

Inside this Issue:

CO Columns	2
Mosquito Research Aids Vaccine Development	4
Enhancing Warfighter Readiness in a Virtual Environment	5
NAMRU San Antonio Investigates Phage Therapy for Potential Applications to Treat Periodontal Infections	6
NAMRU-6 Studies Fever to Predict Pandemics in Developing Countries	7
Enhancing Dental Care for Sailors and Marines	8
Meeting of the 2016 National Neurotrauma Symposium (NNS)	9
Part V: The Mosquito Fighters	11
NSMRL Technical Director Retires	13
NAMRU-San Antonio Tells Their Navy Medicine Story in New Command Video	14
Meet NAMRU-SA's 2016 NREIP Summer Intern	15

Navy Medicine Researchers Develop a Therapeutic Cocktail of Environmental Phages to Overcome an Antibiotic-resistant Infection

By Doris Ryan, Public Affairs Officer, Naval Medical Research Center

SILVER SPRING, Md. – *Acinetobacter baumannii* is recognized as one of the most difficult antimicrobial-resistant gram-negative infections to treat. In a proof-of-concept study, a team from the Naval Medical Research Center (NMRC), in collaboration with the Walter Reed Army Institute of Research (WRAIR), reports success in combating an infection in a laboratory model using bacteriophage therapy. The results will be published in the October 2016 issue of the journal, *Antimicrobial Agents and Chemotherapy*.

The paper, *Personalized therapeutic cocktail of wild environmental phages rescues mice from A. baumannii wound infections*, is available online on the [American Society for Microbiology](#) website.

Combat-related injuries are at high risk for serious infections. Infections have occurred in up to 35 percent of service members who have had combat-related injuries during Operations Iraqi- and

(Continued on page 3)

Deputy Director of the Infectious Diseases Directorate (IDD) at the Naval Medical Research Center (NMRC), Cmdr. Michael Stockelman, shows Petri dishes containing disease pathogens used in Phage research in the microbiology lab at NMRC. (Photo by NMRC Public Affairs)



NMR&D News is a publication of the Naval Medical Research Center, 503 Robert Grant Avenue, Silver Spring, Maryland, 20910. NMR&D News is published monthly by the NMRC Public Affairs Office 301-319-9378.

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NMRC Commanding Officer's Message



It's August and many of us are headed to the annual Military Health System Research Symposium (MHSRS) in Florida! The symposium, sponsored by the Assistant Secretary of Defense for Health Affairs, is the only professional meeting focused specifically on the unique medical needs of the Warfighter.

MHSRS is the DoD's premier scientific meeting known for intense scientific collaborations and sharing of new scientific knowledge about military-unique research and development with a unique focus on optimizing care for military members in operational settings. Military and civilian scientists, along with medical care providers, will spend four action packed days exchanging information with DoD and academic scientists, international partners, and industry representatives on research and related health care initiatives in Combat Casualty Care, Military Operational Medicine, Clinical and Rehabilitative Medicine, and Military Infectious Disease.

The symposium's goal is to maximize research synergy among all of the branches of military service, while acknowledging the need for service-specific medical capabilities. This looks to be a banner year! The Navy Medicine Research & Development enterprise will be presenting over 60 scholarly presentations at both podium and poster sessions as

well as keynote addresses; and new this year, we will be covering many of these events so watch for updates on the [NMRC Facebook page](#).

Congratulations to each of the scientists and researchers who were accepted to present their work at the 2016 symposium. I am excited to know we're going to be sharing the great science we create throughout our enterprise and I am looking forward to hearing about the great collaborations you will bring back; certain to grow and expand our portfolio as we work on improving care and developing products for the warfighter.

**NMRC Commanding Officer sends,
Jacqueline D. Rychnovsky
CAPT, NC, USN**

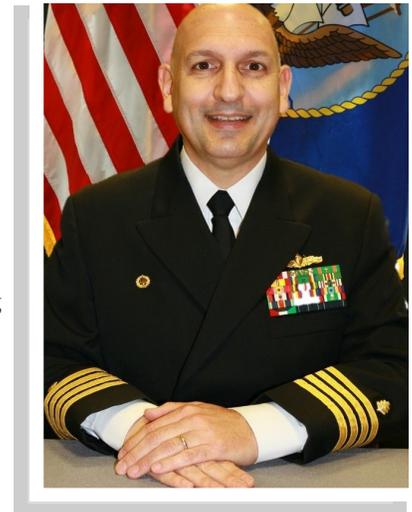
NAMRU-D Commanding Officer's Message

From an early age, our parents, teachers, and coaches ingrain in us the importance of fairness. Well, NAMRU Dayton is not looking to play fair. We are looking for an unfair fight. Before you call my Mom, please let me explain.

The United States has relied on Research and Development (R&D) to deliver the tools of asymmetrical warfare. That is, warfare where one side tips the odds dramatically in its own favor through a unique capability, or "offset". An unfair fight. The U.S. has benefited from a series of offsets. The awesome firepower of nuclear weapons defined the First Offset while the Second Offset appeared with the advent of precision-targeted conventional weapons. Unfortunately, the U.S. is at risk of losing its offset advantage. Precision weaponry is no longer the exclusive purview of the U.S.; and in this era of warfare involving non-state actors like ISIS, nuclear weapons are practically useless. DOD leaders have been looking for a Third Offset: the human being. Enhancing performance by integrating the human seamlessly with machines and other technology. Navy Medicine R&D is positioned perfectly to help answer the call.

At NAMRU Dayton, we are working with the Air Force 711th Human Performance Wing to develop these Third Offset technologies. Expanding the warfighter's ability to safely operate in toxic environments where our adversaries cannot, using The Kraken to develop methods to operate in the most austere motion environments, effective integration of humans into Unmanned Aerial Vehicles, flying computers like the F-35 and other machines all contribute to the Third Offset.

I am so very excited about the future. U.S. military forces may be getting smaller but new technologies from NAMRU-D and sister R&D labs will make each and every warfighter more efficient, more effective and more capable. Yes, we are looking for an unfair fight.



**NAMRU-D Commanding Officer Sends,
Rees L. Lee
CAPT, MC, USN**

Navy Medicine Researchers Develop a Therapeutic Cocktail of Environmental Phages to Overcome an Antibiotic-resistant Infection

(Continued from page 1)

Enduring Freedom. Those infections are often very hard to treat because of multidrug antibiotic-resistance in the bacteria present in the wound.

NMRC worked in collaboration with Navy Medicine's overseas laboratories to collect phages from environmental sources around the world. Using the collection of phages, referred to as a phage library, personalized phage cocktails can be made by selecting multiple individual phages from the phage library to create phage mixes customized to the needs of each patient being treated. NMRC also worked closely with WRAIR's Wound Infections Department to test the phage cocktails in wound infection models and demonstrate that personalized phage cocktails can treat infections.

"Bacteriophages, commonly known as phages, are viruses found in the environment, and are known for their activity against bacteria. This is why they have therapeutic potential, and may be able to treat bacterial infections even when

antibiotics fail," said Cmdr. Michael Stockelman, Deputy Director of the NMRC Infectious Diseases Directorate. "In this study we showed that a phage cocktail can be designed and used to target an infection caused by antibiotic-resistant bacteria in a wound infection experimental model."

Phages have an entirely different way of attacking bacteria compared to the mechanism of action for conventional antibiotics. Phages invade the bacterial cells, replicate in the cells, and destroy the cells when they rupture and release more phages into the body. Because phages multiply when they kill the disease-causing bacteria, the phage treatment actually gets stronger where it is needed most, at the infection site. In addition, phages do not affect bacteria that are not being targeted.

"Our bodies have many different kinds of bacteria that normally live on or in us, and they help our bodies work," said Stockelman. "When we take antibiotics, we impact the whole system, killing good

bacteria along with the bad bacteria. Phages will not damage that community of bacteria called the microbiome the way antibiotics do."

Stockelman explained that phage therapy could allow physicians to treat and manage wound infections in combat casualties who would otherwise need repeated surgeries or other extreme measures, including limb amputations, to control the infections. Successful phage therapy would greatly improve the quality of life for wounded warriors and make it more likely wound infections would not prevent a warfighter's ability to return to duty.

The next step for the team is to plan for clinical trials testing phage cocktails in human volunteers. Initial studies will confirm phage is safe in humans. For future clinical trials, researchers hope to work with treating physicians to eventually make personalized phage cocktails for patients with otherwise untreatable infections, to test the ability of phage therapy to overcome antibiotic resistance in a medical setting, outside the laboratory.

NMRC Celebrates 69th Birthday of Naval Medical Service Corps

By NMRC Public Affairs

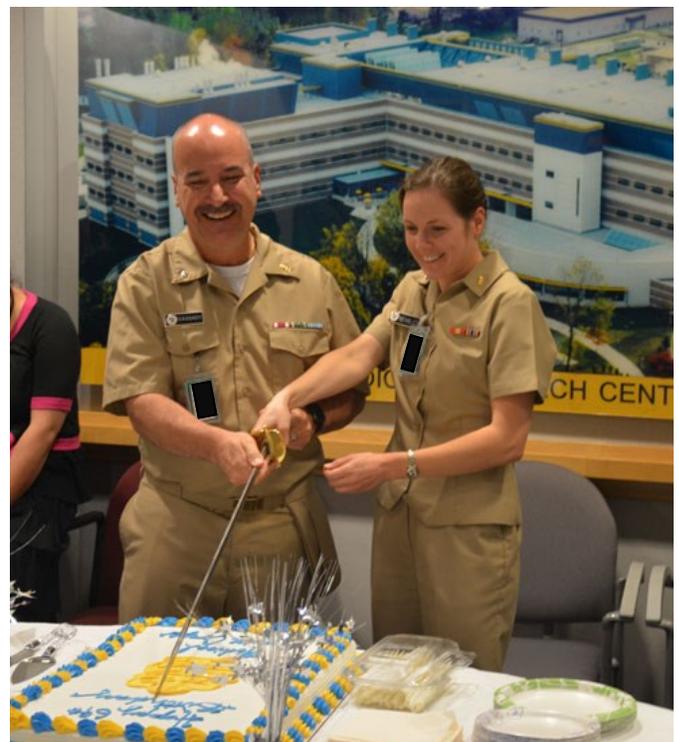
SILVER SPRING, Md.— August 4, 1947, President Harry S. Truman signed Public Law 80-338 creating the Medical Service Corps in the United States Navy.

The crew at the Naval Medical Research Center (NMRC) celebrated the 69th Birthday of the Naval Medical Service Corps (MSC) August 4 with a cake-cutting ceremony meant to observe 69 years of the Navy MSC's dedicated and unwavering service.

NMRC Commanding Officer, Capt. Jacqueline Rychnovsky, gave a brief address on the importance of the celebration followed by the cutting of the cake.

Longest serving MSC officer, Cmdr. Michael Cassidy, and shortest serving officer, Lt. Melissa Mehalick, were given the honors of the ceremonial cutting of the cake.

Cmdr. Michael Cassidy (left) and Lt. Melissa Mehalick (right) join in cutting the cake during a brief ceremony at the Naval Medical Research Center to celebrate the 69th Birthday of the Naval Medical Service Corps. (Photo by NMRC Public Affairs)



Mosquito Research Aids Vaccine Development

By Naval Medical Research Center Public Affairs

SILVER SPRING, Md. – Mosquitos are everywhere and can be hard to avoid. Through the research conducted at the Naval Medical Research Center (NMRC), mosquitos play a vital role in successful vaccine development. Mosquitos can spread many diseases including dengue, West Nile Virus, Zika virus and malaria. Entomologists like Lt. Cmdr. Roxanne Burrus, Ph.D., working for NMRC's Infectious Disease Directorate (IDD), use mosquitos for research to help develop new vaccines for malaria, dengue and Zika virus.

“Many mosquito-borne diseases do not appear to be circulating heavily in the U.S. at this time, although with climate change and global travel patterns of humans and shipments of goods, the potential for that occurring in the future is a concern, we are seeing this now with the concerns related to the Zika virus,” says Burrus.

Mosquito-borne diseases cause debilitating symptoms and could prevent military personnel from completing their mission. Vaccines can protect individuals from becoming infected or reduce the severity of infection if administered prior to being bitten.

According to Burrus, in the case of malaria, mosquitos are used in a laboratory

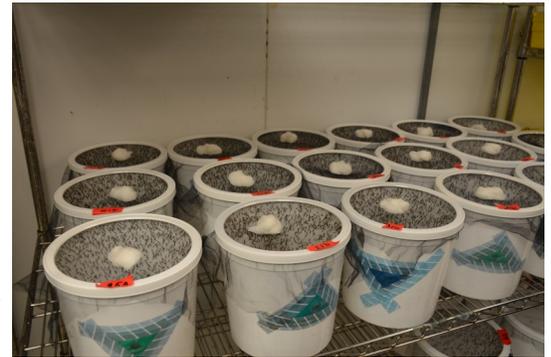
setting to grow batches of malaria sporozoites which are used in studies to develop and test potential vaccines.

The malaria parasite life cycle involves two hosts. During a mosquito blood meal, a malaria-infected female Anopheles mosquito inoculates sporozoites into the human host. Sporozoites infect liver cells and mature into schizonts, which rupture and release merozoites into the blood stream.

“NMRC coordinates closely with the overseas labs through the year and provides support to the labs by assisting them to focus on the most relevant infectious diseases in each overseas laboratory's region,” says Burrus.

“NMRC also sends researchers to visit overseas labs so that we can better understand the research project, interact with collaborating host-nation organizations, and develop plans of action to overcome any challenges that might be faced by an overseas lab,” said Burrus.

The overseas labs which are part of the Navy Medicine R&D enterprise are the U.S. Naval Medical Research Center – Asia in Singapore (NMRC-A) with a detachment in Phnom Penh; the U.S. Naval Medical Research Unit No. 3 in



Mosquitos bred for study in the Walter Reed Army Institute of Research (WRAIR) insectary in Silver Spring, Maryland, to help develop vaccines to fight malaria. (Photo by NMRC Public Affairs)

Cairo, Egypt; and the U.S. Naval Medical Research Unit No. 6 in Lima, Peru (NAMRU-6) with a satellite laboratory in Iquitos, Peru.

The Department of Defense (DoD) has made the development of vaccines to combat mosquito-borne illnesses a top priority, pointed out Burrus. The Military Infectious Disease Research Program (MIDRP) publishes a list of the most important military diseases which is frequently modified to reflect changes in assessed risk levels.

“Malaria and dengue top MIDRP's list of the infectious diseases that are of highest military importance,” says Burrus. “NMRC and its overseas laboratories are continuing efforts to develop vaccines for such illnesses. Research on Zika virus vaccines is in its initial stages.”

When it comes to controlling mosquito populations, Burrus says mosquitos become resistant to pesticides over time, so it is important to constantly develop and test new products for use during military, humanitarian and public-health emergency situations.

For service members and civilians alike, the best defense is to avoid mosquito bites. For service members, using the DoD Insect Repellent System provides the best protection from mosquito bites. It incorporates application of permethrin repellent on the uniform, application of DEET or picaridin repellent on exposed skin, and emphasizes properly worn uniforms and sleeping inside a permethrin-treated bed net.



Lt. Cmdr. Roxanne Burrus (left), entomologist at the Naval Medical Research Center (NMRC), and Dr. Lindsey Garver (right), senior research scientist for the entomology branch of the Walter Reed Army Institute of Research (WRAIR), working with mosquitos in the WRAIR insectary in Silver Spring, Maryland. (Photo by NMRC Public Affairs)

Enhancing Warfighter Readiness in a Virtual Environment

By Pinata Sessoms, Ph.D., research biomedical engineer, Naval Health Research Center

Innovation in military medicine is about saving lives on the battlefield, preventing or mitigating injuries in our warfighters, and ensuring we have a fit and ready force. Innovation is also vital to medical research, which ensures our military leaders and medical providers have the latest information, tools and techniques available to support the health and readiness of our service members.

In the Warfighter Performance Department at the Naval Health Research Center (NHRC), our Physical and Cognitive Operational Research Environment (PhyCORE) team is one group whose innovative research is continually enhancing the physical and cognitive health and readiness of our warfighters. The research we conduct spans a diverse portfolio of topics including:

- Rehabilitation methods for wounded warriors with amputations and mild traumatic brain injuries.
- Evaluation of personal protective equipment.
- Countermeasures for the impact of fatigue on cognitive performance.
- Assessing gear and equipment design variations on performance and survivability.
- Finding objective measures to determine injury level and fitness to return to duty.

One of the unique capabilities of the PhyCORE team is the tool we use to conduct cutting-edge research. Housed in NHRC's Warfighter Performance Lab is the Computer Assisted Rehabilitation Environment (CAREN), an immersive virtual reality system. CAREN is a very large, state-of-the-science piece of technology. When first installed in 2008, the CAREN was initially intended for rehabilitation research in our wounded warriors. Original features of the CAREN included:

- A 9-foot diameter platform, programmable to move in six degrees of freedom, independently or simultaneously.
- A treadmill centered in the platform with integrated force plates to measure ground reaction forces.
- A 180-degree wide, 9-foot tall curved screen surrounding the platform.
- Motion capture cameras integrated with the screen to track user movement.

While the CAREN's off-the-shelf capabilities were well-suited to its original intent of supporting clinical investigation and therapy for the wounded, ill and injured, the PhyCORE team saw the

potential for creating a tool to support additional avenues of research. This research would look beyond rehabilitating wounded warriors and begin studying novel approaches for optimizing the warfighters performance by keeping them healthy and fit.

Expanding the research capabilities of the CAREN required multiple system enhancements. Leveraging our combined expertise (physical therapy, biomechanics, software and hardware engineering, sleep physiology, aerospace experimental psychology, and neurophysiology, among others), the PhyCORE team has modified and improved the CAREN's original configuration to meet military medicine's emerging research needs. Our enhancements include:

(Continued on page 6)



A service member uses the CAREN, a high-tech tool originally designed for rehabilitation and clinical research, which has been enhanced by NHRC scientists to study injury prevention, test equipment and evaluate cognitive performance in a virtual environment. (Photo courtesy of NHRC Public Affairs)

NAMRU San Antonio Investigates Phage Therapy for Potential Applications to Treat Periodontal Infections

Story by Yoon Hwang, Ph.D., NAMRU San Antonio

SAN ANTONIO-- Mission readiness is critical to the success of the U.S. military, and the health of service members is of highest importance. Almost 20 percent of all emergency department visits at a deployed expeditionary medical support facility during Operation Enduring Freedom (OEF) were the result of dental disease.

In deployed military personnel, 12 percent of dental emergencies can be attributed to gingivitis or periodontitis. Furthermore, the emergence and increasing prevalence of bacterial strains that are resistant to available antibiotics poses a serious threat not only to the military, but to world health. The Centers for Disease Control and Prevention reported antibiotic resistance causes two million serious infections and 23,000 deaths each year; adding \$20 billion in excess direct health care costs, in addition to \$35 billion in lost productivity each year.

A critical need exists for the development of novel antimicrobials and alternative strategies for treating bacterial infections. Antimicrobial peptides (AMPs) and bacteriophages have shown promise as potential therapeutics. AMPs are short,

positively charged and amphiphilic peptides, that have been reported to provide potent, broad-spectrum activity against microbial infections and have been considered as potential therapeutic sources for future antibiotics. Bacteriophages

infect and replicate in their specific host bacterial cells, then release progeny phages to infect more bacteria nearby. The therapeutic use of bacteriophages to treat pathogenic bacterial infections is

(Continued on page 7)



Navy scientist, Yoon Hwang, Ph.D., in NAMRU-SA's Craniofacial Health and Restorative Medicine Directorate is checking the expression of visual markers for the development of genetically modified bacteriophages as a novel therapeutic strategy to treat periodontal infections. (Photo courtesy of NAMRU-SA Public Affairs)

Enhancing Warfighter Readiness in a Virtual Environment

(Continued from page 5)

- A programmable scent-delivery system, 3-D projection capability, improved sound system and a high-performance treadmill for destabilizing movements that increase the immersive experience.
- A custom driving cab built in-house that integrates with the system for adaptable, simulated driving scenarios, used as a tool to measure the effects of operational task performance and test fatigue-related countermeasures.
- Surface electromyography (sEMG) to measure muscle activation and fatigue.
- Mobile electroencephalography (EEG) systems to measure brain activity patterns of the user engaged in different tasks to assess real-time cognitive states.
- A laser-based marksmanship training and evaluation system that integrates with the treadmill and motion platform to create a tool ideal for testing cognitive effects of new protective gear and equipment loads on performance in

virtual battlespaces including villages, urban cities, oceans, rivers, fields and mountains.

Thanks to the collaborative efforts of our team, the CAREN is now a single system with multiple research capabilities used to investigate a diverse set of warfighter health and readiness needs in a highly controlled, operationally relevant, virtual environment.

In addition to enhancing the CAREN at NHRC, the PhyCORE team often partners with other Department of Defense (DoD) CAREN sites, and the global community, by sharing information and disseminating research findings. Our team members also run regular DoD CAREN operator meetings, help other sites troubleshoot and advance their systems, and implement new technologies at other sites so the DoD CAREN systems are cross-functional and capable of running multi-site studies.

Looking ahead, the PhyCORE team will continue to embrace the spirit of innovation to improve our research capabilities and provide novel solutions to the unique challenges facing the military health research system and our warfighters.

NAMRU-6 Studies Fever to Predict Pandemics in Developing Countries

Story by NAMRU-6 Public Affairs

LIMA, Peru – Navy Medicine researchers recently published a study reporting for the first time how fever affects mobility during an outbreak of a mosquito-borne pathogen like dengue. The study was published in the *Proceedings of the Royal Society B: Biological Sciences*.

The U.S. Naval Medical Research Unit No. 6 (NAMRU-6), in collaboration with several U.S. universities, studies the transmission dynamics of dengue virus in residents living in the Amazonian city of Iquitos, Peru. The primary importance of these findings is to support mathematical models of dengue virus transmission. Cohort studies of this stature and magnitude only exist in two other locations and have provided critical background information for vaccine deployment and vector control intervention trials.

“Human mobility is an important driver of (disease) transmission dynamics. Mobility increases potential contact in locations where vectors (mosquitoes) are present and enhances transmission and spread of the disease,” said Amy Morrison, program manager and science advisor at the lab. “When study volunteers are diagnosed with dengue we ask them to let us evaluate them clinically throughout their illness, to carry GPS units to track their movements, and to answer questions about symptoms and personal activities. We also collect

mosquitoes in their homes.”

The GPS data showed people with fevers spent more time in their homes, but visit more locations with a far less predictable routine than their U.S. counterparts; an observation the disease modelers suggest enhances disease transmission, according to Morrison.

The majority of dengue infections are mild or asymptomatic, a characteristic shared by the Zika virus. NAMRU-6 researchers are interested in determining the role of how this “majority” of infections are transmitted. Morrison went on to explain the research team compared the movement of people with and without fevers and was able to quantify the impact of fever on movement patterns, and how these observations would affect dengue transmission.

“Conventional wisdom tells us people with more symptoms have more virus in their blood and would be better at infecting mosquitoes. Our fever studies indicate people with fevers move less than we would expect from someone without symptoms,” said Morrison. “Our hypothesis is - even though asymptomatic and mild cases may be less efficient at infecting mosquitoes, asymptomatic people are spreading the virus more quickly to more locations and have higher numbers than symptomatic cases. Asymptomatic cases may account for far more of the disease transmission. Because current control measures often are reactive

to symptomatic cases, they do not effectively control the disease. This is a question now being studied by the NAMRU-6 research team.”

Morrison went on to say, this study is one of the first to generate this type of data in a developing country where dengue is endemic. The study suggests models that ignore differences in mobility due to fever could overestimate the potential for a disease to become pandemic.

NAMRU-6 Executive Officer, Cmdr. Guillermo Pimentel, explained, “This dengue study is another example of the contribution the NAMRU OCONUS (overseas) research laboratories make in generating products and new infectious disease knowledge for Global Health. Accurate disease forecasting and prediction models are crucial to understand and prevent epidemics of public health concerns.”

Dengue is a mosquito-borne viral disease transmitted by female mosquitoes of the species *Aedes aegypti*. There are 4 distinct, but closely related, serotypes of the dengue virus. NAMRU-6 researchers have been studying dengue in Iquitos since 1990. They have characterized the invasion of serotype 1 in 1990, serotype 2 (American genotype) in 1995, serotype 3 in 2001, serotype 4 in 2008, and serotype 2 (Asian American genotype) in 2016. Researchers also added the Zika virus in May 2016.

NAMRU San Antonio Investigates Phage Therapy

(Continued from page 6)

known as phage therapy and has many potential applications in medicine as well as dentistry.

A unique aim of our research within the Craniofacial Health and Restorative Medicine Directorate at Naval Medical Research Unit-San Antonio (NAMRU-SA) is to combine the strength of AMPs and phages together by engineering phages to express AMPs during their lytic cycle. To achieve maximal synergistic effect, a technique to genetically modify phages was developed by inserting an AMP-expressing genetic construct within the phage genome. By replicating within the

targeted bacterial cells and expressing AMPs in the infection site, this novel approach can achieve high local concentrations of both AMPs and lytic phages to target nearby bacteria – even with modest initial phage inoculation – and enhance the range of the original phage infection. Overall, phages and AMPs are less likely to induce bacterial resistance than conventional antibiotics and are considered promising alternatives.

With support from Naval Medical Research Center's Advanced Medical Development Program, ideal candidate AMPs were selected based on their antibacterial activities and safe

concentration against human gingival epithelial cells. Genetic engineering of the phage genome utilizing several recombinering methodologies is currently being conducted. Further testing and development of phage delivery systems are underway to identify new bactericidal treatments for periodontal and wound infections.

Development of genetically engineered phage expressing additional antimicrobial peptides would provide the foundation for rapid, selective and inexpensive treatment methods against periodontal pathogens and help sailors and marines maintain a high level of readiness.

NAMRU San Antonio Dental Researchers Evaluate Data to Enhance Dental Care for Sailors and Marines

Story by Lt. Noel Dickens, D.M.D., M.P.H., Head of Epidemiology and Biostatistics Department, NAMRU-San Antonio

SAN ANTONIO – As the 104th birthday of the U.S. Navy Dental Corps approaches, the time has come again to appreciate the impact of Navy Dental Research on the health of our fellow Sailors and Marines.

Dental caries is one of the most prevalent chronic diseases of people worldwide, and presently represents the number one cause of dental emergencies among deployed military personnel. Prevention is the most effective means for controlling dental caries and attaining an improved state of oral health among Navy Medicine’s beneficiaries. Providing preventive therapy may also lead to a reduction in future restorative dentistry needs that often affect the deployability of service members.

A Caries Risk Assessment is performed on all active duty dental patients during their annual periodic oral examination. Service members who demonstrate moderate and high risk for dental caries development based on oral hygiene, nutritional habits and other caries risk factors, are considered for assignment to a preventative therapy regimen outlined in the U.S. Navy Oral Disease Risk Management (ODRM) Protocol.



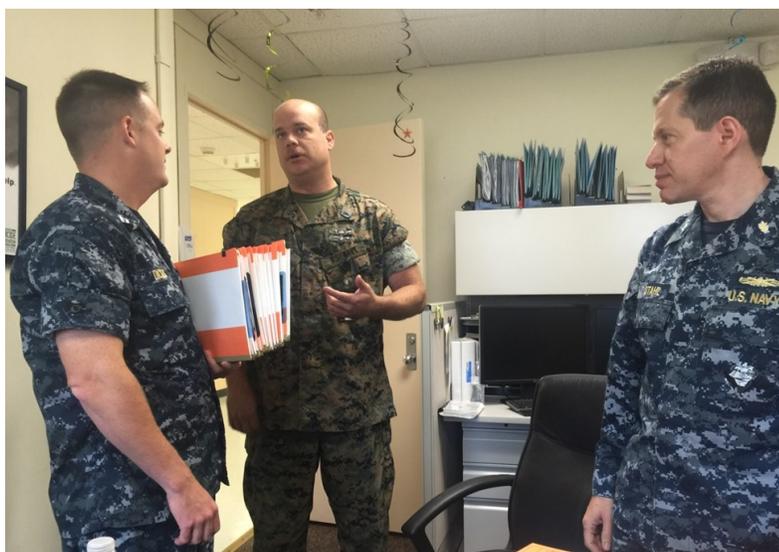
Capt. Jonathan Stahl, D.D.S., Ph.D., M.P.H. (left), head of NAMRU-SA’s Maxillofacial Injury and Disease Department and Lt. Noel Dickens, D.M.D., M.P.H., (right), head of NAMRU-SA’s Epidemiology and Biostatistics Department, in front of 2nd Dental Battalion / Naval Dental Center’s Headquarters at Camp Lejeune, North Carolina during one of their recent data collection trips. (Photo courtesy of NAMRU-SA Public Affairs)

ODRM is aimed at reducing the development of new disease and remineralizing incipient lesions. Dental researchers at Naval Medical Research Unit San Antonio (NAMRU-SA) are

investigating the implementation efficiency of the ODRM Protocol and determining its effect on the Caries Risk Assessment of Active Duty Marines and Sailors.

Data contained in dental records in an observational study (retrospective cohort) are being evaluated to assess the adherence of U.S. Navy dental care providers to the prescribed ODRM program currently established. This study entails physically reviewing dental charts to identify patients who have been classified as moderate and high caries risk according to the Navy’s risk classification criteria.

Using data collected and analyzed from this study, Capt. Jonathan Stahl D.D.S., Ph.D., M.P.H., head of NAMRU-SA’s Maxillofacial Injury and Disease Department, Lt. Noel Dickens, D.M.D., M.P.H., head of the Epidemiology and Biostatistics Department, and other Navy dental researchers from NAMRU-SA, expect to help identify potential modifications to the current ODRM protocol and further enhance the provision of care for Sailors and Marines.



Capt. Jonathan Stahl D.D.S., Ph.D., M.P.H. (right), head of NAMRU-SA’s Maxillofacial Injury and Disease Department and Lt. Noel Dickens, D.M.D., M.P.H., (left), Head of NAMRU-SA’s Epidemiology and Biostatistics Department, discussing patient record management and the study’s inclusion criteria with HMI Berry (center), Leading Petty Officer, at H-1 Dental Clinic, 2nd Dental Battalion / Naval Dental Center, Camp Lejeune, North Carolina. (Photo courtesy of NAMRU-SA Public Affairs)

Meeting of the 2016 National Neurotrauma Symposium (NNS)

By Lt. Cmdr. Peter B Walker, Naval Medical Research Center

From July 26-29, representatives from the Neurotrauma Department (NTD) at the Naval Medical Research Center (NMRC) in Silver Spring, Maryland attended the 2016 National Neurotrauma Symposium (NNS) to showcase the department's ongoing work in clinical and preclinical models of concussion with and without polytrauma, Post-Traumatic Stress Disorder (PTSD), and blast-related neurotrauma. The event hosted more than 500 oral and poster presentations represented by government, industry and academia.

The NTD played a significant role at this annual meeting, offering 10 different oral and poster presentations. These presentations highlight the outstanding ongoing work within the department and are representative of how NTD strives to affect warfighter care in a positive manner.

For example, the work by Dr. Francois Arnaud and her group has sought to better understand the interaction between stress and aeromedical evacuation. In a poster presented on the first two days of the conference, Dr. Arnaud and her group demonstrated that aeromedical evacuation did not appear to worsen outcomes in a rodent model of TBI.

In a related study, Lt. Melissa Mehalick described a computational model of animal survivability following aeromedical evacuation. In this model, the authors demonstrated that, rather than hypobaric conditions, physiologic conditions such as blood loss and increased stress may be more predictive of survivability.

In a study of blood substitute and oxygen therapeutics, Dr. Mullah and Dr. Abutarboush presented safety and efficacy of one of the oxygen therapeutic products

they have screened for use as a phosphorescence quenching oxygen detection method. A different oxygen therapeutic product from the mentioned study was followed up in a lab model and findings were presented in a poster presented by Dr. Haque and Dr. Mullah.

In a blast TB- related vasoactivity study, Dr. Abutarboush, Dr. Mullah and Dr. Lashof-Sullivan presented characterizing the detail vasoactivity of cerebral arterioles following blast TBI in a time and dose dependent manner. This paper was eventually chosen as one of twenty finalists for best abstract at the meeting and is representative of the high-quality science being conducted within the department.

(Continued on page 10)



Standing: (left to right) Lt. Cmdr. Peter B. Walker, Dr. Ashraful Haque and Dr. Saad Mullah. Seated: (left to right) Dr. Anna Tschiffely, Dr. Rania Abutarboush and Lt. Melissa Mehalick. (Photo by NMRC Public Affairs)

Meeting of the 2016 National Neurotrauma Symposium (NNS)

(Continued from page 9)

In a recent addition to the symposium, the conference organizers arranged for a “data blitz.” This format provided several authors a two-minute opportunity to discuss some of the parameters of their studies and to encourage attendees to visit posters and engage with other researchers.

The NTD was represented here by Dr. Rania Abutarboush who presented their work on, “N-acetylcysteine Amide Ameliorates the Blast-Induced Changes in Blood-Brain Barrier Integrity and Intracranial Pressure in a Rat Model.” In this work, Dr. Abutarboush and her colleagues examined the effects of treatment with the antioxidant N-acetylcysteine amide (NACA) after blast exposure on intracranial pressure and the impairment of the blood brain barrier.

The NTD, which has been a part of NMRC since 2005, has made great strides within the research community. The department is comprised of 30 uniformed, government service and contract scientists, and receives competitive funding from sources including the Office of Naval Research, Department of Health Affairs, and others to support their mission to further the understanding neurotrauma.

NTD Presentations at the 2016 National Neurotrauma Symposium (NNS):

- *“Behavioral Assessment following Blast-Exposure in a Rat Model of Mild Traumatic Brain Injury”* Amanda Glueck, Anna Tschiffely, LCDR Peter Walker, LT Jacob Norris, Stephen Ahlers
- *“Exposure to Blast Overpressure Alters Cerebrovascular Reactivity in Rats”* Rania Abutarboush, Saad H. Mullah, Margaret Lashof-Sullivan, Melissa Mehalick, Michael Shaughness, Richard McCarron, Stephen Ahlers
- *“Cerebro-Vascular Reactivity and Brain Oxygenation with Perfluorocarbon Oxycyte after TBI in Rats”* Saad Mullah, Rania Abutarboush, Biswajit Saha, Ashraful Haque, Peter Walker, Françoise Arnaud, Paula Moon-Massat, Anke Scultetus, Charles Auker, Richard McCarron
- *“Examining animal survivability following stress, acute low level blast exposure, and hypobaric conditions”* LT Melissa Mehalick, LCDR Peter Walker, Françoise Arnaud, Eric Maudlin-Jeronimo, Richard McCarron
- *“Does blast overpressure exposure induce functional alterations resulting in behavioral and molecular changes?”* Anna E. Tschiffely, Amanda C. Glueck, LCDR Peter B. Walker, LT Jacob N. Norris, Stephen T. Ahlers
- *“N-acetylcysteine Amide Ameliorates the Blast-Induced Changes in Blood-Brain Barrier Integrity and Intracranial Pressure in a Rat Model”* Rania Abutarboush, Usmah Kawoos, Sydney Zarello, Kevin Nader, Jason Lankasky, Richard M McCarron, Mikulas Chavko
- *“Acute Changes in Vascular Reactivity following Blast TBI at 37, 75, and 140 kPa”* Margaret Lashof-Sullivan, Saad Mullah, Rania Abutarboush, Mike Shaughness, Melissa Mehalick, Richard McCarron, Stephen Ahlers
- *“Effect of Blast and Hemorrhage Injury following Acute Stress in a Rat Model”* Françoise Arnaud, Eric Maudlin-Jeronimo, Georgina Pappas, Richard McCarron
- *“Evaluation of Perfluorocarbon NVX-108 for Brain Tissue Oxygenation after Fluid Percussion Traumatic Brain Injury in Swine”* Ashraful Haque, Biswajit Saha, Brittany Hazzard, Françoise Arnaud, Saad Mullah, Charles Auker, Richard McCarron, Paula Moon-Massat, Anke Scultetus
- *“Polytrauma, Stress and Hypobaric Transport in a Rat Model”* Françoise Arnaud, Eric Maudlin-Jeronimo, Richard McCarron



Part V: The Mosquito Fighters, Solving the Yellow Fever Mystery

By André B. Sobocinski, Historian, BUMED

“The specific agent of the causation of yellow fever exists in the blood of the patient during the first three days after attack after he ceases to be a menace to the health of others. A mosquito of a single species. . .investing the blood of a patient during this infective period, is powerless to convey this disease to another person until twelve days have elapsed but can do so for an indefinite period.”

~Major Walter Reed, MC, USA, 6 February 1901

The very idea bugs, specifically mosquitoes, could transmit disease was nothing new by 1900. As far back as the 1840s, an Alabama-based physician named Josiah Nott—who had lost four children to yellow fever—speculated that insects were intermediate hosts for the disease. Thirty years later, while on a survey expedition to Central America, a U.S. Navy physician named John Bransford noted that mosquito nets could prevent febrile illnesses. But nearly a decade later when Dr. Carlos Finlay put forth his theory of mosquito-based transmission, medical science still turned a blind-eye to a vector-based cause for yellow fever.

Enter the U.S. Army’s Medical Commission.

Organized by the Army Surgeon General George Sternberg in May 1900, the Commission (aka, the Yellow Fever Commission) was established to root out the causes of disease plaguing troops stationed in Cuba. The four-person team was comprised of a former frontier doctor named Walter Reed, his longtime associate Assistant Surgeon James Carroll, and two contract Army medical officers—the bacteriologist Jesse Lazear and the pathologist Aristides Agramante.

When the Commission first convened at the Las Animas Hospital in Havana, it was

a bacterium (*Bacillus icteroides*) and not the mosquito that was the target of their research. Only after blood cultures of patients proved negative for the organism did the Commission finally turn its focus on the mosquito, and Dr. Finlay himself. Finlay, a willing and eager partner to the team, would share decades of research and provide them with the very mosquito eggs that would be the basis of their experiments.

Over the summer and fall of 1900, the Commission took on volunteers, and even two members—Carroll and Lazear—exposed themselves to what they suspected were disease-carrying mosquitoes.

Before falling victim to yellow fever in this first phase of mosquito experiments, Lazear would collect the evidence that would substantiate an existing theory of “extrinsic incubation.” The concept had been proposed two years earlier by a public health doctor named Henry Rose Carter. While serving in a rural Mississippi town hit by yellow fever, Carter observed that people in the same household were more susceptible to the disease two-weeks after the first infection. He held that yellow fever required a period to incubate within an intermediate host before it could be transmitted. Later while stationed as a



Yellow Fever patient in Havana, Cuba, ca. 1898. Courtesy of the National Library of Medicine.

quarantine officer in Havana, Carter would meet with Lazear and provide him with a still unpublished paper outlining his observations.

In November 1900, at a new facility named in honor of their martyred colleague (“Camp Lazear”), the Commission began their second phase of investigations into mosquito transmission and began delving into what was called the “fomite theory.”

(Continued on page 12)

Part V: The Mosquito Fighters

(Continued from page 11)

Some in the medical profession still asserted that yellow fever could be transmitted through so-called “fomites” or objects found in bloodied, soiled and vomit-covered bedding and clothing belonging to the disease victims. To test this theory, the Commission sealed volunteers in an unventilated cabin containing stinking, contaminated articles to see if the infection could be spread. Despite the unpleasantness of the experience, not one volunteer would fall ill. These fomite experiments would be continued in 1901 by U.S. Navy Medical Director John Walton Ross, and Cuban doctor Juan Guiteras at the Las Animas Hospital, adding credence to the Commission’s findings.

The Commission’s experiments concluded in January 1900. A month later, Reed presented their findings to the Pan-American Medical Congress on February 6, 1901 in Havana. Even though history would look upon this as the end of the yellow fever mystery, public and medical opinion did not immediately shift. In fact, Carroll and others would continue investigating mosquito-borne transmission in Havana.

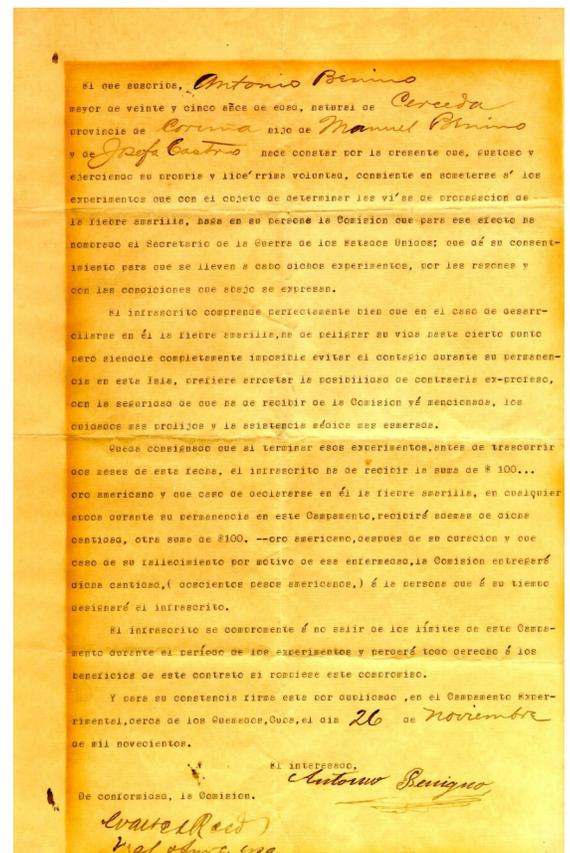
The Commission’s findings would spur new joint-service sanitation regulations in Havana authored by Med. Director Ross and Col. William Crawford Gorgas.

Gorgas would lead the campaign to eliminate mosquito breeding areas throughout the city. The effort would see a reduction of reported yellow fever cases in Havana from 1,400 in 1900 to zero cases in 1902. In 1904, Gorgas would continue the campaign in Panama against malaria-producing mosquitoes helping to ensure successful completion of the Panama Canal.



Above: Yellow Fever Commission. Robert Thom painting showing yellow fever commission members in Cuba. Left to right: William Crawford Gorgas, Aristides Agramonte, Carlos Finlay, James Carroll, and Walter Reed. Courtesy of www.historyofvaccines.org

Right: Yellow Fever Informed Consent Document (In Spanish), November 26, 1900. One of the hallmarks of the Commission’s study was the use of informed consent. Volunteers—Army soldiers, Cuban natives, and even a civil servant—were notified of the risk and each were paid \$100 in gold for participation; an additional \$100 was given if they fell ill. Courtesy of University of Virginia.



NSMRL Technical Director Retires

Story by NSMRL Public Affairs

GROTON, Ct.— Dr. Jerry Lamb, technical director for Naval Submarine Medical Research Laboratory (NSMRL) in Groton, Connecticut, retired July 15 after more than 26 years of federal government service. He has been technical director at NSMRL for 14 years and had prior service at the Naval Underwater Systems Center (NUWC) in New London and Newport, Rhode Island as well as in industry and academia. Most of Dr. Lamb's professional career has been associated with submarine technology, simulator development, training systems and human performance research.

While at NSMRL, Dr. Lamb was responsible for the conception and long-range development of an integrated research program; had general oversight of NSMRL's research program; and was a representative of the laboratory to sponsors at national and international meetings. In that time, he initiated new programs with the Joint Non-Lethal Weapons Directorate for diver deterrent, the United States Coast Guard (USCG) R&D Center for port protection, and NASA for team performance in isolated, confined and extreme environments. He personally led efforts in reducing the vulnerability of high-value maritime assets for the Joint Test and Evaluation command and in developing measures of team performance which were adopted by the submarine force.

After receiving his PhD in Experimental Psychology from the University of Connecticut, Dr. Lamb became Manager of Human Factors for the Electric Boat Division of General Dynamics. The group developed ship control training displays and procedures for submarine crews,



Dr. Jerry Lamb, technical director for Naval Submarine Medical Research Laboratory (NSMRL). (Photo courtesy of NSMRL Public Affairs)

human factors for manned submersibles, conducted multiple psychological studies for Office of Naval Research (ONR), and produced the first computer-based tactical display for submarine use.

After a stint as worldwide head of General Electric's Information Systems Human Factors, Dr. Lamb joined what is now NUWC as a Research Psychologist studying human information processing as applied to digital sonar systems. After a year as a Research Fellow at MIT's Sloan School of Management, he became Program Manager for all submarine training programs where he employed the first Sonar On-Board Trainer integrated with the tactical system.

As Head of the Computer Systems Division, he investigated new display concepts and developed in-house real-time computer display systems. Dr. Lamb personally participated in the design and procurement of a second-generation airborne signal processor. His final position at NUWC was as a Senior Executive Service (SES) department head for submarine fire control systems where he was responsible for the department's efforts for all submarine torpedo and Tomahawk fire control systems development, maintaining configuration of fielded systems, the new Trident class combat systems, and conducting research into new processing/display concepts for submarine use.

After leaving NUWC, Dr. Lamb was President and CEO of Ship Analytics in North Stonington, CT, a simulator and training company specializing in ship bridge simulators and submarine trainers and training for military, commercial, and foreign customers. Dr. Lamb was General Manager of the Systems and Simulation

Division of Contraves USA, a high technology simulation training operation in Tampa, Florida. Products included flight simulators for the U. S. Navy and Taiwanese Air Force and air traffic control and control tower simulators for the Navy, Army and Air Force. While in Florida, he was a member of Tampa's High Technology Council and an advisor to the University of Central Florida's Simulation Institute.

Before joining NSMRL, Dr. Lamb held positions in academia. Dr. Lamb was the Dean for the University of New Haven's Southeastern Campus in New London,

(Continued on page 14)

NAMRU-San Antonio Tells Their Navy Medicine Story in New Command Video

Story courtesy of NAMRU-San Antonio Public Affairs

SAN ANTONIO – The Naval Medical Research Unit San Antonio (NAMRU-SA) announces the release of their first command video. NAMRU-SA, located at JBSA Fort Sam Houston in San Antonio, Texas, unveiled the six-minute video on YouTube in June.

“I think video offers a powerful way to tell our research story and reach audiences of stakeholders and potential collaborators in the academic and military communities that are sometimes difficult to reach,” says Capt. Elizabeth Montcalm-Smith, NAMRU-SA commanding officer.

The video features a combination of NAMRU-SA’s science matter experts as explainers and footage of multidisciplinary Navy researchers engaged in developing technologies and treatments to deliver novel biomedical solutions relevant to the military.

“I think what we’ve shown we’re producing high-quality work that is

innovative and supports readiness by finding solutions to the critical needs of warfighters and the clinicians who treat them,” says Montcalm-Smith.

NAMRU-SA’s research and development narrative includes phage therapy, new treatments that could eradicate antibiotic resistant pathogens, non-pharmaceutical approaches to treating combat wounds,

novel advances in the evaluation of interventions to stem and control hemorrhage, and providing resuscitation to injured warfighters.

This is NAMRU-SA’s first venture into the digital video ecosystem to represent Navy Medicine, while also being the distinct voice sponsors and clients have come to expect from NAMRU-SA.



Capt. Elizabeth Montcalm-Smith, commanding officer, NAMRU-SA (left) completes closing segment of NAMRU-SA’s command video with producer, Mr. Todd Folts, Defense Media Activity, DoD. (Photo courtesy of NAMRU-SA Public Affairs)

“This project has been on our wish list for years, and then finally the time was right. We had the scale, we had the production crew, and we had the stories to tell,” said Montcalm-Smith. “Video challenges your aptitude for telling stories that really represents the essences of scientific research. We hope our command video can be an incredibly useful model for the other research commands to consider,” says Montcalm-Smith.

You can view the NAMRU-SA Command video on [YouTube](#) and on the [NAMRU-SA website](#).

NSMRL Technical Director Retires

(Continued from page 13)

Connecticut. His primary task was to respond to the changing economic climate by developing innovative approaches to the education and re-education of a technically skilled population. Dr. Lamb implemented a unique management training program using computer-simulated case studies and war gaming techniques for a major insurance company, which was then used for multiple other organizations including a local hospital and a Native American tribe.

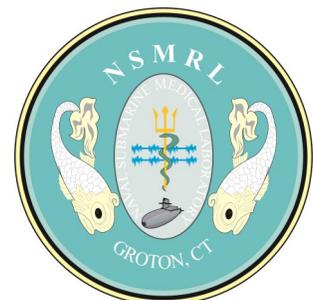
In 1999, Dr. Lamb became Dean of the School of Continuing Education at Eastern Connecticut State University where he had responsibility for all non-credit activities at the university as well as all part-time credit students. In 2001, he transferred to the University of Connecticut as Associate Dean of the College of Continuing Studies. Dr. Lamb led a U. S. Department

of Labor grant for the pharmaceutical Clinical Data Manager training. The program won a national award for excellence from the U.S. Department of Labor Employment and Training Administration in 2004.

Dr. Lamb is a three-time Chair of the Board of Directors President of the Southeastern Connecticut Enterprise Region (seCTer), an economic development non-profit whose predecessor he co-founded in 1985. Dr. Lamb has been an adjunct faculty member at Connecticut College, Mitchell College and the University of Connecticut.

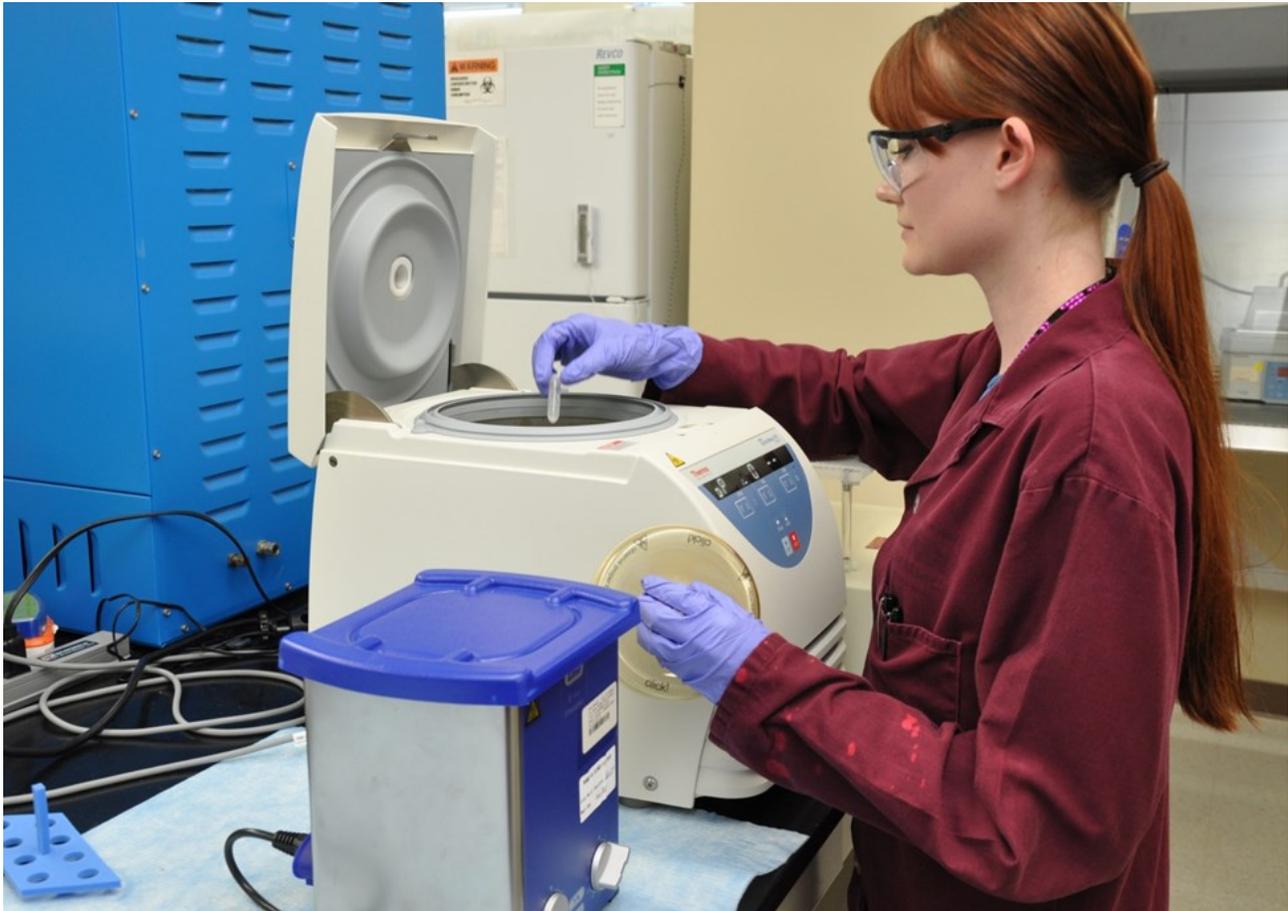
At a retirement luncheon held July 15, Captain Fred Yeo thanked Dr. Lamb for his expertise over the 14 years at NSMRL. “Many lives are better because of Dr. Lamb’s contributions to the Submarine community,” Yeo noted.

When asked what he would miss the most about NSMRL, “As with most people leaving a position they have been working at for some time, I will miss all the people who I interact with on a day-to-day basis,” Lamb replied. “I have enjoyed my tour at NSMRL; the lab is successful due to the hard work and talented people who have come before me and who will come after me. This hard work and talent flows through the lab continuing to make it work and a success.”



Meet NAMRU-SA's 2016 NREIP Summer Intern, Amy Veals

Story by Amy Veals, 2016 NREIP Intern, NAMRU-San Antonio



NAMRU-SA 2016 NREIP summer intern, Amy Veals is centrifuging bacterial cells for the development of a novel antivenom. (Photo courtesy of NAMRU-SA Public Affairs)

SAN ANTONIO --Each year Naval Medical Research Unit San Antonio (NAMRU-SA) sponsors a summer intern through the Naval Research Enterprise Intern Program (NREIP). This 10-week program accepts undergraduate and graduate students giving them an opportunity to gain laboratory experience, explore new areas of research, and encourage careers in STEM fields.

My name is Amy Veals and I am this year's NAMRU-SA NREIP summer intern. Originally fairing from Gilbert, Arizona, I have completed my junior year of a Bachelor of Science in Molecular and Cellular Biology at the University of Arizona. Upon the completion of the NREIP internship, I will return to my studies. My advisor at the University of Arizona introduced me to this opportunity. I realized this was an exciting opportunity

that would broaden my horizons and give me invaluable research experience. I was thrilled when I was informed of my acceptance to the program.

Under the guidance and mentoring of Navy researcher, Yoon Hwang, Ph.D., I have been involved in a project focused on the development of a novel antivenom. This project will lay the groundwork for research into the development of a universal antivenom. At the conclusion of this 10-week project, I will present a poster of my work to the NAMRU-SA Command.

Throughout my time here at NAMRU-SA, I have gained a great deal of critical research experience and knowledge. The NREIP program has provided me with a unique and memorable opportunity and allowed me to gain hands-on experience I

would not have been able to attain elsewhere. It has provided me with valuable insight into the world of research in Navy Medicine. Dr. Hwang and his team, Matthew Kay, Ph.D., and James Titus, M.S., have introduced me to many aspects of research such as daily laboratory work, safety, scientific publication and the grant writing process.

The NREIP program has allowed me to expand my knowledge and skill set, which I can take with me as I finish my Bachelor's and pursue a graduate degree. My goal is to continue on into a Masters in Public Health program. I am so grateful for the opportunity to work with NAMRU-SA. I have had the pleasure of working with many passionate, talented people and gained the skills and confidence I need for a successful career in science.