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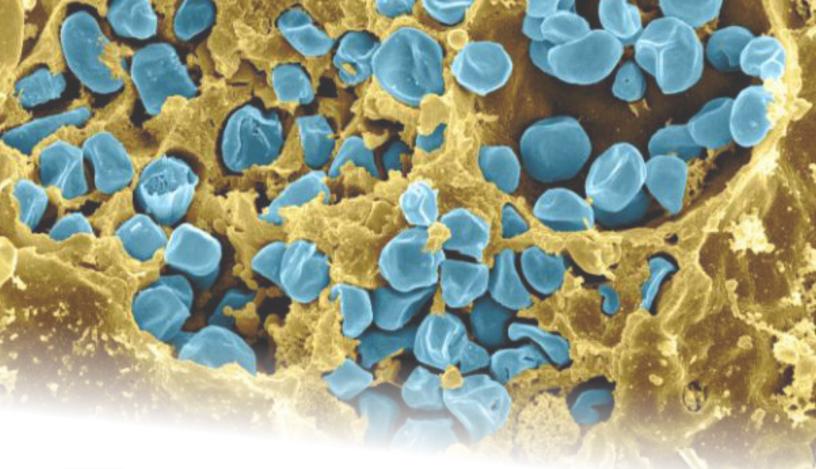
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Testing the Cold Water



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Lead DoD science and technology to anticipate, defend, and safeguard against chemical and biological threats for the warfighter and the nation.

# DEFENSE THREAT REDUCTION AGENCY

Research and Development Directorate Chemical and Biological Technologies Department Joint Science and Technology Office for the Chemical and Biological Defense Program

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Front cover: Soldiers walk off the firing line after executing a Joint Service Lightweight Integrated Suit Technology (JSLIST) stress fire at McGee range on Joint Base Elmendorf-Richardson, Alaska. The JSLIST is the product of a joint service effort to field a common chemical protective clothing ensemble including a lightweight protective garment, multi-purpose overboots, and gloves. (U.S. Air Force photo by Justin Connaher)

Inside cover: Scanning electron micrograph of a murine macrophage infected with *Francisella tularensis* strain LVS colorized in blue. (Photo by National Institute of Allergy and Infectious Diseases)

Back cover: A U.S. Marine boils snow for fresh drinking water near Tolga, Norway, while participating in trainthe-trainer cold weather training. Exercises such as Cold Response enhance familiarity with and proficiency operating in arctic conditions alongside allies and partners. (U.S. Marine Corps photo by Sgt. Robin Lewis)

# The bacteria that causes "rabbit fever" can survive in cold water, which poses a threat to the Joint Force.

esearchers recently discovered the bacterium *Francisella tularensis* that causes the disease tularemia or "rabbit fever"—which could be fatal whether occurring naturally or used as a bioweapon—can survive outside its small animal or insect hosts in cold water during winter months. This poses a threat to the Joint Force deployed in the Northern Hemisphere areas where the bacteria are prevalent.

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The Defense Threat Reduction Agency's (DTRA) Chemical and Biological Technologies Department in its role as the Joint Science and Technology Office (JSTO) for the Chemical and Biological Defense Program invested in research by Northern Arizona University (NAU) in collaboration with the Swedish Ministry of Defence to study the survivability and persistence of *F. tularensis* outside of its known hosts. The disease can result from inhaling as few as 10 *F. tularensis* organisms and is life threatening if untreated. Although tularemia is not passed from human to human, it is highly infectious by drinking water, getting bitten by deer flies or ticks, or eating or handling small animals—such as rabbits, hares, squirrels, and other rodents—that are infected with the bacteria.

# Tularemia can result from inhaling as few as 10 *F. tularensis* organisms and is life threatening if untreated.

Tularemia cases in humans normally occur during late summer and fall months in rural areas exclusively in the Northern Hemisphere. The bacteria typically attack lungs, lymph nodes, skin, and eyes. The resulting disease can be successfully treated with antibiotics, but it can be fatal if untreated, depending on how it was contracted:

- Inhaling the bacteria causes the most severe symptoms in the lungs.
- Being bitten by an insect or tick causes an ulcer that can spread to the lymph glands, producing a fever, chills, headache, and exhaustion.
- Ingesting the bacteria from eating infected food or liquids causes a sore throat with possible mouth ulcers, vomiting, and diarrhea.

The NAU team and their Swedish collaborators researched how *F. tularensis* could survive in the time between usual tularemia outbreaks. One hypothesis was that *F. tularensis* forms biofilms in aquatic environments on sand, rocks, or leaves that supply energy and organic matter to the food chain. The researchers tested this hypothesis using two strains mainly associated with human disease, a Type A strain found only in North America and a Type B strain found throughout the Northern Hemisphere, along with three control strains. They performed the tests in water at 39.2°F and 68°F that represented Scandinavian winter and summer conditions.

# The study produced three general observations on *F. tularensis* survivability in low-nutrient water:

- 1. *F. tularensis* strains do not form biofilms at either 68°F or 39.2°F.
- 2. Both Type A and Type B strains showed increased survival at 39.2°F compared to 68°F.
- 3. Type B but not Type A maintained virulence after 24 weeks at 39.2°F.



Francisella tularensis colonization on Cysteine Heart Agar after 72 hours. (Photo by CDC/Larry Stauffer, Oregon State Public Health Laboratory)

# Northern Hemisphere Type B retained its virulence for 24 weeks and the North America Type A was virulent for 12 to 14 weeks.

The test results show that neither of the Northern Hemisphere test strains formed biofilms, which indicates the two human disease strains do not appear to require biofilm formation to survive. Temperature seems to play a role in survival, with none of the five strains used in the test surviving after 12 to 14 weeks at 68°F. The results show that the virulence of the two human disease strains is aided by lower temperatures: the Northern Hemisphere Type B retained its virulence for 24 weeks, but the North America Type A was virulent for only 12 to 14 weeks.

Whether tularemia cases are the result of a natural outbreak or an intentional release, knowing that a pathogen like *F. tularensis* can survive in water in cold weather for several months can help ensure the Joint Force is properly protected, whether by administering a vaccine or preparing for situational awareness and mission planning in areas where tularemia outbreaks could occur year-round.

> Preventive medicine specialist collects a water sample during an Emergency Deployment Readiness Exercise at Fort Knox, Kentucky. Water samples are taken and processed in a laboratory to determine if the water has been exposed to contaminants. (U.S. Army Photo by Marshall R. Mason)





NAU recently published their findings that describe some conditions in which *F. tularensis* survives outside of a host system.

(Appl Environ Microbiol 87:e02713-20)

Click here to read the article

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# REDUCE. REUSE. REDEFINED

Breathable

Machine Washable

Durable

Flexible

Lightweight

New protective garments make for less wear and tear on the warfighter.

# Adsorbent & Detoxifying



resently, Joint Force protective equipment is effective at neutralizing

chemical warfare agents (CWA), but it is a burden to wear. The Defense Threat Reduction Agency's (DTRA) Chemical and Biological Technologies Department in its role as the Joint Science and Technology Office (JSTO) for the Chemical and Biological (CB) Defense Program partnered with the U.S. Army Combat Capabilities Development Command Soldier Center (DEVCOM SC) and industry performers NuMat Technologies and Guild Associates to create protective overgarments made of lighter materials with improved fit and flexibility for a quick self-detoxifying capability that provides the potential for reuse.

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Metal organic frameworks (MOFs) can be engineered to effectively decompose CWAs, interact with CWAs of a specific size, and be densely incorporated into textiles.

When assuming a Mission Oriented Protective Posture to enter a CB-threat environment, Joint Force servicemembers currently wear an array of protective equipment including a mask, an overgarment, gloves, and overboots to shield them from a full spectrum of CWAs. The overgarment is the Joint Service Lightweight Integrated Suit Technology, which provides effective protection, but it is hot, heavy, and limits breathability. Once the garment reaches sorption capability, it can no longer be used and must be destroyed.

DTRA-JSTO is investing in new self-detoxifying materials for the next generation of CB-protective overgarments that can neutralize CWAs within a day and can be washed and reused, which will increase its service life

The goal of this research is to modernize the materials in the CB-mitigating layer so that it will not only capture and retain chemicals but will also decontaminate CWAs. The researchers' strategy uses two simultaneous approaches. The first approach uses state-of-the-art lamination techniques to make the garment lighter and more flexible, enabling a better fit. The second approach incorporates new materials that destroy CWAs and could enhance or ultimately replace carbon, which enables the creation of a reusable garment. Zirconium hydroxide powders are prepared for microscopic examination of particle and pore size to determine powder quality. (Guild Associates photo)

This new class of materials comes in two forms: (1) zirconium hydroxide and (2) zirconium-based metal organic frameworks (MOFs). Both materials absorb CWAs and use environmental humidity to reactively detoxify CWAs, which could make the garment reusable and extend its service life.

A benefit of zirconium hydroxide powders is that they are inexpensive and easy to manufacture. Garment fabrication with zirconium hydroxide places a layer of carbon powder above a layer of zirconium hydroxide. CWAs contact the carbon powder first, where the agent is retained. Over time, from hours to a day, the CWA will leave the carbon and contact the zirconium hydroxide where it will be detoxified. The garment can then be laundered and reused.

MOFs are a class of materials composed of metal clusters coordinated to form porous structures. Zirconium-based MOFs are used in garment production and work with environmental water to directly neutralize CWAs. These MOFs can be engineered to effectively decompose CWAs, interact with CWAs of a specific size, and be densely incorporated into textiles. MOF manufacturing, however, is expensive.

Industry performers collaborate with DEVCOM SC to assess the capabilities of textile swatches. Methods include CWA permeation and detoxification quantification, breathability, abrasion testing, and robustness after laundering. Swatch testing is ongoing. When prototypes are built, DEVCOM SC will compare prototype garment performance against live CWAs (agent protection) and agent simulants (suit robustness, thermal performance, and overall protection). Self-detoxifying, protective garments will absorb and destroy contamination within 24 hours instead of just absorbing it.

OUTER LAYER

MOF LAYER

This collaboration of industry and Department of Defense laboratories will develop self-detoxifying, protective garments that will absorb and destroy contamination within 24 hours instead of just absorbing it like activated carbon. This greatly reduces the risk of contact exposure for the warfighter and enables garments to be reused, which will increase service life and reduce garment costs. Additionally, these new garments should exhibit reduced thermal burden, provide better fit, and maintain the inherent mobility of the Joint Force.

#### INNER LAYER

ADHESIVE LAYER

Fabricated laminate swatch contains four layers: Inner layer: soft, high-density knit; Adhesive: thermal glue; MOF layer: MOF-based textile; Outer layer: flexible, tough, flame-resistant material. (NuMat Technologies photo)



This fabrication floor allows roll-to-roll manufacture of laminated textiles that contain zirconium hydroxide for prototype protective garment design. (Guild Associates photo)



Within the Defense Threat Reduction Agency's Research and Development Directorate resides the Chemical and Biological Technologies Department performing the role of Joint Science and Technology Office for the Chemical and Biological Defense Program. This publication highlights the department's advancements in protecting the Joint Force and citizens from chemical and biological threats through the innovative application of science and technology.

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