Scientists identify human monoclonal antibodies effective against bird and seasonal flu viruses

February 22 2009

Researchers at the Dana-Farber Cancer Institute, Burnham Institute for Medical Research and the Centers for Disease Control and Prevention have reported the identification of human monoclonal antibodies (mAb) that neutralize an unprecedented range of influenza A viruses, including avian influenza A (H5N1) virus, previous pandemic influenza viruses, and some seasonal influenza viruses.

These antibodies have the potential for use in combination with other treatments to prevent or treat certain types of avian and seasonal flu. The study was published online on February 22 in *Nature Structural and Molecular Biology*.

The antibodies identified by the team of scientists neutralize a broad range of influenza A subtypes because they bind to the highly conserved stem region of H5 type hemaglutinin (HA). Binding to the stem prevents a conformational change in the protein that is necessary for viral entry into the host cell, thereby preventing further infection of host cells and the rise of escape mutants.

"The head portion of hemaglutinin is highly mutable, leading to the rise of forms of the virus that can evade neutralizing antibodies," said Robert Liddington, Ph.D., professor and director, Infectious and Inflammatory Disease Center at Burnham and one of the investigators on the study. "However, the stem region of hemaglutinin is highly conserved because

it undergoes a dramatic conformational change to allow entry of viral RNA into the host cell. It's very difficult to get a mutation that doesn't destroy that function, which explains why we aren't seeing escape mutants and why these antibodies neutralize such a variety of strains of influenza."

While more costly to produce than existing influenza drugs, therapeutic antibodies can be readily manufactured and stockpiled. In the event of a pandemic, the antibodies could be used in combination with antiviral therapies to contain the outbreak until a vaccine became available. The production of a new influenza vaccine takes six to nine months using conventional methods.

"There are clear settings where human monoclonal antibodies can be used strategically for both the prevention and early treatment of influenza infection and disease," said Wayne A. Marasco, M.D., Ph.D., associate professor of medicine at Dana-Farber and Harvard Medical School. "At-risk individuals, such as first responders and medical personnel, exposed family members and coworkers and patients who cannot make antibodies because of pre-existing medical conditions or advanced age, could all benefit from this new type of therapy."

In the study, the team of scientists used a human antibody phage display library to identify 10 mAb that bind to the stem of H5 type HA, the influenza protein responsible for viral entry into the host cell. The scientists determined the X-ray crystal structure of the mAb bound to the H5N1 HA, which showed that the heavy chain of the mAb inserts into a highly conserved pocket in the HA stem, inhibiting the conformational change required for membrane fusion and viral entry into the cell.

The scientists further showed that an unprecedented number of different types of bird flu and seasonal influenza viruses were inhibited and the mAb protected mice that were exposed to H5N1 virus. "Our human

monoclonal antibody protected mice from the lethal H5N1 virus even when injected three days after infection. This is good news, but many antibodies can do this. What surprised us is that the same antibody protected mice from a lethal infection with a very different virus such as the H1N1 subtype that causes seasonal human infections; this is really remarkable," said Dr. Ruben Donis, chief of the Molecular Virology and Vaccines Branch at CDC.

Vaccines consisting of attenuated or killed virus do not typically stimulate antibodies against the stem, perhaps because it is less accessible than the head region. In this study, the scientists used recombinant purified protein, not virus, so the antigenic part of the virus recognized by the antibodies was fully exposed.

Seasonal influenza occurs each year, causing mild to severe illness. Worldwide, more than 250,000 deaths from seasonal influenza occur annually. The best protection from seasonal influenza is yearly vaccination.

Influenza pandemics are worldwide outbreaks of disease that occur when a new influenza virus emerges for which people have little or no immunity. The disease spreads easily person-to-person, causes serious illness, and can spread across the country and around the world in a very short time. Health professionals are concerned that the continued spread of a highly pathogenic avian influenza A (H5N1) virus across eastern Asia and other countries represents a significant threat to human health. While vaccines can control influenza, they are not always effective because the vaccine must be updated each year. Vaccines against H5N1 in development have shown promise, but none has been reported to elicit a broad response in humans that would cover a broad range of different H5N1 virus strains. Antiviral medications, including the neuraminidase inhibitor oseltamivir (Tamiflu ®), is the primary treatment method, but has limited effectiveness if administered more than 24-48 hours after

symptom onset.

More information: J Sui et al. Structural and functional bases for broad-spectrum neutralization of avian and human influenza A viruses. *Nature Structural & Molecular Biology* DOI: 10.1038/nsmb.1566 (2009).

Source: Burnham Institute