Peering inside the skull of a mouse to solve meningitis mystery

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NYU Langone Medical Center scientists and their collaborators at the Scripps Research Institute in La Jolla, Calif., have discovered an unexpected cause for the fatal seizures seen in mice with viral meningitis, an infection of the central nervous system, according to a study published in the journal Nature. The finding may lead to a new way of thinking about how the human immune system responds to viral diseases.

The NYU researchers, Michael L. Dustin, Ph.D., the Irene Diamond Professor of Immunology and Professor of Pathology at NYU School of Medicine, and Jiyun V. Kim, Ph.D., a scientist in Dr. Dustin's laboratory, employed intravital two-photon microscopy to peer inside the skulls of infected mice. This breakthrough technology allows scientists to take moving pictures of immune cells in action. The cells are tagged with a protein that glows fluorescent green when activated by infrared light, which is able to penetrate living tissue without damaging it.

Drs. Dustin and Kim collaborated with Dorian McGavern, Ph.D., Associate Professor of Immunology and Silvia Kang, Ph.D., at Scripps Research Institute, who provided virology expertise and performed many critical experiments that supported the unexpected findings of the study.

A Disease Driven by the Immune System

The scientists used lymphocytic choriomeningitis virus (LCMV), which

is relatively harmless in humans with a healthy immune system. Other viruses that cause meningitis usually are associated with mild symptoms. By contrast, bacterial meningitis is a much more contagious and serious disease, particularly in young children. If not treated promptly with antibiotics, it may lead to hearing loss, brain damage, and even death.

Mice infected with LCMV suffer fatal seizures. It was known that these seizures are not caused by the virus itself, but by the immune system's response to the infection. Something sets off a chain of events that begins with leakage of fluid from blood vessels into the meninges, the protective covering of the brain and spinal cord, followed by swelling, which in turn leads to seizures. "T-cells, which are designed to attack the virus, were thought to be the bad guys, but no one understood the exact cellular dynamics involved in infection-induced seizures," explains Dr. Kim, who did the intravital two-photon microscopic imaging in the study, which will be published in the January 8, 2009, issue of *Nature*. It appeared online last month.

This sort of overreaction by immune cells, called immunopathology, is a factor in numerous conditions in humans, ranging from allergies and autoimmune diseases to stroke and viral infections.

High-Tech Imaging Shows Cells in Action

As the NYU researchers watched the behavior of the T-cells, they noticed something strange. Rather than attacking cells infected with the virus, the T-cells wandered around, apparently unable to recognize their targets. "Up to a point, the T-cells did everything they should do," Dr. Dustin explains. "They made copies of themselves and migrated to where the virus was, but when they got there, they couldn't do the right thing. At least they didn't do what we expected them to, which was to stick tightly to the infected cells."

Intravital two-photon microscopy employs an oscillating infrared laser yielding high-resolution moving pictures. Immune cells appear as bright green lights in the tissue covering the brain of a living mouse. Using surgical methods perfected by at NYU's Skirball Institute for Biomolecular Medicine by Wen Biao Gan, Ph.D., the microscopy produces time-lapse "movies," capturing activity that is not evident in still images made from slices of tissue viewed on a microscope slide.

"A series of frozen images gave the misleading impression that the T-cells were engaging with the infected cells, but intravital microscopy clearly showed that the immune cells appeared to overrun the infected cells", notes Dr. Dustin.

Source: NYU Langone Medical Center