RESEARCH ABSTRACT

SWINE HEALTH

Title: **Comparison of Serological Assays in Diagnosing Differing Serotypes of Swine Influenza Virus (SIV) - NPB #03-162**

Investigator: Eileen Thacker, DVM, PhD, DACVM

Institution: Iowa State University

Date Received: August 29, 2005

Abstract:
Swine influenza virus (SIV) is an economically important pathogen of swine. Recently, new subtypes of the virus have emerged among US swine herds. In addition, apparent changes in the genetic structure of this virus have resulted in strains that often differ antigenically within subtypes. As a result of these changes in the virus, our ability to diagnose SIV infection and thus determine appropriate vaccine strategies has become increasingly difficult in the field. The study reported here compares the ability of a subtype-specific commercial enzyme-linked immunosorbant assay (ELISA), hemagglutination inhibition (HI), and the serum neutralization (SN) assays to detect antibodies elicited by different isolates within multiple subtypes of SIV. Pigs were infected with genetically different isolates of the 3 major circulating subtypes within swine population (H1N1, H1N2, and H3N2). When all pigs within a group collectively reached HI reciprocal titers ≥160 against the homologous virus for that group, serum was collected for use in the study. The serum was assayed and statistical analyses performed to compare the ability of these three different antibody assays to detect antibodies that were produced. Differences were found between these assays in the cross-reactivity among isolates and the ability of the each assay to accurately detect antibodies. These differences will provide important information to diagnostic laboratories, veterinarians and swine producers as they attempt to evaluate serological results based on antibodies produced following infection with SIV.

SIV infection can be diagnosed using methods that detect the virus including: virus isolation, antigen detection by immunoassay, and molecular-based assays such as RT-PCR. Diagnosis of infection by virus isolation can be very difficult as the virus is shed for a very short period of time following infection, often for only 3 to 5 days making timing of sample collection critical (9). Diagnosis of SIV infection can also be determined by measuring antibody production using; the hemagglutination inhibition (HI), ELISA, or serum neutralization (SN) assays (14). Detection of serum antibodies is the most common method used to diagnose infection and to determine the subtype of virus inducing the immune response. Currently, the HI test is the assay most commonly used to detect SIV antibodies, and is based on the ability of the hemagglutinin (HA) protein present on the viral surface to agglutinate red blood cells (RBC). This test has a moderate level of sensitivity depending on whether the test antigen used in the assay is recognized by the antibodies (3). Typically, the diagnosis of an SIV infection by the HI assay requires a four-fold increase in titer in paired acute and convalescent serum samples. Moreover, the HI assay is subject to nonspecific serum inhibitors, and frequent antigenic drift may lead to false negative results (3). A commercial ELISA (IDEXX HerdChek SIV) has been licensed for both the H1N1 (6, 7); and the H3N2 subtypes. However their ability to differentiate antibodies elicited against different isolates has not been well documented. The SN assay, which measures antibodies that are capable of neutralizing the virus, is labor intensive, virus specific, and the correlation to antibodies detected by the other assays is largely unknown (1, 13). Production of neutralizing antibodies is important in clearing the virus from the host as well as preventing infection (12, 18) and therefore a more complete understanding of the relationship between these three assays is of interest.

The central hypothesis of the proposed research is that antibodies elicited against genetically distinct isolates within the same or different subtypes of SIV may react differently in various serological assays, making the diagnosis of SIV infection difficult to interpret for producers and practitioners. In addition, correlation between influenza antibody assays has not been well defined. The reduced ability to accurately detect and classify the specific virus circulating within a herd makes diagnostic interpretation and vaccine selection difficult under field conditions. The study reported here is investigating the level of cross-reactivity between viruses of the same subtype using the three previously described antibody detection assays. These results will enable us to improve diagnostic accuracy as well as enable the industry to determine if new vaccines need to be developed for the control of individual viral isolates