Understanding Avian Influenza Virus: Infection, Pathogenicity and Prevalence

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Abstract

Influenza is a highly contagious respiratory disease, caused by influenza viruses that result in significant morbidity and mortality in humans and animals worldwide. Wild waterfowl are considered natural reservoirs for type A avian influenza viruses (AIV) and the source of influenza viruses that affect humans and species of high economic importance (poultry, swine and equine). During our DHS Summer Internship at the FAZD Center, Texas A&M University, we investigated the presence of human (α 2-6) and avian (α 2-3) sialic acid (SA) residues on the respiratory tract of several species (domestic turkey, wild turkey, chicken, quail, and pig) that could act as intermediate species for the adaptation of wild-bird influenza viruses to humans. Our results indicate the presence of both human and avian specific receptors in the examined species, suggesting that they may play an important role in interspecies transmission (Project 1).

We also examined the pathogenicity of AIV-mutants (altered at the neuraminidase and hemagglutinin genes). We measured the host immune response and identified lesions in the hosts' lungs and trachea. As preliminary results, two of the mutant viruses showed similar pathogenicity to that of the wild type virus while in the other mutant viruses the pathogenicity was reduced (Project 2).

In addition, we participated in the "Influenza A Infections in Wild Birds and GIS Decision Support System" surveillance program sponsored by the CSREES Avian influenza Coordinated Agricultural Program (AICAP) at Texas A & M University. The strategies that we learned are currently being used to screen resident and migratory wild birds in Puerto Rico to determine their role as potential carriers or intermediate hosts for the virus. Preventing the introduction and adaptation of wild-bird origin AIV to non-commercial and commercial poultry would minimize the impact on public health and poultry industry (Project 3).

Introduction

In addition to causing annual epidemics, type A influenza viruses have periodically caused serious pandemics that spread rapidly on a global scale. Wild waterfowl are considered natural reservoirs for type A influenza viruses and thought to be the source of all influenza viruses (Perez, et al., 2003; Wan and Perez, 2008). Occasionally, avian influenza viruses (AIV) are transmitted to other hosts such as pigs, horses, and humans. In 1997, H5N1 (by pathogenic) influenza viruses were transmitted directly from chickens to humans without an intermediate host (Perez, et al., 2003; Wan and Perez, 2008). Influenza virus infections are mediated by specific interactions between the viral glycoprotein hemagglutinin (HA) and cell oligosaccharides, which contain a terminal residue of a nine-carbon sugar named sialic acid (SA). Most AIV isolated from avian species, bind preferentially to the N-acetylgalactosamine (Mannose; a2-3) linkages. In contrast, influenza viruses isolated from human and swine are known to bind receptors with N-acetylgalactosamine (a2-6) linkages. Receptor specificity has been postulated as to provide the most effective host barrier for crossed-influenza virus infections. Our hypothesis was that human (a2-6) sialic acid (SA) residues would be present in different wild species.

The objectives were: Project 1: to identify human (α 2-6) and avian (α 2-3) sialic acid (SA) residues on the respiratory tract (trachea) of several species with the potential to act as virus reservoirs; Project 2: to study the influence of AIV-mutants in the pathogenesis of the virus by means of the host immunological response; and Project 3: to learn techniques to assist in the development of an early detection system for AIV in Puerto Rico, in order to prevent the introduction and adaptation of the virus to other hosts, including humans.

As our future goals, we plan to establish a AIV Surveillance Program in Puerto Rico. We also plan to coordinate research efforts, with local (PR) and National Agencies including, Interagency National Early Detection System for Highly Pathogenic AI Virus in Wild Birds in the United States, NIH Centers for Excellence on Influenza virus Research and CSREES AICAP among others.

Project 1

Tracheas collected from different animal species

White Leghorn
Rio Grande Turkey
Bobwhite quail
Jungle Fowl
White Leghorn
Feral Pig

Sialic acid (SA) residues were identified using biotin-conjugate Lectin-based staining. Lectines identify SA residues specific for human (α2-6): SNA and avian (α2-3): MAA influenza virus receptors. Presence and location of tubules, in respiratory ciliated cells, was confirmed by immuno-staining.

Project 2

1 day-old SPF White Leghorn

9-day-old chicken embryo

Egg Inoculation

Allantoic fluid collection

Sample collection

AV FluDetect

Hemagglutination Test

Hemagglutination Inhibition Test

Project 3

Chicken trachea sections stained with DAB. A: Sialool Lectin (α2-6); B: MAA Lectin (α2-3); C: Negative Control. D: Tubulin: identification of ciliated epithelial cells. E: Negative Control.

Literature cited

1. Perez, D.R., R.J. Webby, E., Hoffmann, R.G., Webster. 2003. Land-based birds as potential carriers or intermediate hosts for the virus. Preventing the introduction and adaptation of wild-bird origin AIV to non-commercial and commercial poultry would minimize the impact on public health and poultry industry (Project 3).

Conclusions

• Our results indicated the presence of both human and avian associated receptors in the white leghorn chicken, bobwhite quail, pig, and Rio Grande turkey. These findings suggested that these hosts may play an important role in AIV inter-species transmission (Project 1).

• Two of the mutant viruses showed similar pathogenicity to that of the wild type virus while in the other mutant viruses the pathogenicity was reduced (Project 2).

• As a result of the DHS Summer Internship, we gained experience on procedures required to carry out AIV surveillance in wild birds which we are currently using to screen resident and migratory birds in Puerto Rico to evaluate those as potential carriers and potential intermediate hosts for the virus (Project 3).

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