across the continent in 2022 and southwards to Central and South America.

2. HPAI H5 in wildlife of South America, 2022 to 2023: rapid spread and conservation impact

Since its first detection in wild birds in Colombia on 18 October 2022 (WAHIS, 2022a), HPAI H5 virus spread rapidly in wildlife populations across Central and South America. In Central America, HPAI H5 was first detected in December 2022 in Panama (Promed mail, 2022) and Honduras (WAHIS, 2022b), followed by Costa Rica (WAHIS, 2023g) and Guatemala (WAHIS, 2023i) in January 2023. In February 2023, HPAI H5 was reported in several captive wild birds at the Zoological Garden of Havana, Cuba (WAHIS, 2023h). Overall, reported wild bird mortalities in Central America were relatively low (hundreds) and mostly restricted to brown pelicans (*Pelecanus occidentalis*).

Initially, HPAI H5 detections in South America were largely restricted to the Pacific coast, spreading through Ecuador and Peru (November 2022), and then Chile (December 2022), corresponding to the Pacific Americas flyway. In the affected regions, which are partially separated from the rest of South America by the physical barrier of the Andes mountains, there was extensive mortality of wild birds (mostly seabirds) and aquatic mammals associated with HPAI H5 virus detections in the subsequent months. By 21 July 2023, the Peruvian government reported the deaths of 519,541 seabirds, with 229,554 Peruvian boobies (Sula variegata), 201,047 guanay cormorants (Leucocarbo bougainvillii), 57,335 Peruvian pelicans (Pelecanus thaqus), 5,573 Inca terns (Larosterna inca), in addition to other species of boobies, penguins, gulls, and others (Peru Ministerio de Salud, 2023). In Chile, by 9 May 2023, the Agricultural and Livestock Service (Servicio Agrícola y Ganadero – SAG) reported the deaths of 27,977 seabirds, with 6,506 Peruvian boobies, 6,380 guanay cormorants, 4,594 kelp gulls (Larus dominicanus), 4,509 Peruvian pelicans, 1,016 grey gulls (Larus modestus), 726 neotropic cormorants (Phalacrocorax brasilianus), 640 Humboldt penguins (Spheniscus humboldti), in addition to other species of cormorants, gulls, vultures, swans, petrels, shorebirds, and others (Chile Servicio Agricola y Ganadero, 2023). Additionally, by 21 July 2023, the Chilean National Fisheries and Aquaculture Service reported the deaths of 2,517 Humboldt penguins and 460 Magellanic penguins (Spheniscus magellanicus) (Chile Servicio Nacional de Pesca y Acuicultura, 2023). Aquatic mammals were also heavily affected in both countries, with Peru reporting the deaths of 9,414 marine mammals by 21 July 2023, of which 9,314 were South American sea lions (Otaria flavescens) (Peru Ministerio de Salud, 2023), and Chile reporting the deaths of 16,856 South American sea lions, 61 pinnipeds of other species, 39 otters, and 106 cetaceans by 21 July 2023 (Chile Servicio Nacional de Pesca y Acuicultura, 2023). HPAI H5 virus infection was confirmed in two species of otters in the region: marine otter (Lontra felina) and southern river otter (Lontra provocax) (Chile Servicio Nacional de Pesca y Acuicultura, 2023). HPAI H5 virus infection also was confirmed in three species of cetaceans in the region: Chilean dolphin (Cephalorhynchus eutropia), common dolphin (Delphinus delphis), and spiny porpoise (Phocoena spinipinnis) (Chile Servicio Nacional de Pesca y Acuicultura, 2023; Peru Ministerio de Salud, 2023). Because only a small proportion of animals found dead along the coast of Peru and Chile were tested for HPAI H5, it is possible that some mortalities were due to causes other than HPAI H5. Nevertheless, the numbers of seabirds, sea lions and otters found dead in these countries were orders of magnitude greater than during the same months in previous years, and it is reasonable to presume the virus was responsible for the majority of these deaths. Also, the numbers of wild animals reported dead are undoubtedly only a fraction of the numbers of individuals that died, many probably at sea, so that these figures greatly underestimate the actual level of wildlife mortality from HPAI H5.

The spread of HPAI H5 in South America to the east of the Andes was not accompanied by the same level of reported wildlife mortality as along the Pacific coast of the continent. Although HPAI H5 was initially recorded on the coast of Venezuela on 17 November 2022 (WAHIS, 2022c), with the deaths

of 172 brown pelicans, no further detections were reported to the east of the Andes until the virus was detected in Bolivia (January 2023) (WAHIS, 2023c), and then in northern Argentina (WAHIS, 2023a) and Uruguay (WAHIS, 2023j) (February 2023). It is still unclear whether the virus was transported to this region from the Caribbean across the Amazonian region, e.g. via the Occidental or Central Amazon flyways (de Oliveira et al, 2016) or from Peru or Chile across the Andes. Following the arrival of HPAI H5 in northwestern Argentina (first case in Andean geese [Chloephaga melanoptera]), spread to Uruguay likely occurred through aquatic birds migrating along the westeast flyways from the lower Paraná River valley to the coastal wetlands of south Brazil and Uruguay (Antas, 1994). During the subsequent months, HPAI H5 was detected in aquatic birds at multiple sites in Bolivia, Argentina and Uruguay, and later reported from Paraguay (June 2023), without associated reports of high mortality in wildlife populations. Black-necked swans (Cygnus *melancoryphus*), ducks and coots were species in which HPAI H5 was most frequently detected at the time in these countries. Brazil reported its first detection of HPAI H5 in seabirds in May 2023 (WAHIS, 2023d), followed by a number of reported HPAI H5 detections in seabirds along its eastern and southeastern coastline in subsequent months. At the time of early cases, HPAI H5 detections in Brazil were mainly in Cabot's terns (Thalasseus acuflavidus) and royal terns (Thalasseus maximus), which were congregating to breed on coastal islands, but there were also sporadic detections in other species of aquatic birds (e.g. swans, gulls, cormorants) and birds of prey (Brasil Ministério da Agricultura e Pecuária, 2023). Relatively small numbers of seabirds were found at some of the affected sites in Brazil and are still under investigation, but no large-scale wildlife mortalities have been reported in South America to the east of the Andes. A large-scale mortality (thousands) of penguins and other marine animals was recorded along the coast of Uruguay in mid July 2023, however preliminary diagnostic tests and necropsy results from a small subset of individuals suggest that the primary causes of mortality were starvation and parasitism. The only reports of HPAI H5 detections in wild mammals east of the Andes have been 7 of 21 South American sea lions found dead in Río Grande (53.5°S), Tierra del Fuego, Argentina, that tested positive for HPAI H5 on 11 August 2023 (Argentina Ministerio de Economía, 2023a), and two South American sea lions—one with neurological signs, the other found dead—in a large sea lion breeding colony in Punta Berneja (41.1°S), province of Río Negro, Argentina, that tested positive for HPAI H5 on 16 August 2023 (Argentina Ministerio de Economía, 2023b) (Figure).

The impacts of this HPAI H5 outbreak on wildlife in South America are enormous. Firstly, the immense numbers of lives lost: more than 500,000 wild birds and more than 20,000 wild mammals, involving at least 80 different species, reported dead so far, with actual mortality likely many times larger. This has a direct conservation impact on multiple wild bird and mammal species that are already threatened from other causes. This includes species listed by the International Union for Conservation of Nature (IUCN) as being under threat of extinction in the near future in nine categories, including "near-threatened" (likely to become endangered in the near future), "vulnerable" (high risk of extinction in the wild), and "endangered" (higher risk of extinction in the wild), and based on criteria such as rate of decline, population size, area of geographic distribution, and degree of population and distribution fragmentation (<u>https://www.iucnredlist.org/</u>). Based on the number of individuals found dead, the most significant conservation impact for seabirds has been on Humboldt penguins (vulnerable), with estimated losses of about 13% of the Chilean population, and Peruvian pelicans (endangered in Peru), with loss of about 36% of the Peruvian population. There is also conservation concern due to the loss of individuals of near-threatened species, such as red-legged cormorant (*Poikilocarbo gaimardi*), sooty shearwater (*Ardenna grisea*), elegant tern (Thalasseus elegans) and Inca tern. Among mammal species, a significant conservation impact has been on the South American sea lion, with a loss (based on reported deaths alone) of at least 9% of the populations in Peru and Chile (Cárdenas-Alayza et al, 2016). The highest conservation impact among mammals is for endangered species such as the marine otter and the southern river otter, and of near-threatened species such as the Chilean dolphin and the spiny porpoise.

3. Detection of HPAI H5 at the southern tip of South America, and risk for spread to Antarctica

HPAI H5 virus has spread to the southern tip of South America very rapidly and there is a substantial risk it will continue southwards and in the near future reach Antarctica and its offshore islands, which provide breeding habitat for over 100 million birds every spring, and which are inhabited by 6 species of pinnipeds and 17 species of cetaceans (Harris et al, 2015; Wilson & Reeder, 2005). This risk may be increased in coming months due to the spring migration of wild birds from South America to breeding sites in the Antarctic. The negative impact of this virus on Antarctic wildlife could be immense—likely worse than that on South American wildlife—because many Antarctic species of birds and mammals are closely related to species in Eurasia and the Americas that are known to have suffered high mortality from HPAI H5 virus infection, and because many Antarctic species occur in dense colonies of up to thousands of pinnipeds and hundreds of thousands of birds, allowing efficient virus transmission among individuals.

After its detection in October 2022 in Colombia, the northernmost country in South America, HPAI H5 virus has spread rapidly to the southern tip of South America (south of 40°S latitude), traversing the distance of more than 6000 km in just three months. The first detection of HPAI H5 in the southern tip of South America was on 18 January 2023, when the virus was found in a kelp gull in the city of Ancud, Chile (41.9°S) (WAHIS, 2023f) (Figure). No further infections were recorded in the region until late March, when the virus was detected in Rawson, Argentina (43.3°S) (WAHIS, 2023b), and in Puerto Natales, Chile (51.7°S) (WAHIS, 2023f). Additional cases were recorded in the region during April and May 2023, demonstrating the circulation of the virus in aquatic birds (swans, gulls, skuas, ducks, grebes, terns, and coots), terrestrial birds (parakeets and caracaras) and aquatic mammals (sea lions and river otters) in southern Chile and in aquatic birds (swans, ducks, and gulls) in southern Argentina. In early April 2023, the virus was detected in black-necked swans at Estela (53.4°S), representing the first detection in Tierra del Fuego (WAHIS, 2023f). The southernmost reported detection to date was 14 June 2023, when two South American sea lions were found dead near each other at Puerto Williams, on the Beagle Channel in Chile (54.9°S), one of which tested positive for HPAI H5 (Polar Comunicaciones, 2023) (WAHIS, 2023e). This places the virus approximately 550 km southwest of the Falkland Islands (Islas Malvinas), 950 km northwest of the South Shetland Islands (Islas Shetland del Sur) and 1080 km northwest of the Antarctic Peninsula.

Recent risk assessments by the Antarctic Wildlife Health Network of the Scientific Committee on Antarctic Research have analysed the risks of incursion of HPAI H5 into Antarctica from nearby regions where it is present, including South America and South Africa (Dewar et al, 2023). Antarctica, together with Oceania (including Australia and New Zealand), are the only continents still free of HPAI H5 virus (Dewar et al, 2022; Wille & Klaassen, 2023)

From its current presence in South America, the risk of HPAI H5 incursion into Antarctic/Subantarctic wildlife is considered greatest for the Falkland Islands (Islas Malvinas), due to their relative proximity to mainland South America (circa 500 km; Figure) and the potential movement of aquatic birds, in addition to the presence of large resident populations of wild Anseriformes (including black-necked swans, one of the species most affected by HPAI H5 in southern Chile) and pinnipeds (including South American sea lions). Falkland Islands (Islas Malvinas) also has an endemic species of Anseriform, the Falkland flightless steamer duck (*Tachyeres brachypterus*); given HPAI H5 impacts on other species of steamer ducks in South America (Fuegian steamer ducks, *Tachyeres pteneres*, in Chile), the virus could pose a significant threat to the endemic Falkland steamer duck. Unlike most other islands in the Subantarctic region, Falkland Islands (Islas Malvinas) are permanently inhabited by people (c. 3660 inhabitants) and have small-scale poultry farming, which could also contribute to the spread and impacts of HPAI H5. The presence of birds of prey such as vultures, caracaras, hawks

and falcons is also of concern, since these birds could become exposed to HPAI H5 through the consumption of infected wildlife, as has occurred on other continents.

The Scotia Arc is also of particular concern with regards to the potential spread of HPAI H5 in the Southern Ocean. The Scotia Arc is a chain of islands between the southern tip of South America and the Antarctic Peninsula, including South Georgia Islands (Islas Georgia del Sur), the South Sandwich Islands (Islas Sándwich del Sur), the South Orkney Islands (Islas Orcadas del Sur), and the South Shetland Islands (Islas Shetland del Sur) (Figure). Although South Georgia (Islas Georgia del Sur) is remote (c. 2000 km from mainland South America), its eastward location implies a greater similarity in terms of wildlife composition with South America and the Falkland Islands (Islas Malvinas) than other areas located further south. It is also worrying in that South Georgia (Islas Georgia del Sur) has a substantial resident population of non-endemic Anseriformes (yellow-tailed pintails, Anas georgica, and speckled teal, Anas flavirostris), which could also contribute to the spread and impact of HPAI H5 on the island. The Antarctic Peninsula and the remainder of the Scotia Arc-South Shetland Islands (Islas Shetland del Sur), South Orkney Islands (Islas Orcadas del Sur), and South Sandwich Islands (Islas Sándwich del Sur)—have more barren terrestrial environments that lack resident populations of Anseriformes, sea lions or birds of prey. Nevertheless, these areas are home to large colonies of seabirds (especially penguins), Antarctic fur seals (Arctocephalus gazella) and southern elephant seals (Mirounga leonina), which are known or likely to be susceptible to HPAI H5. The circulation of low pathogenicity avian influenza (LPAI) strains (subtypes H4N7, H5N5, H6N8, and H11N2) was previously demonstrated in Adélie (*Pygoscelis adeliae*) and chinstrap penguins (Pygoscelis antarcticus), southern giant petrels (Macronectes giganteus), snowy sheathbills (Chionis albus), and brown skuas (Stercorarius antarcticus) (Barriga et al, 2016; De Seixas et al, 2022; Hurt et al, 2016; Hurt et al, 2014; Ogrzewalska et al, 2022; Petersen et al, 2017). This indicates that these species provide favourable epidemiological conditions for the transmission and persistence of influenza viruses, and suggests that if introduced, HPAI H5 strains could spread and cause significant impacts on these populations.

A number of avian species have been identified as potential vector species for viral introduction and spread through the region, including skuas, gulls, giant petrels, and sheathbills. LPAI has already been detected in many of these species, demonstrating susceptibility to influenza viruses. Many of these species are known to traverse the Drake Passage and connect South America with the Sub-Antarctic Islands and Antarctic Peninsula through nomadic and migratory movements (Figure). This includes spring migration of several seabird species from South America to the Antarctic. For example, snowy sheathbills and southern fulmars (Fulmarus glacialoides), may spend the Southern Hemisphere winters in South America, and the Southern Hemisphere summers in the Antarctic (Quintana & Travaini, 2000; Zamora et al, 2023). Predatory behaviour of skuas and giant petrels leads to visitation of numerous sites along the southern tip of South America, Sub-Antarctic Islands and the Antarctic Peninsula. These predatory/scavenging habits and frequent incursions into breeding colonies of other species, suggest these species are also likely to play a significant role in the spread of HPAI H5 among sites in the Subantarctic and Antarctic regions. For example, the breeding range of the southern giant petrel extends from c. 40°S (Gough Island) to nearly 68°S in West Antarctica. There are some differences in the breeding chronology of southern and northern populations, but usually birds arrive at colonies from July/August through September, depending on latitude and location. At the Antarctic sites, the southern giant petrel breeds annually in loose colonies, with eggs generally laid from mid-October to mid-November (ACAP, 2010). Brown skuas are fully migratory and are found on or around Subantarctic Islands populated by burrow-nesting seabirds or penguins. They are highly predatory, feeding mainly on other birds but will also scavenge around fishing boats and feed at sea. Breeding begins in October and November. Birds are loosely colonial but highly territorial, nesting on grass, gravel or bare rock (Anderson et al, 2009). Brown skuas are commonly recorded in high numbers in Antarctica; Argentina; Australia; Falkland Islands

(Islas Malvinas), New Zealand; South Georgia (Islas Georgia del Sur) and the South Sandwich Islands (Islas Sándwich del Sur).

The potential for human activities (e.g. tourism or research) to accidentally contribute to the dissemination of HPAI H5 to Antarctica and Subantarctic islands should not be disregarded.

The remainder of the Subantarctic and Antarctic region may also be vulnerable to the spread of HPAI H5. Several seabirds such as albatross species are known to perform circumpolar movements in the Southern Ocean, and therefore could support the longitudinal transport of HPAI H5 among Subantarctic islands (e.g. vagrant/visitor individuals) or transmission during interactions at sea (e.g. albatrosses and petrels aggregating at foraging sites or near fishing vessels). Concurrently, some species of Charadriiformes (e.g. arctic terns, Sterna paradisaea, long-tailed jaegers, Stercorarius longicaudus, south polar skuas, Stercorarius maccormicki), perform transhemispheric migrations that could also provide opportunities for the latitudinal transport of HPAI H5 to the Subantarctic and Antarctic regions. For example, the south polar skua spends the Southern Hemisphere winter in the North Pacific and North Atlantic and breeds on relatively snow-free areas in Antarctica in the Southern Hemisphere summer (Kopp et al, 2011). It is usually reliant on fish, with predation on seabirds being of variable importance. It can, however, survive exclusively on penguins where a breeding colony is associated with a penguin rookery. Endemic species of Anseriformes are present at Crozet, Kerguelen, Auckland, and Campbell Islands, and could be vulnerable to HPAI H5 outbreaks. Kerguelen terns (Sterna virgatta), which are endemic to Kerguelen, Prince Edward and Crozet Islands, could also be at risk considering the severe impacts that HPAI H5 has had on tern populations in southern Africa and Europe. Although cormorants were not traditionally considered significant hosts of influenza viruses, the large-scale mortality of these birds during HPAI H5 outbreaks in southern Africa and Peru raises concern about the potential impacts that HPAI H5 could have on endemic species of cormorants present at Kerguelen, Campbell, Auckland, Macquarie, and Bounty Islands. Similarly, although parakeets are not usually considered highly susceptible to avian influenza, at least two clusters of mortality of slender-billed parakeets (Enicognathus leptorhynchus) were attributed to HPAI H5 in southern Chile in April and May 2023; the potential threat posed by HPAI H5 to parakeet species that are endemic to the Auckland and Antipodes Islands should therefore not be overlooked. HPAI H5 poses a potential conservation threat to other avian species that are partly or completely restricted to the Subantarctic and Antarctic regions and are already under threat of extinction from other causes, such as the southern rockhopper penguin (Eudyptes chrysocome) (vulnerable), the emperor penguin (Aptenodytes forsteri) (near-threatened), and the sooty shearwater (near-threatened), among others (https://www.iucnredlist.org/).

HPAI H5 could also have a severely negative impact on pinniped and cetacean populations present in the Subantarctic and Antarctic region. All six Antarctic pinniped species belong to the families Otariidae or Phocidae, and 5 of 17 Antarctic cetacean species belong to the families Delphinidae or Phocoenidae (https://www.iucnredlist.org/). Pinnipeds and cetaceans have been reported with fatal infection due to HPAI H5 virus along the Pacific coast of South America, or along the Atlantic coasts of North America and Europe (European Food Safety Authority et al, 2023). Pinnipeds of concern include the South American fur seal, the endangered New Zealand sea lion (*Phocarctos hookeri*), the Antarctic fur seal, all belonging to the family Otariidae, and the leopard seal (*Hydrurga leptonyx*) and the crabeater seal (*Lobodon carcinophaga*), both belonging to the family Phocidae. The Falkland Islands (Islas Malvinas) population of the South American fur seal, with 7,500 individuals (Cárdenas-Alayza et al, 2016), is of concern given the high mortality of this species from HPAI H5 virus infection in Peru (Gamarra-Toledo et al, 2023a). The New Zealand fur seal is of concern because of its close relationship to the South American fur seal, which suffered mortality from HPAI H5 virus infection in Peru (Gamarra-Toledo et al, 2023a), and because it is one of the world's rarest sea lion species, with a population of around 10,000 individuals at New Zealand's subantarctic Auckland and Campbell

Islands (Chilvers, 2015). The Antarctic fur seal is of concern because of a similar close relationship to the susceptible South American fur seal and its clustered distribution, with approximately between 4.5 and 6.2 million individuals, 95% of the global population, around South Georgia Island (Islas Georgia del Sur) (Croxall, 1992). The leopard seal is of concern because of its close relationship to the harbour seal (*Phoca vitulina*), which suffered high mortality from HPAI H5 virus infection in the USA (Puryear et al, 2023), and because a considerable proportion of its diet consists of birds, particularly penguins, so that it would be at risk from predation on infected birds. Its population numbers about 35,500 individuals and is widely distributed in Antarctic and Subantarctic waters of the Southern Hemisphere (Hückstädt, 2015). The crabeater seal also is of concern because of its close relationship to the susceptible harbour seal. The crabeater seal has a circumpolar distribution around the coast of Antarctica, and is by far the most abundant seal species in the world, consisting of at least 7 million and possibly as many as 75 million individuals (Adam, 2005). HPAI H5 virus outbreaks in these species would likely cause thousands to millions of deaths, as well as knock-on effects on the ecosystems to which they belong.

4. Options for response regarding potential incursion of HPAI H5 into Antarctica

Now that HPAI H5 virus is circulating in wild birds and mammals and has spread across South America, there is little that can be done to stop it from spreading to Antarctica and from causing morbidity and mortality in Antarctic wild birds and mammals. Measures that can be applied to poultry farms, such as housing orders, biosecurity barriers, and vaccination, generally do not apply to wildlife populations. Vaccination of critically endangered species may be an exception, as is being planned for the California condor (*Gymnogyps californianus*) in North America (USFWS, 2023). Mass vaccination using live virus (vector) vaccines for colonial wildlife should be regarded as an urgent priority for research. Furthermore, the development of vaccines that can be delivered via Remotely Piloted Aircraft Systems (RPAS) could be a valuable approach for vaccination of colonial wildlife in Antarctica, always with careful consideration of the potential disturbance and special regulations of RPAS operation in the Antarctic region (Antarctic Treaty System, 2018; COMNAP, 2017; Hodgson & Koh, 2016). Nevertheless, until this technology has been developed there are other options for response in Antarctica that can be considered. More details about these options are provided below, with possible solutions to challenges of implementing them in Antarctica.

a. Recording information of mortality events associated with HPAI H5

Current surveillance systems for HPAI in wildlife report the detection of HPAI virus but not necessarily the number of wild animals found diseased or dead. Well-documented descriptions of HPAI outbreaks in wildlife are important to evaluate the impact of this disease on wildlife populations. This information can help wildlife managers to adapt conservation plans, and can help policymakers with planning to prevent future HPAI outbreaks, not only in poultry and humans, but also in wildlife.

When visiting a site with unusual mortality, it is relevant to collect the following information and samples:

- To determine the extent of the outbreak, record:
 - Number of sick and dead birds/mammals and total number present. Carcasses can be counted from outside the colonies using binoculars and/or from photographs. If a decision is made to enter the colony, and when possible, corpses should be marked to avoid double counts (e.g. clipping primaries of birds or spraying paint; carcasses do not have to be touched either way).
 - \circ Species.
 - o Age.

- o Sex.
- Any ring numbers on birds or tags on mammals.
- Photos of sick and dead animals as supporting evidence.
- To substantiate the cause of the outbreak:
 - If testing is available, collect a number of fresh carcasses for pathological and/or virological analysis in plastic bags (after consultation with relevant authorities); as an alternative to whole carcasses, swabs may be collected for virological analysis.
 - \circ $\;$ Describe and/or record on video the clinical signs in affected birds/mammals.

Potential challenges for collection, storage, and analysis of samples to diagnose HPAI at remote field sites may include the absence of specialized consumables, equipment and infrastructure, trained personnel and restrictions of exporting/importing samples to certified facilities outside of Antarctica/Subantarctic regions. However, there are possible solutions for collecting and storing samples for HPAI diagnosis (EWDA avian influenza diagnosis card; Reperant et al, 2021) using more generally available consumables and storage conditions in resource-limited settings. These include the following:

- While the preferred storage medium is RNA Shield, RNALater or similar RNA stabilization solution (Wille et al, 2017), dracon/polyester or cotton-tip swabs (e.g. from throat and cloaca of birds, from nose and throat of mammals) can be collected in 100% ethanol and stored for up to several months at room temperature and still provide good results for detection of viral RNA by PCR (Krafft et al, 2005).
- While the preferred sample for detection of antibodies against AI in blood is serum obtained by centrifugation of clotted blood, blood samples also can be absorbed onto filter paper strips and stored for several months at room temperature and still provide good results for antibody detection (Dusek et al, 2011).

If people trained in pathology are available, postmortem tissue samples (e.g. brain, trachea, lung, heart, pancreas, liver, spleen, kidney; EWDA avian influenza diagnosis card) for pathological diagnosis of AI can be collected in 10% neutral-buffered formalin, transferred after two weeks to 70% ethanol, and stored for up to several months at room temperature and still provide good results for both pathological diagnosis and next-generation sequencing of AI (Bodewes et al, 2015).

If people trained in virology are available and it is possible to access a MinION sequencer, it is possible to perform rapid and advanced virus sequencing on-site in the Antarctic environment (Johnson et al, 2017). Both clinical and environmental samples can be analysed on-site by use of a Nanopore MinION-based influenza sequencing approach making it possible to sequence the whole genome of AI viruses, and determine the subtype and clades (Miah et al, 2023).

Overall, it is imperative that relevant permits are in place, including CITES (Convention on International Trade in Endangered Species of Wild Fauna and Flora) for international shipment of samples from listed species, that testing and/or sample transport pathways are identified prior to sample collection, and that any testing performed (in Antarctica) is done within biosafety standards of the relevant jurisdiction. It is also necessary to follow relevant health and safety guidelines, including use of appropriate personal protective equipment (PPE), to avoid human infection (WOAH 2022).

b. Limiting amplification of an HPAI H5 outbreak at a site by removing carcasses.

Prior to consideration of bird carcass removal, a thorough risk assessment needs to be performed. Disadvantages include disturbance of birds or mammals, with a chance that animals abandon the colonies or haulout sites; human-caused spread of virus (see below), and a (very small) risk of

infection of people who visit the breeding colony when wearing appropriate protective equipment. An additional complication in Antarctica is the difficulty of carcass disposal, and in the case of mammals, the biomass of animals. Carcasses may not be buried or composted, and there are no, or few, incineration facilities available on the continent. Therefore, carcass removal should only be considered for birds and if disposal mechanisms are available, which may include relocation of carcasses from colonies to low-risk settings, or collection for disposal off the continent. Finally, carcass removal should only be considered when trained personnel and appropriate PPE are available. The main advantage of carcass removal is to potentially reduce virus spread as carcasses that may remain infectious for days to months (Bregnballe et al, 2023; Ramey et al, 2020; Yamamoto et al, 2017), and there is limited evidence from tern breeding colonies in the northern hemisphere that it can improve survival of birds (Rijks et al, 2022). However, further research and validation is required to confirm that this activity is indeed beneficial across species and ecological settings.

If bird carcass removal is being undertaken, appropriate hygiene measures are important:

- Wear adequate PPE, and perform adequate washing and disinfection afterwards (including footwear) to prevent contamination of other areas. Use most direct route to infected carcasses, to prevent spread to non-infected areas of the site.
- Suitable disinfectants for HPAI virus include agents based on ethanol (e.g. Sterillium) or sodium hypochlorite (e.g. Clorox), after washing with soap and water and removing all organic material.
- Transport carcasses in double plastic bags to the place designated by the relevant authorities for further disposal.
- For specific details on PPE and other hygiene measures, see websites of the relevant authorities.

c. Reducing risk of human-caused spread of HPAI H5

In HPAI outbreaks limited to poultry, people may spread virus from one poultry farm to another through contaminated clothing or footwear, or through contaminated equipment such as vehicles. In the current HPAI H5 outbreak that also involves wildlife, similar risks are present for people working with, or close to, wildlife. The following prevention options reduce the risk for human-caused HPAI H5 incursion into Antarctica and for HPAI H5 spread between different locations in Antarctica. More detailed information on preventative and response options can be found in the report by the Antarctic Wildlife Health Network of the Scientific Committee on Antarctic Research "The risk of avian influenza in the Southern Ocean: A practical guide for operators interacting with wildlife" (Dewar et al, 2022)

- Clean and disinfect clothing, footwear, and field equipment prior to arrival in Antarctica and in between visits to different wildlife areas.
- Observe wildlife from a distance for signs of illness (in particular neurologic signs) and mortality suggestive of HPAI H5 before visiting a wildlife area, and cancel the visit if HPAI H5 is suspected and report the event to the relevant authorities.
- Wear adequate PPE when working around wildlife.

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Figure. Map of the southern tip of South America and western Antarctica, indicating localities and island groups mentioned in the text. Notable locations in South America mentioned in the text are denoted by a red circle.

