Drug Delivery Innovation: Breaking Through the Blood Brain Barrier

Warfighters may be better protected from neurotoxic exposure to chemical and biological threats through a more sophisticated system of transporting countermeasures through the human blood-brain barrier (BBB).

A team of investigators from the University of Wisconsin including the principal investigator Dr. Eric Shusta, and Dr. Brian Pate, program manager from the Defense Threat Reduction Agency’s Chemical and Biological Technologies Department (DTRA CB), may soon discover a way to break through the BBB. Their efforts utilize human pluripotent stem cells as a means to identify new antibodies and their cognate human BBB transporters, such as insulin or transferrin receptor systems, capable of delivering efficient and specific CB countermeasures to their desired neurological targets.

The BBB is an anatomical feature consisting of tightly packed endothelial cells that form the walls of brain capillaries (vasculature) whose primary function is to prevent molecules from getting into the brain. This vascular structure is a critically important feature as it contains specialized transport pathways that allow the uptake of nutrients from the blood necessary for brain function.

The supporting cells that surround the BBB are unique in that they induce the barrier phenotype. Therapeutic access to the brain is restricted because the phenotype consists of tight junctions that serve to limit paracellular entry, and the lack (continued on page 2)
Understanding Q Fever Risks...

In the journal *Frontiers in Microbiology*, a review co-authored by Dr. Sara Ruiz and Dr. Daniel Wolfe highlights key aspects of Q Fever. The article discusses the risk the disease poses to military personnel, the recent advances in vaccines and a strategic approach to provide a next-generation vaccine against Q Fever for the warfighter.

Beyond the risk posed by Q Fever from a biodefense perspective, the disease also presents a concern for the military due to natural exposures. In an analysis of U.S. troops deployed to an area of Iraq with known Q Fever outbreaks, 7.2 percent were suspected to have been naturally exposed to *C. burnetii* during the deployment.

In a more comprehensive study, deployed warfighters admitted to hospitals with fever and other non-specific symptoms were monitored to determine which pathogens may have caused the illness. Approximately 10 percent of these individuals tested positive for Q Fever.

As chronic cases of Q Fever are difficult to diagnose and treat with antibiotics, vaccination strategies have been considered a viable alternative for at-risk populations. Q-Vax is a formalin-inactivated whole cell vaccine that is currently being utilized in Australia, demonstrating that effective, vaccine-mediated protection is possible. However, this vaccine cannot be administered to individuals that have previously been exposed to *C. burnetii*, which can represent a large percentage of deployed forces. Given this limitation, a next-generation vaccine against Q Fever that could be administered to the warfighter regardless of their exposure history is desired.

Relative to other biodefense pathogens, there are a fairly large number of Q Fever cases around the world each year. While unfortunate, these cases could provide key insights into the discovery of bacterial proteins that should be included in a safe and effective vaccine. Moving forward, development of these vaccines will continue to rely on animal models for testing and evaluation prior to clinical trials. However, clinical trials could be conducted in at-risk populations such as veterinarians and abattoir workers to determine how effective these vaccines are in reducing the disease burden associated with Q Fever.

The full article was published online in *Frontiers in Microbiology* on 16 December 2014, doi: 10.3389/fmicb.2014.00726.

Drug Delivery...

of fenestrae and low pinocytotic uptake impedes trafficking of small molecule drugs and large molecule therapeutics to and from the brain.

The BBB also contains efflux transporters that are capable of recognizing a broad range of substrates and serve to pump unwanted molecules back into the bloodstream.

The development of successful non-invasive medical countermeasures to chemical and biological threats that affect the central nervous system has been dependent upon the ability to effectively deliver small molecule pharmaceutical, biologic, and nano medicines to brain tissue. Current antibody-based approaches have yielded limited brain uptake because the antibody targeting reagents have restricted or low BBB permeability.

Shusta’s approach leverages quantitative engineering principles, tools, and applications along with medical expertise to rapidly enable the translation of these stem cell based therapeutics. The proposed methodology utilizes a large non-immune library of human antibody fragments that is displayed on the surface of phage particles. This library will be mined against a human BBB model developed in the Shusta laboratory. Because the model is based on human induced pluripotent stem cells, it has advantages in that it is human-sourced, forms a uniquely impressive physical barrier, and is well suited for transcytosis-based screening; therefore, the resultant identified antibody-transporter combinations will have human relevance.

Results from this work will ultimately result in the identification of new antibodies that target human BBB transport systems and may serve as more efficient carriers of chemical and biological countermeasures that mitigate the effects of neurotoxic exposure.

POC: Dr. Daniel Wolfe, daniel.wolfe@dtra.mil

POC: Dr. Brian Pate, brian.pate@dtra.mil
The Defense Threat Reduction Agency’s (DTRA) Research and Development (J9) Directorate, Chemical and Biological (CB) Technologies Department, serves as the Joint Science and Technology Office for Chemical and Biological Defense. This publication highlights the organization’s accomplishments to protect warfighters and citizens through the innovative application of science and technology research.