Phage Therapy in Military Medicine: Concept and Preliminary in-vivo Results

Daniel Gelman
The Hebrew University of Jerusalem, Israel
Advisors: Dr. Ronen Hazan and Dr. Shaul Beyth

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Disclosures

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Learning Objectives

At the conclusion of this activity, the participant will be able to:

1. Identify the impending threat of antibiotic resistance in general and specifically in military medicine.
2. Explain the advantages of Bacteriophage Therapy in response to the threat of antibiotic resistance.
3. Describe the evolution of Bacteriophage Therapy until now.
4. Describe the potential future use of Bacteriophage Therapy as discovered in our research.
Obtaining CME/CE Credit

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http://amsus.cds.pesgce.com
Infectious Diseases in Military Medicine
The Decrease in Casualties of War

- Hygiene
- Vaccines
- Antibiotics
"...There may be a danger, though, in under-dosage. It is not difficult to make microbes resistant to penicillin..."
Antibiotic Resistance
Antibiotic Resistance

Increase in antibiotic resistance among pathogens from 1990-2002.

A decreasing number of novel antibiotics approved for clinical use.

Source: Clinical Microbiol Infect 2004; 10 (Suppl. 4): 1-9

Source: Nat Biotech 2006 24: 1521
Antibiotic Resistance – Military Medicine

In Harm’s Way: Infections in Deployed American Military Forces

Naomi E. Aronson,¹ John W. Sanders,² and Kimberly A. Moran³

¹Infectious Diseases Division, Uniformed Services University of the Health Sciences, and ²Infectious Diseases Service, National Naval Medical Center, Bethesda, Maryland; and ³Infectious Diseases Service, Walter Reed Army Medical Center, Washington, D.C.
Antibiotic Resistance – Military Medicine

![Graph showing susceptibility % of Acinetobacter spp. against various antibiotics from 2002 to 2005. The antibiotics include Amikacin, Amp/Sulbactam, Cefepine, Ceftazidine, Ciprofloxacin, Gentamicin, Imipenem, Levofloxacin, Pip/Tazo, and Tobramycin. The graph includes data for 2002, 2003, 2004, and 2005.]
“…A female Washoe County resident in her 70s developed septic shock and died…”
“…the isolate was resistant to 26 antibiotics, including all aminoglycosides and polymyxins tested, and intermediately resistant to tigecycline…”

2016: A Nevada woman dies of a superbug resistant to every available antibiotic in the US

But Recently...
Mode of Action

Bacteriophages work exactly where antibiotics fail

Easy Isolation  Combat Resistance  Biofilm Eradication

Khalifa et al. AEM. 2015
Bacteriophage Therapy
A brief Historical Review

• 1915 – The first report of bacterial viruses
• 1917 – Beginning of the Bacteriophage era (World War I)
• 1919 – The first clinical application of bacteriophages, Paris
• 1923 – Establishment of the Eliava Institute, Tbilisi, Georgia
"The therapeutic effects reported may or may not have been due to bacteriophage itself"

"...most of the work reported has been done without adequate controls, and..."
The Fall of Phage Therapy

1. Due to the striking success of antibiotics

2. Due to the unknown nature of phages relative to the simplicity of antibiotics

3. Due to historical and psychological reasons
The Eliava Institute
Phage therapy gets revitalized

The rise of antibiotic resistance rekindles interest in a century-old virus treatment.

2014
Bacteriophage Therapy Research Rises Again

PubMed Publications: Phage Therapy

Recent Clinical Applications

Tom woke up from his coma
Development and use of personalized bacteriophage-based therapeutic cocktails to treat a patient with a disseminated resistant *Acinetobacter baumannii* infection

Running head: Successful Bacteriophage Therapy

March 2016

Robert T. Schooley, M.D.1, Biswajit Biswas, Ph.D.2,3, Jason J. Gill, Ph.D.4,5, Adriana Hernandez-Morales, M.S.6, Jacob Lancaster5, Lauren Lessor5, Jeremy J. Barr, Ph.D.7,15, Sharon L. Reed, M.D.1,8, Forest Rohwer, Ph.D.7, Sean Benler, Ph.D.7, Anca M. Segall, Ph.D.7, Randy Taplitz, M.D.1, Davey M. Smith, M.D., M.A.S.1, Kim Kerr, M.D.1, Monika Kumaraswamy, M.D.1, Victor Nizet, M.D.9,10, Leo Lin, Ph.D.9, Melanie D. McCauley, M.D.1, Steffanie A. Strathdee, Ph.D.1, Constance A. Benson, M.D.1, Robert K. Pope, Ph. D.11, Brian M. Leroux11, Andrew C. Picel, M.D.12, Alfred J. Mateczun, M.D.2, Katherine E. Cilwa, Ph.D.14, James M. Regeimbal, Ph.D.2, Luis A. Estrella, Ph.D.2, David M. Wolfe, Ph.D.2, Matthew S. Henry, M.S.2,3, Javier Quinones, M.S.2,3, Scott Salka13, Kimberly A. Bishop-Lilly, Ph.D.2,3, Ry Young, Ph.D.5,6, Theron Hamilton, Ph.D.2
Use of bacteriophages in the treatment of colistin-only-sensitive *Pseudomonas aeruginosa* septicaemia in a patient with acute kidney injury—a case report

Serge Jennes¹, Maia Merabishvili², Patrick Soentjens³, Kim Win Pang³, Thomas Rose¹, Elkana Keersebilck¹, Olivier Soete¹, Pierre-Michel François¹, Simona Teodorescu¹, Gunther Verween², Gilbert Verbeken², Daniel De Vos² and Jean-Paul Pirnay⁴

Queen Astrid Military Hospital, Brussels, Belgium. November 2016
• 61-year-old man, septicaemia with colistin-only-sensitive *P. aeruginosa*

- Blood cultures turned negative
- CRP levels dropped
- Fever disappeared
- Kidney function recovered
- No unexpected adverse events
Military Uses of Phage Therapy
Military Uses of Phage Therapy

From Battlefront to Homefront

- Wound Infections
- Burn Infections
- Foodborne Illnesses
- Dental Infections
- Preventative Tool for Biological Warfare
- Secondary Osteomyelitis
Military Uses of Phage Therapy

Bacteriophages can be applied in every infectious condition in which the main pathogens are known
The Soviet Experience
Phage Therapy in Military Medicine: Wound Infections

Phage Therapy in Military Medicine: Foodborne Illnesses

Tbilisi, Georgia 1963–1964
30,769 children (6 months to 7 years old)

Of these, children on one side of the streets (17,044 children) were given *Shigella* phages orally (once every 7 days), and the children on the other side of the streets (13,725) did not receive phages.

Alexander Sulakvelidze et al. AAC. 2001;45:649-659
1307 patients with suppurative bacterial infections caused by multidrug-resistant bacteria of different species were treated with specific bacteriophages (BP). BP therapy was highly effective; full recovery was noted in 1123 cases (85.9%).

…septicemia, purulent otitis media, peritonitis, pyogenic arthritis and myositis, Osteomyelitis of the long bones, pyogenic infections of burns, purulent mastitis…
Modern Research
Phage Therapy in Military Medicine: 
**Burn Infections**

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**September 2015:** 
**Bacteriophage Therapy Re-enter Clinical Trials**

- The first randomized and monitored phase I/II single-blind trial, evaluating the tolerance and effectiveness of phage therapy in fighting sensitive antibiotic-resistant infections
- conducted in 11 major burns units in France, Switzerland and Belgium
Phage Therapy in Military Medicine:
Wound Infections

Personalized Therapeutic Cocktail of Wild Environmental Phages Rescues Mice from *Acinetobacter baumannii* Wound Infections

James M. Regeimbal, Anna C. Jacobs, Brendan W. Corey, Matthew S. Henry, Mitchell G. Thompson, Rebecca L. Pavlicek, Javier Quinones, Ryan M. Hannah, Meron Ghebremedhin, Nicole J. Crane, Daniel V. Zurawski, Nimfa C. Teneza-Mora, Biswajit Biswas, and Eric R. Hall
Phage Therapy in Military Medicine: Osteomyelitis

Phage therapy of staphylococcal chronic osteomyelitis in experimental animal model

Chandan Kishor, Raghvendra Raman Mishra, Shyam K. Saraf, Mohan Kumar, Arvind K. Srivastav, and Gopal Nath
Phage Therapy in Military Medicine: Biological Warfare Prevention

<table>
<thead>
<tr>
<th>Category</th>
<th>Disease</th>
<th>Agent</th>
</tr>
</thead>
<tbody>
<tr>
<td>Anthrax</td>
<td>Bacillus anthracis</td>
<td></td>
</tr>
<tr>
<td>Botulism</td>
<td>Clostridium botulinum toxin</td>
<td></td>
</tr>
<tr>
<td>Plague</td>
<td>Yersinia pestis</td>
<td></td>
</tr>
<tr>
<td>Smallpox</td>
<td>Variola major</td>
<td></td>
</tr>
<tr>
<td>Tularemia</td>
<td>Francisella tularensis</td>
<td></td>
</tr>
<tr>
<td>Viral hemorrhagic fevers</td>
<td>Filoviruses (e.g. Ebola, Marburg) and arenaviruses (e.g. Lassa, Machupo)</td>
<td></td>
</tr>
</tbody>
</table>
Phage Therapy in Military Medicine: Biological Warfare Prevention
Bioterrorism Prevention and Therapy

Foodborne and Waterborne Diseases

Osteomyelitis

Burn injuries infections

Wound Site Infections
Mouse Peritonitis Model

- Female, 4-6-week-old, outbred ICR mice
- *E. faecalis V583 - 10*LD$_{50}$
- SRFE 50%
Bacterial Target

VANCOMYCIN-RESISTANT ENTEROCOCCUS (VRE)

- 20,000 drug-resistant enterococcus infections
- 1,300 deaths from drug-resistant enterococcus infections
- 66,000 enterococcus infections per year

Some enterococcus strains are resistant to vancomycin, leaving few or no treatment options.

Threat level: serious

This bacteria is a serious concern and requires prompt and sustained action to ensure the problem does not grow.
- Untreated

- **Standard Antibiotic Regimen**
  Ampicillin (12.5 mg/Kg)

- **Phage Therapy**
  EFDG1-EFLK1 Cocktail (MOI=0.025)

- **Combined Phage – Antibiotic Treatment**
Survival Rates – 1 Hour Delay

- Combined Treatment (N=15)
- Phages (N=15)
- Antibiotics (N=15)
- Untreated (N=10)

Gelman et al. (In Preparation)
Survival Rates – 6 Hours Delay

![Graph showing survival rates for different treatments with time (hours) on the x-axis and survival rate on the y-axis. The treatments include Combined Treatment (N=5), Phages (N=5), Antibiotics (N=5), and Untreated (N=10).]
Clinical Scoring

- Level of consciousness
- Activity
- Eyes
- Respiration Rate
- Respiration Quality
- Provoked Reaction
Clinical Scoring

Phage

Antibiotics

Combined
Clinical Scoring - Survivors

Gelman et al. (In Preparation)
Complete Blood Count

Gelman et al. (In Preparation)
Bacterial Load

[Graph showing bacterial load with labels for CFU/g and categories for Control, AB, Phage, and Phage & AB.]

Gelman et al. (In Preparation)
Bacteriophage Prevalence

Combined therapy reduces bacteriophage prevalence

Gelman et al. (In Preparation)
There is a positive correlation between the bacterial load and the prevalence of bacteriophages.
Key Points

- Phage Therapy successfully resolved severe septic peritonitis
- Bacteriophages are appropriate for use as a “single shot” medication
- Phage – Antibiotic Combination allows a rapid elimination of the phage
- Direct Influence on the Immune Response?
- Anti-Phage Immunization?
Phage Therapy has a very high potential as an alternative and as an addition to antibiotics.

Phage Therapy can be applied in many aspects of military medicine.

The integration of Phage Therapy to the standard of care in the military medicine is an important and a required step towards preventing the arrival of the post antibiotic era.
Thank you.

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Contact Us: Daniel.Gelman@mail.huji.ac.il
Questions?