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Water-Borne Transmission of Influenza A Viruses?

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Summary. The isolation of influenza A viruses from unconcentrated lake water and from fecal samples on the shore of these lakes is reported for the first time. Influenza A viruses, representative of most of the major antigenic subtypes, co-circulate in ducks on the lakes.

Influenza A viruses from wild birds [1] have been implicated in the origin of the Asian [A/Singapore/1/57 (H2N2)] and Hong Kong [A/HK/1/68 (H3N2)] strains of human pandemic influenza [2-4]. A puzzling aspect of this postulate concerns the interspecies transmission of these influenza viruses since influenza is widely held to be an air-borne infection requiring close contact. Recent studies [5] in Alberta, Canada, have shown a prevalence of antigenically diverse influenza A viruses concurrently circulating in feral ducks. We report here additional studies of feral ducks in the same geographical area which were done last year to determine (i) the method

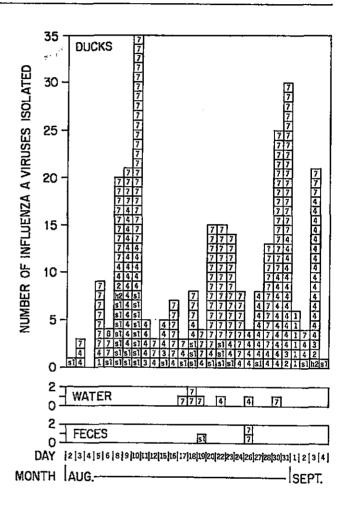
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of virus transmission, as well as (ii) the extent of antigenic diversity of these influenza viruses and (iii) evidence for continual circulation of viruses from year-to-year.

Our studies began after the breeding season and prior to migration, when the ducks were assembling in marshalling areas. From August 2 to September 4, 1977, cloacal samples were obtained from 2,046 waterbirds trapped on 16 lakes in Vermillion River County, Alberta, Canada. We visited each lake (average area 40 km²) approximately ten times during the month. Ducks were trapped 30-60 m from the shore in water 0.6-1.0 m deep; water was collected within 3 m of the traps, and feces were collected from the shore. Influenza A viruses were isolated from each of the three sources (fig. 1). Viruses were detected in the birds every day, with the exception of one, and on each of the 16 lakes. Viruses were most frequently isolated from the cloacae of healthy mallard ducks, the most common waterfowl

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Fig. 1. Influenza A viruses isolated from lake water, feces and ducks in Canada during August 2-September 4, 1977. 10- to 11-day-old embryonated chicken eggs were inoculated with samples from duck cloacae, feces, or lake water; after incubation at 35° for 72 h, viruses were detected by hemagglutination. The number in each block indicates an avian hemagglutinin subtype of the influenza virus isolated (e.g., 7 = Hav7); s1 and h2 refer to hemagglutinins related immunologically to the Hswl and H2 subtypes. ? indicates an unclassified hemagglutinin subtype. Influenza A viruses were characterized by hemagelutinin-inhibition [13] and neuraminidaseinhibition [14] tests with monospecific antisera [15] to the isolated antigens of reference strains [16-18]. The species of ducks from which viruses were isolated included mallard (Anas platyrhynchos), pintail (Anas acuta), green-winged teal (Anas carolinensis), blue-winged teal (Anas discors), gadwall (Anas stepera), canvasback (Aythya valisineria), redhead (Aythya americana), and widgeon (Mareca americana).



species of North America [6]; 26% of the juvenile (less than 1 year old) mallard ducks yielded virus. Unconcentrated water samples from six different lakes and droppings from the shores of three other lakes also yielded influenza viruses – marking the first time that influenza viruses have been isolated from these natural sources.

The surface antigens (hemagglutinin and neuraminidase) of these viruses comprised 18 different antigenic combinations and were related to 9 of the 15 reference hemagglutinin

subtypes (one hemagglutinin was not classified) and all 10 neuraminidase subtypes. The predominant antigenic combinations of viruses from the ducks were the only ones detected in water and feces and included Hsw1N1, Hav7Neq2, and Hav4Nav1. All of the major hemagglutinin and neuraminidase subtypes of human strains were represented among these duck isolates: Hsw1, which is related to the HO, H1, and Hsw1 subtypes [5]; H2, which is antigenically indistinguishable from the human H2 [3]; Hav7, which is related to H3

[2]; and both N1 and N2, which correspond to the neuraminidase subtypes of human strains. In addition, antigenic counterparts of these duck viruses (Hav1Nav2 [7], Hav7Nav1 [8], Hav4Neq2 [9], and Hav8Nav4 [10]) have been associated with disease outbreaks in domestic avian species in North America.

The continued circulation of many antigenically diverse influenza viruses in this feral bird population from 1976 [5] to 1977 supports the suggestion that wild ducks are a natural reservoir of most, if not all, influenza A viruses [2]. The findings of infectious virus in lake water and droppings and a concomitant high incidence of influenza viruses in the ducks suggest that water supplies, contaminated with feces from infected birds, may be a natural medium for the spread of virus infection among wild birds. Laboratory studies [11] have shown that (i) influenza viruses replicate in the cells lining the intestinal tract of ducks, (ii) high titers of infectious virus are excreted in the feces, and (iii) water inoculated with these feces retains infectious virus for 4 days at 22° and for over 30 days at 0°.

These findings add support to our contention that shared water supplies constitute an available source of virus for wild ducks, as well as other species. Domestic animals that drink contaminated water or share habitats with these ducks along the migration routes could become infected and could then transmit the virus to other animal species and, perhaps, to man. That interspecies transmission of influenza A viruses is a natural phenomenon has been established by the passage of viruses between pigs and man [12]. Hence, contamination of water supplies with feces of infected, yet healthy, feral ducks offers a mechanism for maintaining a wide variety of influenza viruses within the duck population and for introducing these viruses into other species.

Acknowledgments

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References

- 1 Easterday, B.C.: in Kilbourne, The influenza viruses and influenza, pp. 449-481 (Academic Press, New York 1975).
- 2 Webster, R. G. and Laver, W. G.: in Kilbourne, The influenza viruses and influenza, pp.269-314 (Academic Press, New York 1975).
- 3 Webster, R.G.; Laver, W.G., and Tumova, B.: Virology 67: 534-543 (1975).
- 4 Scholtissek, C.; Rohde, W.; von Hoyninger, V., and Rott, R.: Virology 87: 13-20 (1978).
- 5 Hinshaw, V.S.; Webster, R.G., and Turner, B.: J. gen. Virol. (in press).
- 6 Anderson, D.R. and Henny, C.J.: US Dept. of the Interior, Fish and Wildlife Service. Bureau of Sport Fisheries and Wildlife, No. 105 (1972).
- 7 Beard, C.W. and Helfer, D.H.: Avian Dis. 16: 1133-1136 (1972).
- 8 Yaseen, S.A.: Serotypes of neuraminidase of influenza A viruses isolated from birds in Canada; thesis, Univ. of Guelph, Ontario, Canada (1975).
- 9 Johnson, D.C. and Maxfield, B.G.: Avian Dis. 20: 422-424 (1976).
- 10 Lang, G.; Tumova, B., and Schild, G.C.: Bull. Wld Hlth Org. 47: 515-519 (1972).
- 11 Webster, R.G.; Yakhno, M.; Hinshaw, V.S.; Bean, W.J., and Murti, K.G.: Virology 84: 268– 278 (1978).
- 12 Hinshaw, V.S.; Bean, W.J.; Webster, R.G., and Easterday, B.C.: Virology 84: 51-62 (1978).
- 13 Advanced Laboratory Techniques for Influenza Diagnosis. US Dept. of Health, Education and Welfare. Immunology Series No. 6, pp. 25-62 (1975).
- 14 WHO Report: Bull. Wld Hlth Org. 48: 199-203 (1973).
- 15 Webster, R.G.; Isachenko, V.A., and Carter, M.: Bull. Wld Hlth Org. 51: 324-332 (1974).
- 16 WHO Report: Bull. Wld Hlth Org. 45: 119-124 (1971).
- 17 Webster, R.G.; Morita, M.; Pridgen, C., and Tumova, B.; J. gen. Virol. 32: 217-225 (1976).
- 18 Webster, R.G.; Tumova, B.; Hinshaw, V.S., and Lang, G.: Bull. Wld Hlth Org. 54: 555-560 (1976).