BioOne COMPLETE

Serologic Evidence of Influenza A and B Viruses in South American Fur Seals (Arctocephalus australis)

Authors: Blanc, Andrea, Ruchansky, Dora, Clara, Mario, Achaval, Federico, Le Bas, Alfredo, et al.

Source: Journal of Wildlife Diseases, 45(2) : 519-521

Published By: Wildlife Disease Association

URL: https://doi.org/10.7589/0090-3558-45.2.519

BioOne Complete (complete.BioOne.org) is a full-text database of 200 subscribed and open-access titles in the biological, ecological, and environmental sciences published by nonprofit societies, associations, museums, institutions, and presses.

Your use of this PDF, the BioOne Complete website, and all posted and associated content indicates your acceptance of BioOne's Terms of Use, available at www.bioone.org/terms-of-use.

Usage of BioOne Complete content is strictly limited to personal, educational, and non - commercial use. Commercial inquiries or rights and permissions requests should be directed to the individual publisher as copyright holder.

BioOne sees sustainable scholarly publishing as an inherently collaborative enterprise connecting authors, nonprofit publishers, academic institutions, research libraries, and research funders in the common goal of maximizing access to critical research.

Downloaded From: https://complete.bioone.org/journals/Journal-of-Wildlife-Diseases on 28 Mar 2024
Terms of Use: https://complete.bioone.org/terms-of-use

Journal of Wildlife Diseases, 45(2), 2009, pp. 519–521
© Wildlife Disease Association 2009

Serologic Evidence of Influenza A and B Viruses in South American Fur Seals (*Arctocephalus australis)*

Andrea Blanc,[1] Dora Ruchansky,[1] Mario Clara,[2] Federico Achaval,[2] Alfredo Le Bas,[3] and Juan Arbiza[1,4]
[1] Sección Virología, Facultad de Ciencias-Universidad de la República, Iguá 4225, CP: 11400 Montevideo, Uruguay; [2] Sección Zoología, Facultad de Ciencias-Universidad de la República, Iguá 4225, CP: 11400 Montevideo, Uruguay; [3] Sección Fisiología, Facultad de Ciencias-Universidad de la República, Iguá 4225, CP: 11400 Montevideo, Uruguay; [4] Corresponding author (email: jarbiza@fcien.edu.uy)

ABSTRACT: The aim of this work was to detect serologic evidence of influenza virus infections in South American fur seals (*Arctocephalus australis*) that inhabit the Uruguayan coast. In 29 of 37 serum samples that were analyzed, we identified antibodies to at least one of the following antigens: H1N1 (A/NewCaledonia/20/99), B/Beijing/184/93-like viruses, B/Hong Kong/330/01, and B/Sichuan/379/99 by means of the hemagglutination inhibition assay (HAI). Results confirmed that influenza A viruses circulate in marine mammals and also showed, for the first time, indirect evidence of influenza B infections in *Arctocephalus australis*.

Key words: *Arctocephalus australis*, hemagglutination inhibition assay, influenza virus, South American fur seals, Uruguay.

Influenza viruses belong to the family Orthomyxoviridae, which have enveloped and segmented, single-stranded negative-sense RNAs. There are three types of influenza viruses: A, B, and C, of which only types A and B cause widespread outbreaks in humans.

Influenza A virus infects a variety of avian and mammalian species including humans, horses, swine, and marine mammals such as seals and cetaceans (Webster et al., 1992; Murphy and Webster, 1996). It was believed that influenza B virus only infects humans, but in 2000 an influenza B virus isolate from an infected harbor seal (*Phoca vitulina*) was reported (Osterhaus et al., 2000).

Since the end of the 1970s, evidence of influenza A infections in marine mammals has been reported based on virus isolation (Lvov et al., 1978; Lang et al., 1981; Webster et al., 1981; Geraci et al., 1982; Hinshaw et al., 1982, 1984, 1986; Callan et al., 1995) or on antibody detection (de Boer et al., 1990; Steuen et al., 1994; Danner and McGregor, 1998; Nielsen et al., 2001; Ohishi et al., 2002, 2004, 2006; Fujii et al., 2007). However, very few have been reported for influenza B (Osterhaus et al., 2000; Ohishi et al., 2002).

Uruguay has one of the largest colonies of both *Arctocephalus australis* (South American fur seal) and *Otaria byronia* (South American sea lion) in the world. These species inhabit the Lobos Island, located in the Atlantic Ocean 8.5 km from the coast (35°1′24″S, 54°52′54″W). The islands have a rocky surface that covers 41 ha.

Serum samples were collected in September 2004 from 37 South American fur seals (12 female and 25 male) estimated to be 10 mo old. The presence of antibodies against influenza A and B virus was tested using the hemagglutination inhibition assay (HAI) according to the WHO Manual on Animal Influenza Diagnosis and Surveillance (2002a). Briefly, all serum samples were incubated overnight in a 37 C waterbath with receptor-destroying enzyme (RDE) in a relationship 1:3, respectively, to remove nonspecific inhibitors and then heated at 56 C during 30 min to inactivate the RDE. After that, the serum samples were absorbed with turkey red blood cells to inactivate nonspecific agglutinins. The HAI was performed in microtiter plates using turkey erythrocytes at a concentration of 0.5% in phosphate-buffered saline. A sample was considered antibody positive when the HA titer was ≥80, as reported by Ohishi et al., 2004.

Downloaded From: https://complete.bioone.org/journals/Journal-of-Wildlife-Diseases on 28 Mar 2024
Terms of Use: https://complete.bioone.org/terms-of-use

TABLE 1. Hemagglutination-inhibition assay (HAI) of fur seal serum samples with influenza A and B virus antigens. The highest titers for each antigen tested appear in boldface.

Influenza virus antigens	Positive rate[a] with titer<80	Positive rate with titer=80	Positive rate with titer=160	Positive rate with titer=320	Positive rate with titer=640
IA/H1N1/NewCaledonia/20/99	16/37(43.3%)	10/37(27%)	10/37(27%)	**1/37(2.7%)**	0/37(0%)
IA/H3N2/ Panamá/2007/99	37/37(100%)	0/37(0%)	0/37(0%)	0/37(0%)	0/37(0%)
IB/Beijing/184/93	12/37(32.4%)	13/37(35.2%)	10/37(27%)	**2/37(5.4%)**	0/37(0%)
IB/Hong Kong/330/01	17/37(46%)	14/37(37.8%)	**6/37(16.2%)**	0/37(0%)	0/37(0%)
IB/Sichuan/379/99	13/37(35%)	4/37(11%)	8/37(22%)	10/37(27%)	**2/37(5%)**

[a] Number positive/number tested (positive percent).

We used the following influenza virus antigens: A/New Caledonia/20/99(H1N1), A/Panamá/2007/99(H3N2), B/Beijing/184/93-like viruses, B/Yamanashi/166/98, B/Hong Kong/330/01, and B/Sichuan/379/99 (WHO Influenza Reagent Kit, Center for Disease Control and Prevention, Atlanta, Georgia, USA, given by Departamento de Laboratorios de Salud Pública of Ministerio de Salud Pública).

Antibodies to influenza were detected in most seal-serum samples tested (Table 1). Thus, among 37 serum samples, only eight did not inhibit hemagglutination of any antigen, while five inhibited hemagglutination of one antigen assayed; and three, five, and 16 sera inhibited two, three, and four antigens, respectively. None of them reacted with all the antigens considered, but 17 reacted with all B antigens tested and 16 of them with H1N1 antigen.

Regarding influenza A virus, we detected antibodies reaching HAI titers of 320 only in one (3%) serum sample against H1 subtype. On the other hand, we found antibodies against three influenza B virus strains that have been previously detected in humans in Uruguay. The Beijing strain circulated in 1999 (WHO, 1999), the Sichuan strain in 2001 (WHO, 2001), and the Hong Kong strain in 2002 (WHO, 2002b). The highest HAI titers (640) were found against the Sichuan strain in 5% of the seals studied (Table 1), but the HAI titers of some sera against antigens of the other strains were also high (320 against Beijing in 5% of the samples and 160 against Hong Kong in 16% of the samples studied). The results were consistent with those proposed by Osterhaus et al. (2000) and Ohishi et al. (2002), which suggested that influenza B virus was not restricted to humans.

To our knowledge, this is the first report about influenza virus A and B circulation in this marine mammal species (*Arctocephalus australis*).

We thank Alberto Ponce de Leon and his team from Dírección Nacional de Recursos Acuáticos-Ministerio de Agricultura y Pesca, Montevideo, Uruguay for their cooperation in capturing the South American fur seals at Lobos Island. This research was funded by projects from Comisión Sectorial de Investigación Cientifica-Universidad de la República and Laboratorios Clausen SA, Montevideo, Uruguay and Laboratorios Clausen SA.

LITERATURE CITED

CALLAN, R. J., G. EARLY, H. KIDA, AND V. S. HINSHAW. 1995. The appearance of H3 influenza viruses in seals. Journal of General Virology 76: 199–203.

DANNER, G. R., AND M. W. MCGREGOR. 1998. Serologic evidence of influenza virus infection in a ringed seal (*Phoca hispida*) from Alaska. Marine Mammal Science 14: 380–384.

DE BOER, G. F., W. BACK, AND A. D. M. E. OSTERHAUS. 1990. An ELISA for the detection of antibodies against influenza A nucleoprotein in humans and various animal species. Archives of Virology 115: 47–61.

FUJII, K., C. KAKUMOTO, M. KOBAYASHI, S. SAITO, T. KARIYA, Y. WATANABE, Y. SAKODA, H. KIDA, AND M. SUZUKI. 2007. Serological evidence of influenza A infection in Kuril harbor seals (*Phoca vitulina stejneri*) of Hokkaido, Japan. Journal of Veterinary and Medical Sciences 69: 259–263.

Downloaded From: https://complete.bioone.org/journals/Journal-of-Wildlife-Diseases on 28 Mar 2024
Terms of Use: https://complete.bioone.org/terms-of-use

GERACI, J. R., D. J. ST. AUBIN, I. K. BARKER, R. G. WEBSTER, V. S. HINSHAW, W. J. BEAN, H. L. RUHNKE, J. H. PRESCOTT, G. EARLY, A. S. BAKER, S. MADOFF, AND R. T. SCHOOLEY. 1982. Mass mortality of harbor seals: Pneumonia associated with influenza A virus. Science 215: 1129–1131.

HINSHAW, V. S., G. M. AIR, A. J. GIBBS, L. GRAVES, B. PRESCOTT, AND D. KARUNAKARAN. 1982. Antigenic and genetic characterization of a novel hemagglutinin subtype of influenza A viruses from gulls. Journal of Virology 42: 865–872.

———, W. J. BEAN, R. G. WEBSTER, J. E. REHG, P. FIORELLI, G. EARLY, J. R. GERACI, AND D. J. ST. AUBIN. 1984. Are seals frequently infected with avian influenza viruses? Journal of Virology 51: 863–865.

———, J. R. GERACI, P. FIORELLI, G. EARLY, AND R. G. WEBSTER. 1986. Characterization of two influenza A viruses from a pilot whale. Journal of Virology 58: 655–656.

LANG, G., A. GAGNON, AND J. R. GERACI. 1981. Isolation of an influenza A virus from seals. Archives of Virology 68: 189–195.

LVOV, D. K., V. M. ZDANOV, A. A. SAZONOV, N. A. BRAUDE, E. A. VLADIMIRTCEVA, L. V. AGAFONOVA, E. I. SKLJANSKAJA, N. V. KAVERIN, V. I. REZNIK, T. V. PYSINA, A. M. OSEROVIC, A. A. BERZIN, I. A. MJASNIKOVA, R. Y. PODCERNJAEVA, S. M. KLIMENKO, V. P. ANDREJEV, AND M. A. YAKHNO. 1978. Comparison of influenza viruses isolated from man and from whales. Bulletin of the World Health Organization 56: 923–930.

MURPHY, B. R., AND R. G. WEBSTER. 1996. Orthomyxoviruses. *In* Fields virology, 3rd Edition, Vol. 1, B. N. Fields, D. M. Knipe, and P. M. Howley (eds.). Lippincott-Raven Publishers, Philadelphia, Pennsylvania, pp. 1397–1445.

NIELSEN, O., A. CLAVIJO, AND J. A. BOUGHEN. 2001. Serological evidence of influenza A infection in marine mammals of Arctic Canada. Journal of Wildlife Diseases 37: 820–825.

OHISHI, K., A. NINOMIYA, H. KIDA, C. PARK, T. MARUYAMA, T. ARAI, E. KATSUMATA, T. TOBAYAMA, A. BOLTUNOV, L. KHURASKIN, AND N. MIYAZAKI. 2002. Serological evidence of transmission of human influenza A and B viruses to Caspian seals (*Phoca caspica*). Microbiology and Immunology 46: 639–644.

———, N. KISHIDA, A. NINOMIYA, H. KIDA, Y. TAKADA, N. MIYAZAKI, A. BOLTUNOV, AND T. MARUYAMA. 2004. Antibodies to human-related H3 influenza A viruses in Baikal seals (*Phoca sibirica*) and ringed seals (*Phoca hispida*) in Russia. Microbiology and Immunology 48: 905–909.

———, T. MARUYAMA, A. NINOMIYA, H. KIDA, H. ZENITANI, T. BANDO, Y. FUJISE, K. NAKAMATSU, N. MIYAZAKI, AND A. BOLTUNOV. 2006. Serologic investigation of influenza A virus infection in cetaceans from the western north Pacific and the southern oceans. Marine Mammal Science 22: 214–221.

OSTERHAUS, A. D. M. E., G. F. RIMMELZWAAN, B. E. E. MARTINA, T. M. BESTEBROER, AND R. A. M. FOUCHIER. 2000. Influenza B virus in seals. Science 288: 1051–1053.

STEUEN, S., P. HAVE, A. D. M. E. OSTERHAUS, J. M. ARNEMO, AND A. MOUSGTAARD. 1994. Serological investigation of virus infections in harp seals (*Phoca groenlandica*) and hooded seals (*Cystophora cristata*). Veterinary Record 134: 502–503.

WEBSTER, R. G., V. S. HINSHAW, W. J. BEAN, K. L. VAN WYKE, J. R. GERACI, D. J. ST. AUBIN, AND G. PETURSSON. 1981. Characterization of an influenza A virus from seals. Virology 113: 712–724.

———, W. J. BEAN, O. T. GORMAN, T. M. CHAMBERS, AND Y. KAWAOKA. 1992. Evolution and ecology of influenza A viruses. Microbiology Reviews 56: 152–179.

WORLD HEALTH ORGANIZATION (WHO). 1999. Recommended composition of influenza virus vaccines for use in the 2000 influenza season. Weekly Epidemiological Record 39: 321–328.

———. 2001. Recommended composition of influenza virus vaccines for use in the 2002 influenza season. Weekly Epidemiological Record 40: 313.

———. 2002a. WHO manual on animal influenza diagnosis and surveillance. WHO Global Influenza Programme, pp. 1–98.

———. 2002b. Recommended composition of influenza virus vaccines for use in the 2003 influenza season. Weekly Epidemiological Record 41: 347.

Received for publication 27 June 2008.

Downloaded From: https://complete.bioone.org/journals/Journal-of-Wildlife-Diseases on 28 Mar 2024
Terms of Use: https://complete.bioone.org/terms-of-use