SEROLOGICAL SURVEY OF INFLUENZA IN AVIAN SPECIES USING ENZYME-LINKED IMMUNOSORBENT ASSAY (ELISA)

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An enzyme-linked immunosorbent assay (ELISA) was used to detect antibodies against influenza A virus in chickens and in domestic and wild ducks.

Purified influenza A viruses were disrupted with Triton X-100 and used as antigen for ELISA. Antibodies in chicken antisera to each subtype (HA1-13, NA1-9) of influenza A viruses were detected by ELISA with A/PR/8/34 (H1N1) or A/Aichi/2/68 (H3N2) as antigen. Antibodies in sera from chickens experimentally infected with A/duck/Hokkaido/5/77 (H3N2) were also detected by ELISA with a different subtype virus, A/PR/8/34 (H1N1), as antigen. Sera from 711 chickens were examined for antibodies against influenza A virus by ELISA with A/PR/8/34 virus as antigen. Of these, 19 (2.7%) birds were shown to be seropositive.

The cross-reactivity between 7.8S immunoglobulin from Peking ducks and from wild ducks of ten species was tested by gel-diffusion using rabbit antiserum against 7.8S immunoglobulin from Peking ducks. It was shown that the antigenicity of 7.8S immunoglobulin from Peking duck is common to those of mallard, wigeon, teal and shoveler, and partially common to those of eastern scap-duck, spot-bill duck, falcated-teal, pintail, pochard and tufted duck. Therefore, horseradish peroxidase conjugated rabbit IgG to Peking duck 7.8S immunoglobulin could be used in ELISA to test the other ten species of wild ducks.

Antibodies in the sera of ducks experimentally infected with A/duck/Hokkaido/8/80 (H3N8) and A/duck/Alberta/157/77 (H4N6) viruses were detected by ELISA with A/PR/8/34 (H1N1) of a different subtype virus as antigen. Sera from four ducks experimentally infected with A/duck/Hokkaido/5/77 (H3N2), which did not inhibit hemagglutination of the homologous virus, also showed antibody positive by ELISA with A/PR/8/34 (H1N1) virus as antigen. Sera from 214 ducks were examined for antibodies against influenza A virus by ELISA with A/PR/8/34 virus as antigen. Of these, 25 (11.7%) birds were shown to be seropositive in this ELISA system. This is the first seroepidemiological evidence indicating a high rate of infection of wild ducks with influenza A virus.

An attempt was made to detect antibodies in chicken antisera against influenza A virus and in sera of chickens and ducks experimentally infected with influenza A virus by ELISA of the Biotin-Avidin System using biotin-labeled A/PR/8/34 (H1N1) virus antigen. By this assay, antibodies in antisera and sera of chickens experimentally infected were detected at low titers, but they were not detected in the sera of the experimentally infected ducks.