Protein 'tubules' free avian flu virus from immune recognition

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A protein found in the virulent avian influenza virus strain called H5N1 forms tiny tubules in which it "hides" the pieces of double-stranded RNA formed during viral infection, which otherwise would prompt an antiviral immune response from infected cells, said Baylor College of Medicine researchers in an online report in the journal *Nature*.

Two domains or portions of the protein NS1 combine to form tiny tubules where double-stranded RNA is hidden from the immune system, said Dr. B. V. Venkataram Prasad, professor of biochemistry and molecular biology, molecular virology and microbiology at BCM and his student, Dr. Zachary A. Bornholdt (now of the Scripps Research Institute in La Jolla, California).

"Once we confirm the importance of this structural information, we should be able to design drugs to block this action," said Prasad. "There are other things the protein could do to interfere with different immune mechanisms. We don't know if this is the only mechanism or if there are others that also come into play during influenza virus infection."

The two researchers had already recognized the importance of the protein NS1 in the virulence of influenza viruses and particularly, H5N1, a form of avian flu associated with more than half the deaths in a 2004 "bird flu" outbreak that resulted in 50 human cases and 36 deaths in Vietnam, China and Thailand. In all but one case, experts ruled out human-to-human spread of the virus.
In a previous report, Prasad and Bornholdt described the structure of an

area of the protein called the effector domain. In this report, a series of elegant experiments designed and carried out over eight months by Bornholdt allowed the two scientists to "crystallize" the entire protein.

By doing this, they were able to determine its structure using a technique called X-ray crystallography. This technique enables scientists to determine the three-dimensional structure of proteins and other bio-molecules by scattering X-rays through a crystal of the molecule. They substantiated their structure with cryo-electron microscopy, which makes images of tiny frozen structures using an extremely powerful electron microscope.

That structure revealed a previously unsuspected idiosyncrasy of NS1 in H5N1 that could explain the virus' virulence. In most cases, when an infected cell is exposed to a virus, double-stranded RNA molecules are formed triggering a potent anti-viral response that involves production of interferon.

However, the two domains of NS1 in this H5N1 interact to form tiny tubules. The double-stranded RNA is hidden or sequestered in these structures. The cell never sees a significant length of the RNA and does not marshal its immune forces to the fight the virus. Prasad and Bornholdt believe also that cellular factor binding sites found on the surface of the tubules also play a role in fooling the immune system.

"This is only one structure," said Prasad. "We need to see if this holds up with other NS1 structures from other influenza viruses."

Bornholdt's technique for crystallizing the protein will prove valuable in pursuing this work, said Prasad.

"Is this a common mechanism for eluding the immune system?" he said. He said hopes to build a library to NS1 structures to facilitate future

studies designed to fight influenza worldwide.

While H5N1 is not usually transmitted from human-to-human at this point, a small change in its genetic structure – perhaps an exchange of genes with a more easily transmitted flu virus – could change that, he said. Developing drugs to fight the virus could prove life-saving in a pandemic.

Source: Baylor College of Medicine