Trauma-Related Infections Research Area

2018 Annual Report
The Trauma-Related Infections Research Area is part of the Infectious Disease Clinical Research Program (IDCRP) based at the Uniformed Services University of the Health Sciences (USU), Department of Preventive Medicine and Biostatistics. There are three primary research protocols: 1) Trauma Infectious Disease Outcomes Study (TIDOS); 2) Trauma-Associated Osteomyelitis; and 3) Invasive Fungal Wound Infection (IFI) Molecular Diagnostics.

TIDOS is the centerpiece protocol of the Trauma-Related Infections Research Area and involves investigators from a variety of disciplines, including infectious disease, trauma surgery, orthopedics, surgical pathology, epidemiology, statistics, microbiology, and molecular biology. Both TIDOS and Trauma-Associated Osteomyelitis are made possible through the cooperative research of investigators and personnel across multiple clinical sites. In addition, TIDOS involves collaborations with the U.S. Army Institute of Surgical Research, Walter Reed Army Institute of Research, Naval Medical Research Center, and the United Kingdom Wound Infection Surveillance Programme. The IFI Molecular Diagnostics protocol also includes collaborations with the DoD Joint Pathology Center and the University of Texas Health Science Center at San Antonio.

TIDOS was developed with the following objectives:

- Establish a cohort of DoD beneficiaries and active-duty personnel with trauma-related injuries to determine short- and long-term outcomes and potential risk factors associated with infections.
- Describe the infectious disease epidemiology of trauma-related injuries or other nosocomial infections in the cohort population.
- Establish a database and bacterial/fungal isolate repository to support future approved studies focused on informing clinical management, disease prevention, or clinical trial design.
- Inform DoD efforts to develop real-time tools for combat-related health event/outcome analysis secondary to trauma-related infections during wartime.

TIDOS has been supported by U.S. Navy Bureau of Medicine and Surgery (BUMED) Wounded, Ill, and Injured Program (WII), the National Institute of Allergy and Infectious Diseases (NIAID), the DoD Global Emerging Infections Surveillance (GEIS), the Military Infectious Diseases Research Program (MIDRP) through the Defense Health Program, and U.S. Army Defense Medical Research and Development Program (DMRDP) funding. Trauma-Associated Osteomyelitis has been supported by BUMED WII and NIAID. The IFI Molecular Diagnostics protocol has been supported by DMRDP. The IDCRP was formed through an Interagency agreement between NIAID and USU and is supported by the Henry M. Jackson Foundation pursuant to a cooperative agreement.

The views expressed are those of the authors and do not reflect the official views of USU, Henry M. Jackson Foundation, National Institutes of Health or the Department of Health and Human Services, DoD or the Departments of the Army, Navy or Air Force.
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Letter from the Trauma-Related Infections Research Area Director

The close of 2018 brings about the end of another successful year for the Trauma-Related Infections Research Area as we move into our 10th year of collaborative research. As part of the Trauma Infectious Disease Outcomes Study (TIDOS), three core manuscripts presenting the methodology and main findings from the overall study population’s experience during the initial post-trauma hospitalization, the subsequent experience following initial discharge in the overall DoD TIDOS cohort, and lastly, the VA-specific ongoing infection complication concerns as the personnel transition from active duty to veterans. Multiple analyses were also completed under the TIDOS Multidrug-Resistant and Virulent Organisms (MDR/VO) Trauma Infections Initiative. New analyses are underway to examine the bacterial microbiome, antimicrobial resistance emergence, interaction of common wound bacteria (e.g., ESKAPEE pathogens), biofilm dispersal agents, and clinical outcomes in relation to wound microbiology and biofilm production.

In 2018, the DoD Technical Report with the findings of the Invasive Fungal Infection (IFI) Molecular Diagnostics Study, assessing PCR-based methods to support earlier identification of fungal elements, was finalized and presented to the Joint Trauma System for incorporation into clinical practice guidelines for use within U.S. military hospitals. Follow-on assessment of diagnostic techniques and guidance on future clinical application is underway, as well as finalizing a manuscript detailing the initial PCR analyses. A comprehensive review of patients with laboratory evidence of fungus in wound cultures or tissue specimens in relation to temporal relationship with surgical findings has led to refinement of the IFI definition and classification scheme. A manuscript detailing this revised classification system, as well as an assessment of IFI epidemiology, was submitted for journal consideration.

The Trauma-Associated Osteomyelitis Study also had successes over the past year. The tibia case-control and case-case analyses were published and accepted for publication, respectively. The open femur fracture case-control analysis was accepted for publication and a manuscript presenting findings from the upper extremity case-control analysis is under journal consideration. Manuscripts with findings from the femur and upper extremity case-case analyses are nearing finalization. These analyses expand upon the available knowledge of injury- and clinical management-specific factors related to initial and recurrent osteomyelitis. A major effort undertaken at the Veterans Affairs St. Louis Health Care System has extended follow-up assessment (up to 10-15 years from initial injury) through review of VA health records, as well as added the analysis to assess impact on comorbidities. The analysis of this additional data will be completed in 2019.

In the upcoming year, TIDOS analyses will continue to focus on blast-related wound infections, wound microbiology and multidrug resistance, and clinical practice guideline adherence/refinement and antibiotic stewardship. Of note, a Military Medicine supplement on the accomplishments of the Trauma-Related Infections Research Area is in development.

The Trauma-Related Infection Research Area’s aims and objectives remain high priorities for military medicine and continue to have significant clinical relevance during inter-war periods for the purpose of improved understanding for ongoing issues among wounded personnel, and to enhance efforts with evidence-based approaches in preparation for the next conflict.

David R. Tribble, MD, DrPH
Science Director, IDCRP
Director, Trauma-Related Infections Research Area
The Infectious Disease Clinical Research Program (IDCRP) has expanded understanding of treatment strategies for combat-related trauma infectious complications. Infections in these complex wounds remain a major challenge, requiring well-designed clinical research in the areas of prevention and management. Treatment strategies have been complicated by the emergence of multidrug-resistant bacterial organisms and aggressive new threats, such as invasive molds. Furthermore, infections following open fractures (including hardware-related) often create challenges due to potential for chronic or recurrent infections.

During 2017, the strategic aims of the Trauma-Related Infections Research Area were revised to reflect research priorities for the Military Health System. Four research aims were identified to advance the Research Area in the desired direction, in line with IDCRP’s overall mission, vision, and goals.

**Aim 1:** Describe the epidemiology, clinical characteristics, and outcomes among combat blast-related wounds and infections

**Aim 2:** Compare clinical outcomes and antibiotic exposure to specific microbiological factors in colonizing or infecting organisms isolated from trauma patients (Multidrug-Resistant and Virulent Organisms Trauma Infections Initiative)

**Aim 3:** Evaluate short- and long-term health impacts of combat-related infections through ongoing care in DoD and/or VA following initial discharge

**Aim 4:** Assess adherence and outcomes to Joint Trauma System Clinical Practice Guidelines and antibiotic stewardship in support of the U.S. strategy in Combating Antibiotic Resistant Bacteria

Within each of the different trauma-related study protocols, a great deal of progress has been made to complete these research aims.
Blast-Related Trauma and Wound Infections

With the high incidence of blast-related trauma during the wars in Iraq and Afghanistan, assessing the potential long-term impact of these serious injuries is a priority of the Military Health System. In particular, research on the long-term morbidity and disability related to wound infections following blast trauma is of high importance. During 2018, the 3rd Japan-US Technical Information Exchange Forum on Blast Injury (JUFBI) was held to bring together both military and civilian experts from various regions of the world to share their knowledge and experiences related to blast trauma (Photo 1).

At the 2018 JUFBI, Dr. David Tribble (IDCRP/USU) presented an overview of blast-related wound infection epidemiology and microbiology with emphasis on the high proportion of wound colonization and infections with multidrug-resistant organisms. In particular, approximately one-fourth of blast casualties developed at least one trauma-related infection with an average of two infections per patient. Extremity wound infections are the most frequent complication with ~57% of skin and soft-tissue infections being polymicrobial and ~37% isolating multidrug-resistant organisms. Approximately half of blast-related osteomyelitis are also polymicrobial with a high proportion of multidrug-resistance.

As a follow-on to the 2016 DoD Blast Injury Research Program’s International State-of-the-Science Meeting on Minimizing the Impact of Wound Infections Following Blast-Related Injuries, a session at the 2018 Military Health System Research Symposium (MHSRS) focused on discussing advances in blast injury and wound infection research to fill critical knowledge gaps (Photo 2). Findings from multiple TIDOS analyses were presented at the MHSRS session (1 oral presentation and 6 posters) with topics on invasive fungal wound infections (see Page 5), wound microbiology (see Pages 9-10), and antibiotic practice patterns related to extremity wound infections following blast trauma (see Page 4).

Furthermore, TIDOS investigators were invited to prepare a chapter reviewing current TIDOS research efforts regarding blast-related wound infections for the DoD Blast Injury Research Program Coordinating Office’s Annual Report to the Executive Agent for Medical Research for Prevention, Mitigation, and Treatment of Blast Injuries.

In the forthcoming year, blast-related trauma and resultant infections will continue to be a high priority of the Trauma-Related Infections Research Area. The next steps will focus on the examination of best practices regarding antibiotic prophylactic prevention and medical/surgical treatment of blast wound infections, along with the comprehensive assessment of infection risk factors and clinical outcomes for both extremity and non-extremity infections. Factors related to social, mental, and physical health well-being among cohort enrollees following the initial trauma hospitalization will also be evaluated.

Photo 1: (from left) Mr. Michael Leggieri, Dr. David Tribble, Lt. Gen Norihiko Yamada, and Dr. Nariyoshi Shinomiya at 2018 JUFBI

Photo 2: Dr. Laveta Stewart presenting at the 2018 MHSRS
Extremity wound infections contribute the highest proportion to the infectious disease burden among combat casualties. In particular, deep soft-tissue infections (DSTIs) are a common infectious complication of combat-related open extremity wounds and are often associated with short and long-term disability, resulting from multiple surgical procedures and limb loss. Moreover, DSTIs have the potential of progressing to become osteomyelitis, which requires prolonged use of antibiotics. Due to their seriousness and the possible involvement of multidrug-resistant organisms, broad-spectrum antibiotics are frequently prescribed to manage DSTIs. Nevertheless, medical management in relation to extremity wound infections (DSTIs and osteomyelitis) during the recent wars has not been well described.

As a result, Dr. Laveta Stewart (HJF; IDCRP/USU) led the TIDOS analysis to characterize antibiotic treatment associated with combat-related extremity wound infections and to identify frequent practice patterns for use in the assessment of clinical outcomes. Receipt of antibiotics was assessed in three periods surrounding the infection diagnosis date: 4-10 days prior to diagnosis; ±3 days of diagnosis; and 4-10 after diagnosis (Figure).

Carbapenems were the most frequently prescribed antibiotics in the week of diagnosis, while it was fluoroquinolones and vancomycin in the pre-diagnosis and post-diagnosis period, respectively. A higher proportion of patients also received at least two antibiotics during the week of diagnosis compared to the other periods. Overall, the findings demonstrate substantial variability in antibiotic prescribing practices. These findings were presented at the 2018 Military Health System Research Symposium and a manuscript has been submitted for journal consideration.

Outcomes of 259 patients with DSTIs were examined with regards to whether they received a carbapenem (without vancomycin), vancomycin (without carbapenem), carbapenem plus vancomycin, or only other antibiotics for at least two days (Flowchart).

Similar injury mechanisms among patients in the antibiotic regimen groups were observed (>90% with blast injuries); however, the patients who received ‘other’ antibiotics had lower injury severity and reduced rates of amputations compared to patients in the groups that received carbapenem and/or vancomycin. In addition, patients in the ‘other’ antibiotics group were less frequently admitted to the intensive care unit at either Landstuhl or the U.S. hospitals. To assess the optimal effectiveness of treatment strategies following DSTI diagnosis, an analysis comparing outcomes (e.g., timing of wound closure) among the antibiotic regimen groups is underway. These data, along with other published reports, will be utilized to support the development of recommendations for the management of DSTIs.
Early diagnosis combined with aggressive surgical debridements and directed antifungal chemotherapy are integral for improving outcomes of patients with invasive fungal wound infections (IFIs). As a result, Dr. Anuradha Ganesan (HJF; IDCRP/USU; WRNMMC) led the retrospective analysis evaluating the utility of a polymerase chain reaction (PCR)-based assay for rapid detection and increased accuracy of fungal elements in formalin-fixed paraffin-embedded surgical pathology specimens to support early diagnosis of IFI. When compared to histopathology as the reference standard, the PCR-based assay was found to be highly specific (99%); however, with lower sensitivity (63%). Sensitivity improved to 83% in tissues with angioinvasion. These findings were presented at the 2018 Military Health System Research Symposium and the 2018 IDSA ID Week (Photo).

In 2018, following review by subject-matter experts, the Tissue-Based Molecular Diagnostics Evaluation in Combat-Related Invasive Fungal Wound Infections Technical Report was finalized and submitted to the Defense Medical Research and Development Program and the Military Infectious Diseases Research Program (MIDRP). The report was also submitted to the Joint Trauma System to support clinical practice guideline recommendation refinement. A manuscript with the findings is nearing finalization.

Follow-on analysis through a grant from MIDRP incorporating specimens collected from IFI subjects with positive wound cultures and negative histopathology are planned to further assess the utility of the PCR-based assay.

Following a comprehensive review of the TIDOS database, the IFI definition/classification scheme was refined based on the timing of laboratory fungal evidence to include categories of IFI, High Suspicion of IFI, and Low Suspicion of IFI. Criteria for an IFI classification was laboratory evidence (histopathology and/or culture) of a fungal infection at the time (or after) wound necrosis is observed. Patients with laboratory evidence of a fungal infection who did not meet IFI criteria were classified as High Suspicion if they had an infection event and receipt of antifungal therapy for ≥10 days. If the patient received antifungal therapy for <10 days, they were still classified as High Suspicion if there was an amputation within 10 days of injury or the patient died. Patients with fungal laboratory evidence who did not meet the criteria of an IFI or High Suspicion were classified as Low Suspicion.

Among 1932 combat causalities with open wounds sustained in Afghanistan and admitted to participating U.S. hospitals between 2009 and 2014, 246 (13%) had laboratory evidence of a fungal infection, of which 94 were classified as an IFI (Flowchart). Patients classified as an IFI and High/Low Suspicion were comparable regarding mechanism of injury and occurrence of traumatic amputations (Figure). Nevertheless, a larger proportion of IFI patients required >20 units of blood within 24 hours post-injury. In general, fungi from the order Mucorales were more frequently detected in wounds classified as IFI compared to both High and Low Suspicion wounds. A manuscript with these findings has been submitted for journal consideration.

Figure: Patient characteristics stratified by IFI classification
Patients with open fractures are at risk of developing osteomyelitis, which may result in substantial morbidity, including the need for multiple surgical procedures. Characteristics of patients with open fractures of the tibia, femur, and long bones of the arm were assessed to identify risk factors for the disease in case-control studies. In the femur case-control risk factor analysis (led by LCDR Louis Lewandowski), 103 patients with open femur fractures and osteomyelitis were compared to 64 controls. Patients with osteomyelitis had a significantly longer duration to definitive surgery compared to controls. Being injured between 2003 and 2006, use of antibiotic beads, Gustilo-Anderson fracture classification (modified to include transfemoral amputations), foreign body with implants at fracture site, and Orthopaedic Trauma Association Open Fracture Classification for muscle loss and dead muscle were significantly associated with osteomyelitis risk. The findings demonstrated that patients with open femur fractures resulting in significant muscle damage have the highest risk for developing osteomyelitis. Case-case analyses for each of the bone sites were also conducted to examine the risk of recurrence of the disease following completion of treatment for the initial osteomyelitis.

Findings from the tibia case-control analysis were published in the Journal of Orthopaedic Trauma. Manuscripts reporting on the femur case-control analysis (Journal of Orthopaedic Trauma) and the tibia case-case analysis (Clinical Orthopedics and Related Research) were accepted for publication. A manuscript with findings from the arm long bones case-control analysis also was submitted for journal consideration. Manuscripts with findings from the femur and arm long bones case-case analyses are in development.

Through a bidirectional data use agreement with the Veterans Affairs (VA) St. Louis Health Care System, data abstraction of patients who entered VA healthcare was completed with 532 patients having sufficient information for analysis. The follow-up period for many subjects spans a full decade. Presently, cases of osteomyelitis identified in electronic medical records during VA follow-up have been assessed and verified in accordance with standardized diagnostic criteria. Analysis of the data is underway.
Non-Extremity Trauma-Related Infections

While orthopedic trauma is the predominant injury pattern, complex non-extremity wounds also result from blast trauma with the potential for infectious complications. Approximately 9% of wounded military personnel sustained genitourinary trauma between 2009 and 2014 (Figure). A recent TIDOS analysis, led by Dr. Stephen Liang (Washington University School of Medicine [WUSM]) and Dr. Jay McDonald (St. Louis VA Medical Center, WUSM), assessed the association of genitourinary trauma and urinary tract infections among patients in the TIDOS-VA cohort and a manuscript has been submitted for journal consideration.

Abdominal injuries also account for a high proportion of battlefield trauma (Figure) with the majority being penetrating wounds. While colon injuries, splenectomy, and large-volume blood transfusions are risk factors in civilian trauma, less information is known about the risk of infection with combat-related abdominal trauma. As a result, risk factors for the development of intra-abdominal infections after combat-related abdominal trauma are being assessed in a TIDOS analysis (led by CDR Matthew Bradley; WRNMMC). The study population is restricted to wounded personnel who underwent downrange exploratory laparotomy as part of their initial care. Presently, data abstraction is nearing completion.

Long-Term Quality of Life

Combat-related infections may develop long after the initial hospitalization period. Nonetheless, the relationship between long-term infectious complications and mental health factors is not well understood. In addition, less is known about the mental and physical well-being of wounded warriors after returning to civilian life following discharge from a hospital or recovery center.

As part of the collaboration with the St. Louis VA Medical Center (led by Dr. Jay McDonald), mental health and social factor information have been collected from TIDOS cohort enrollees who have received VA healthcare. This includes information on post-traumatic stress disorder, depression, alcohol use, tobacco use, opioid use, relationship status, living situation, and employment status. These data will be examined to assess the relationship between mental health diagnoses and the risk of infection. Furthermore, TIDOS enrollees have also provided responses to the SF-8 Health Survey during follow-up, which includes questions related to quality of life, physical health, and mental health. These data are presently being evaluated to determine how physical and mental well-being are impacted by injury severity, pattern of injury (e.g., amputation), and trauma-related infections.

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<th>SF-8™ Health Survey—Example Questions</th>
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<tr>
<td>1. Overall, how would you rate your health during the <strong>past 4 weeks</strong>?</td>
</tr>
<tr>
<td>Excellent</td>
</tr>
<tr>
<td>2. During the <strong>past 4 weeks</strong>, how much did physical health problems limit your physical activities (such as walking or climbing stairs)?</td>
</tr>
<tr>
<td>Not at all</td>
</tr>
<tr>
<td>3. During the <strong>past 4 weeks</strong>, how much difficulty did you have doing your daily work, both at home and away from home, because of your physical health?</td>
</tr>
<tr>
<td>Not at all</td>
</tr>
</tbody>
</table>
A priority of the Trauma-Related Infections Research Area is the professional development of clinical research skills for future infectious disease clinicians through the support of Graduate Medical Education (GME). During the past year, multiple residents and infectious disease fellows have conducted analyses, in collaboration with the TIDOS team, under the mentorship of TIDOS investigators: Maj Dana Blyth (BAMC) and Dr. Katrin Mende (HJF; IDCRP/USU; BAMC).

In one analysis, Capt Sarah Schall (BAMC resident) examined the clinical epidemiology of *Clostridium difficile*-associated diarrhea (CDAD) among wounded military personnel. Approximately 1% of patients met criteria for confirmed (laboratory-based) or presumptive (diarrhea with treatment for *C. difficile* in absence of laboratory confirmation) CDAD. The majority of patients sustained blast injuries, resulting in high injury severity. Approximately 74% of the patients had at least one infection diagnosis prior to diagnosis of CDAD, primarily skin and soft-tissue infections and pneumonia. Prior to CDAD diagnosis, 96% of the patients were prescribed antibiotics with 1st generation cephalosporins being predominant, followed by tetracyclines, vancomycin, carbapenems, and fluoroquinolones. Although CDAD was severe or fulminant in 50% of the patients, there were no deaths, and, overall, the rate was very low.

Capt Joseph Yabes (ID Fellow, BAMC) and LTC Paul Robben (ID Fellow, WRNMMC) examined the risk of acute kidney injury in critically-ill trauma patients with ≥48 hours of concomitant vancomycin and extended spectrum β-lactams (piperacillin-tazobactam or other) use. Acute kidney injury was defined by meeting any of the standardized clinical criteria (e.g., RIFLE) 3-7 days after initiation of antimicrobials. Patients who received vancomycin plus piperacillin-tazobactam or vancomycin and other extended spectrum β-lactams were matched 1:3 for analysis. Among patients who received vancomycin plus piperacillin-tazobactam, ~15% met the acute kidney injury criteria, while it was 7% among the patients who received other broad-spectrum β-lactams in combination with vancomycin.

The Sequential Organ Failure Assessment (SOFA) score has been shown to have value in predicting outcomes (e.g., sepsis and mortality) in critically ill civilian populations; however, less is known about the utility of the score with trauma patients. Therefore, Maj Shannon McCarthy (ID Fellow, BAMC) assessed the application of serial SOFA scores in relation to infectious outcomes among wounded military personnel. Among patients admitted to the intensive care unit at Landstuhl (within 4 days of injury) and at participating hospitals in the United States, ~58% were diagnosed with at least one infection following U.S. hospital admission (largely skin and soft-tissue infections and pneumonia). Admission SOFA scores at Landstuhl were independently associated with the development of subsequent infections and had a higher predictive value when compared to injury severity scores.

The findings of the CDAD and acute kidney injury analyses were presented at the 2018 IDSA ID Week (Photo), while the SOFA assessment was presented at the 2018 U.S. Army/U.S. Air Force American College of Physicians Annual Chapter Meeting and the 48th Critical Care Congress (Feb 2019). Additional GME-supported analyses involving *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, and antifungal activity of cerium nitrate are discussed in Microbiological Analyses (pages 9-10).
Multidrug resistance, virulent organisms, and the polymicrobial nature of wounds add to the challenge in caring for wounded warriors with combat trauma-related infections. Led by Dr. Katrin Mende (HJF; IDCRP/USU; BAMC), the TIDOS Multidrug-Resistant and Virulent Organisms (MDR/VO) Trauma Infections Initiative seeks to further the understanding of the microbiology of complex combat-related wounds. The MDR/VO Trauma Infections Initiative is a collaboration between multiple DoD laboratories: Brooke Army Medical Center (BAMC), Naval Medical Research Center (NMRC), U.S. Army Institute of Surgical Research (USAISR), and Walter Reed Army Institute of Research (WRAIR). The goals of the Initiative are to further the understanding of the combat trauma-related wound infection microbiome, assess the impact of multidrug resistance on clinical outcomes, and develop a novel antimicrobial countermeasure evaluation pipeline within the DoD laboratories. An update on the TIDOS MDR/VO Initiative was presented at the 2018 ASM Microbe annual meeting and the 2018 Military Health System Research Symposium (MHSRS) (Photos 1).

Through collaboration with the NMRC and WRAIR Wound Infections Departments, Enterococcus spp. isolates from the TIDOS Microbiological Repository were re-evaluated to confirm or correct species-level identification using an advanced mass spectrometer technique called Matrix-Assisted Laser Desorption/Ionization-Time of Flight (MALDI-TOF). Use of MALDI-TOF improved species level identification for 43 previously unspeciated Enterococcus isolates and corrected the speciation of 2 previously misidentified isolates. Overall, Enterococcus faecium was the greatest contributor followed by Enterococcus faecalis (Table). Following species identification, Enterococcus isolates were screened for antagonistic activity using spot plating and examined for inhibitory activity against planktonic and biofilm phases of the pathogens. The final step involved evaluating the ability of the isolates to antagonize virulence in the Galleria waxworm infection model and in the mouse topical skin infection model (Figure 1).

### Table: Identification of Enterococcus isolates using MALDI-TOF

<table>
<thead>
<tr>
<th>Species</th>
<th>Frequency</th>
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<tr>
<td>E. faecium</td>
<td>77%</td>
</tr>
<tr>
<td>E. faecalis</td>
<td>14%</td>
</tr>
<tr>
<td>E. gallinarum</td>
<td>6%</td>
</tr>
<tr>
<td>E. mundtii</td>
<td>2%</td>
</tr>
<tr>
<td>E. hirae</td>
<td>1%</td>
</tr>
<tr>
<td>E. cassiflavus</td>
<td>1%</td>
</tr>
</tbody>
</table>

In collaboration with USAISR, Enterococcus isolates were also assessed for the capability of forming biofilms. Among initial Enterococcus isolates collected from clinical infection work-ups, 9% exhibited biofilm formation in the presence of human plasma, while it was 14% of Enterococcus isolates from surveillance cultures (colonization).
Under the mentorship of Maj Dana Blyth (BAMC) and Dr. Katrin Mende, CPT Mary Ford (Internal Medicine Resident, BAMC), CPT John Kiley (ID Fellow, BAMC) and MAJ Heather Pomerantz (ID Fellow, BAMC) conducted TIDOS MDR/VO Initiative analyses as part of Graduate Medical Education.

In the first analysis, CPT Ford examined *Pseudomonas aeruginosa* isolates collected from wound and blood cultures from combat casualties (90% with blast trauma). Approximately 10% of patients had *P. aeruginosa* isolates classified as MDR (all with blast trauma), which was associated with higher injury severity and longer duration on ventilators, as well as length of hospitalization. For her *P. aeruginosa* analysis, CPT Ford was awarded 1st Place in the Resident Research Podium Competition at the San Antonio Uniformed Services Health Education Consortium Research Day.

Due to the challenge posed by increasing antibiotic resistance, the characteristics of combat casualties with *Klebsiella pneumoniae* infections were assessed by CPT Kiley. Patients with *K. pneumoniae* infections primarily sustained blast-related injuries (82%), resulting in high injury severity. Approximately half of the patients with *K. pneumoniae* infections had isolates classified as MDR; however, there were no significant difference in characteristics between the patients with and without MDR *K. pneumoniae*. Resistance patterns were also examined by CPT Kiley. The majority of infecting *K. pneumoniae* isolates were collected from respiratory specimens, followed by wound and blood cultures (Figure 2). Isolates were largely susceptible to meropenem, ertapenem, and amikacin, while susceptibility was low for cefazolin, amoxicillin-clavulanate, and ceftriaxone.

Lastly, the *in vitro* antifungal activity of cerium nitrate (topical antiseptic utilized in burn care) against molds and yeasts recovered from the wounds of combat casualties was evaluated by MAJ Pomerantz.

In general, cerium nitrate had very little activity against the mold isolates. The one exception was *Mucor circinelloides* where there was a time-dependent decrease in colony-forming units (CFUs) over a 24-hour period (Figure 3). Cerium nitrate was more effective against *Candida* spp. with time-dependent killing occurring at all tested concentration. MAJ Pomerantz was awarded 2nd Place in the podium presentation competition at the 2018 Annual Meeting of the Texas Infectious Diseases Society.

**Figure 2:** *Klebsiella pneumoniae* isolate sources

**Figure 3:** *Mucor circinelloides* CFUs after exposure to cerium nitrate

LCDR Mark Simons (NMRC) and Dr. Lee Mangum (USAISR) presented findings from the *Enterococcus* spp. and biofilm analyses, respectively, at the 2018 MHSRS (Photos 1). CPT Ford presented *P. aeruginosa* findings at 2018 MHSRS. CPT Kiley and MAJ Pomerantz presented their findings at the 2018 IDSA ID Week (Photos 2).

**Photos 2:** [from left] CPT Ford at 2018 MHSRS; CPT Kiley and MAJ Pomerantz at 2018 IDSA ID Week
Publications in 2018


Petfield JL, Tribble DR, Potter BK, Lewandowski LR, Weintrob AC, Krauss M, Murray CK, Stinner DJ, the Trauma Infectious Disease Outcomes Study Group. Is Bone Loss or Devascularization Associated with Recurrence of Osteomyelitis in Wartime Open Tibia Fractures? Clinical Orthopaedics and Related Research. Accepted for publication.


Posters and Presentations in 2018

San Antonio Uniformed Services Health Education Consortium Research Day, 26 April 2018, JBSA Fort Sam Houston.

* Ford M. Clinical Characteristics and Resistance Patterns of Pseudomonas aeruginosa Isolated from Operations Enduring Freedom and Iraqi Freedom Trauma Patients. [1st place in Resident Research Podium competition]


Tribble DR, Ganesan A, Mende K, Stewart L. Blast Trauma Wound Infection Epidemiology and Microbiology.

2018 ASM Microbe. 7-11 June 2018, Atlanta, GA

2018 Annual Meeting of the Texas Infectious Diseases Society, 8-10 June 2018, Lost Pines, TX.

* Pomerantz HS, Beckius ML, Blyth DM, Tribble DR, Mende K. Antifungal Activity of Cerium Nitrate against Mold Isolates Associated with Combat-Related Injuries Including Burns. [2nd place in competition podium presentation]

Military Health System Research Symposium, 20-23 August 2018, Kissimmee, FL.


Stanbo J, Reinhart A, Lipinski M, Watters C, Demons ST, Tyner SD, Simons MP, Mende K, Tribble DR. Antagonism of ESKAPE Pathogens by Enterococcus Isolates from the Multidrug-Resistant and other Virulent Organisms (MDR/VO) Trauma Infections Initiative. [Poster #216]


U.S. Army / U.S. Air Force American College of Physicians Chapter Meeting, 6-8 September 2018, Lackland Air Force Base


Blyth D. Lessons from the Field: An update from the Trauma Infectious Disease Outcomes Study.

ID Week, A Joint Meeting of IDSA, SHEA, HIVMA, and PIDS, 3-7 October 2018, San Francisco, CA.

* Schall SE, Li P, Merritt T, Carson ML, Whitman TJ, Petfield JL, Tribble DR, Blyth DM. Clinical Characteristics of Military Trauma Patients with Clostridium difficile Infections. [Poster #483]


* Pomerantz HS, Beckius ML, Blyth DM, Akers KS, Tribble DR, Mende K. Antifungal Activity of Cerium Nitrate Against Mold Isolates Associated with Combat-Related Injuries Including Burns. [Poster #1376]


* Presentation/publication from a Graduate Medical Education-supported analysis.
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