The Burden of Avian Influenza Viruses in Community Ponds in California

Zin Htway, Doctor of Philosophy, Public Health

Dr. Richard Jimenez, Chair; Dr. LaToy Johnson, Member; Dr. Rodney Bowden, URR


Abstract
This research was a baseline study of the proportion of influenza A virus (IAV) in urban and rural communities in California. The population was artificial recirculating water ponds in the geographic locations of rural and urban communities. Surface water samples were collected from artificial recirculating ponds in California.

Research Questions
(R1): Is there a difference in the burden of influenza A virus in rural ponds compared to urban ponds?
(R2): Is there an association between geographic location, altitude, estimated surface area and observed presence of waterfowl, and the proportion of influenza A virus in rural ponds compared to urban ponds?
(R3): Is there an association between water pH, salinity, or water temperature and influenza A virus infectivity in rural and urban water ponds?
(R4): Are HSN1 influenza A viral subtypes detected in the sample of rural and urban ponds novel gene sequences or have the gene sequences been previously identified elsewhere?

Relevant Literature
Aquatic virus reservoirs is a human-animal interface. Franklin et al. (2011) suggested aquatic virus reservoirs give rise to indirect transmission, which would alter the transmission dynamics, beyond just direct interactions between infectious and susceptible individuals. The literature suggests novel influenza virus reservoirs originate in the Southern China region where migratory waterfowl become infected (Webster et al., 1992).

Procedures
A representative sampling from each of the 21 counties (rural areas), and 37 counties (metropolitan) in California were attempted. Surface water samples were collected from artificial recirculating ponds. IAV (+) was verified by real-time RT-PCR, MDCK cells for virus infectivity, nucleotide sequencing of the RNA genome, and phylogenetic analysis of IAV HSN1 strains. pH, salinity, and temperature of water samples were analyzed. Pond surface areas were calculated using a laser rangefinder, GARMIN® GPS, or Google Earth.

Limitations
The findings are limited to geographical locations along the migratory flyways of the waterfowl within the boundaries of California. Severe drought conditions limited available water ponds for sampling. Summer 2014 wildfires in N. California made several water locations unsafe for travel and field research. Unsafe smoke and fire across regions are likely to have diverted migratory waterfowl migration towards safer flight paths and stopovers.

Data Analysis
Included proportions, bivariate and multivariate logistic regression analysis. The IVs included: geographic community location (rural or urban), GPS location (latitude and longitude), altitude, approximate water pond surface area. The DVs included: IAV detection, IAV infectivity endpoint titer, IAV (HSN1) characterization. Variables evaluated as possible mediators and moderators included: water pH at collection, water salinity at collection, water temperature at collection, and presence or absence of waterfowl (Anseriformes) and shorebirds (Charadriiformes) at collection.

Findings
Showed an association in the burden of influenza A virus to geographical location. The data favored a greater burden of IAV in urban ponds compared to rural ponds. The data analysis—MDCK plaque assays, real time RT-PCR, and nucleotide sequencing—supported the proposal by Franklin et al. (2011) that aquatic systems may serve as reservoirs and sources of infection for both wild birds and mammals. Thus, the aquatic habitats sampled for this study were potential sites of the human-animal interface of IAV. Interestingly, the analysis of the data did not fully support the well-accepted theory of the influenza virus human-animal interface by Webster et al. (1992).

Social Change Implications
Impact of this study extends to the international level, the national and state level, the population level, and at the individual level. The results of this study show artificial water ponds in communities can be sources of IAV. Artificial water ponds in communities can be used for IAV surveillance and monitoring, expand and improve upon the IAV strain library for vaccine development, and may bring greater awareness to the individual, and thus, leading to improved vaccination rates.