GEIS Respiratory Disease Surveillance Newsletter

DoD Center for Deployment Health Research
Naval Health Research Center, San Diego

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Sponsored by the DoD Global Emerging Infections System (GEIS), the Naval Health Research Center (NHRC) collaborates with numerous federal and non-federal institutions to conduct surveillance for several respiratory pathogens, including adenovirus, influenza, respiratory syncytial virus (RSV), parainfluenza, *Streptococcus pyogenes*, and invasive *Streptococcus pneumoniae*. Please visit our website at http://www.nhrc.navy.mil/geis for additional information.

Current Study Updates

Pneumococcal Vaccine Trial

In collaboration with Centers for Disease Control and Prevention, the Mayo Clinic and Foundation, and Wyeth-Lederle vaccines, NHRC and 4 recruit training sites are conducting a double blind, placebo-controlled trial of a 23-valent pneumococcal vaccine to assess the vaccine's clinical effectiveness among the military trainee population. The study is currently underway at Fort Jackson, Fort Leonard Wood, NRTC Great Lakes, and MCRD Parris Island. The study will eventually enroll over 191,000 trainees. The participants will be actively followed during recruit training for pneumonia and passively followed (by tracking inpatient and outpatient military medical databases) for pneumonia and acute respiratory disease after recruit training until the end of the study period (up to 3 years) for pneumonia and acute respiratory disease. To date, study site personnel have enrolled more than 134,000 recruits with an enrollment rate of approximately 70 percent.

Evaluation of PCR Testing Using Room Temperature Specimens

One of the biggest challenges the Febrile Respiratory Illness (FRI) sites face is keeping viral specimens frozen at ultra-low temperatures for long periods before and during shipping. A sub-study at Fort Jackson has begun to evaluate the performance of room temperature specimens in comparison with frozen viral cultures. The study, in collaboration with researchers at the Armed Forces Institute of Pathology, uses PCR techniques to test for adenovirus and influenza. If these tests prove successful, the specimen collection portion of FRI surveillance could be simplified and the surveillance expanded to regions where freezing specimens is not feasible. The first specimens collected during the 2001-2002 flu season yielded promising results. However, further testing is needed and specimen collection will resume in December 2002 for a 4-month period.

Febrile Respiratory Illness (FRI) in a Mexican population

In collaboration with the Mexican Institute of Public Health Services, FRI surveillance at a community clinic in Ensenada, Mexico continues. Lab results for the first specimens collected indicate that both flu and adenovirus are present in the population.

We have recently confirmed an early influenza B isolate for the 2002-2003 flu season. The sample was collected in Ensenada, Mexico in late September. Subtyping is underway.

Association Between Adenovirus Infections Among Military Personnel and the Development of Obesity

This case control study, which began in October 2001, investigates if an association exists between weight control problems (obesity) and adenovirus-36 exposure in a population of active-duty Navy personnel. The study, which is being conducted at the Clinical Trials Center and at other military commands in San Diego, is currently enrolling cases (individuals with a Body Mass Index (BMI) of >30) and controls (BMI <25).

New Studies

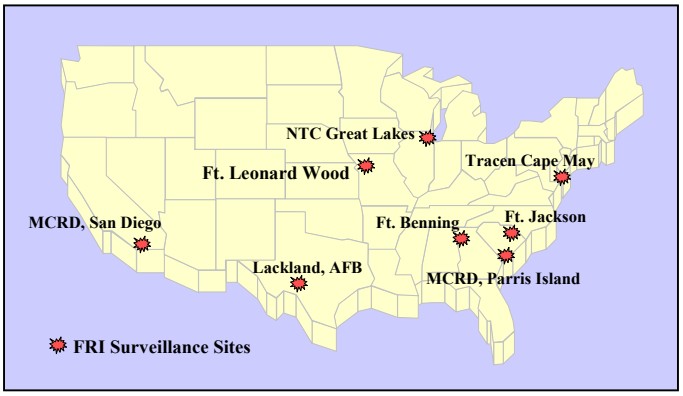
Laboratory Based Surveillance for Febrile Respiratory Illness Aboard Floating Platforms

Laboratory supported surveillance for respiratory pathogens aboard U.S. military ships and submarines in the Pacific Rim has been sparse. Viral illness outbreaks have repeatedly occurred in these settings. This study, currently being initiated, will examine respiratory illness aboard a variety of floating platforms, as well as submarines, home based in the San Diego area. Viral throat culture specimens will be obtained from sailors presenting with FRI to determine FRI etiology aboard ships, both at home and while deployed.

Continued from page 1

Real-time Diagnostic Capabilities on Aircraft Carriers – Real-time PCR techniques for influenza and adenovirus detection are currently being optimized for use on aircraft carriers. This capability will utilize existing RAPID equipment currently found aboard Aircraft Carriers. Routine use of this equipment while testing for respiratory pathogens, will help to maintain technical competency.

Febrile Respiratory Illness Surveillance



Current Progress – Febrile Respiratory Illness surveillance is currently being conducted at eight military training sites. From June 2001 to June 2002, 2422 specimens were collected from trainees presenting with symptoms matching the definition of FRI. Specimens are tested for adenovirus, influenza A and B, respiratory syncytial virus (RSV), and parainfluenza 1, 2 and 3. The specific trends observed for each of these viruses are described in the following sections.

FRI Specimens Tested per Site
June 1998 – September 2002

SITE	SPECIMENS TESTED
Fort Benning	1545
Fort Jackson	3057
Fort Leonard Wood	1418
NRTC, Great Lakes	1388
MCRD, San Diego	662
MCRD, Parris Island	323
CGTC Cape May	510
Lackland AFB	957
Total	9860

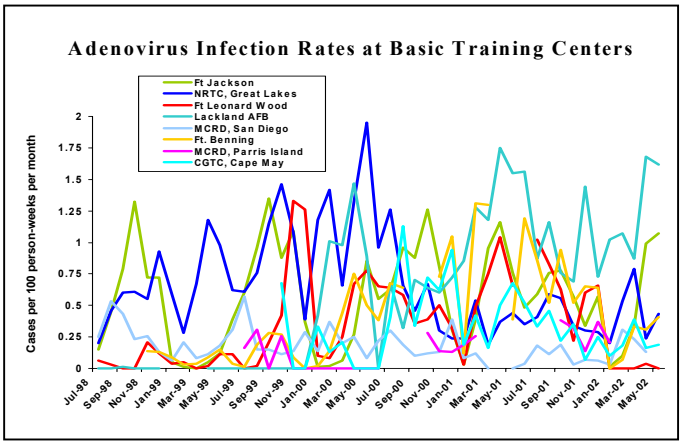
Geographic Trends – Since January 1, 2002, five of the eight surveillance sites have reported FRI rates above the epidemic threshold, which is defined as 1.5 cases per 100 trainees per week. The distribution of viral test results by site demonstrates that adenovirus is responsible for the majority of FRI at all locations.

Temporal Trends – During the first eight months of 2002, FRI rates showed no discernable seasonal trends, as epidemics occurred at various times throughout this period.

Adenovirus

Current Progress – In the absence of vaccine, adenovirus remains the leading cause of FRI among trainees. More than 61% of the 9860 throat cultures collected for the FRI study between June 1998 and June 2002, and 1482 (61.2%) of the 2422 collected between June 2001 and June 2002 tested positive for adenovirus, Approximately 97% of all adenovirus isolates collected to date have been type 4.

Geographic Trends – From January 1, 2002 to May 31, 2002, the amount of FRI morbidity caused by adenovirus remained high at all sites, ranging from 41.1% at Fort Jackson, to 79.5% at Lackland AFB. Please see accompanying charts for adenovirus infection rates by site.



Influenza

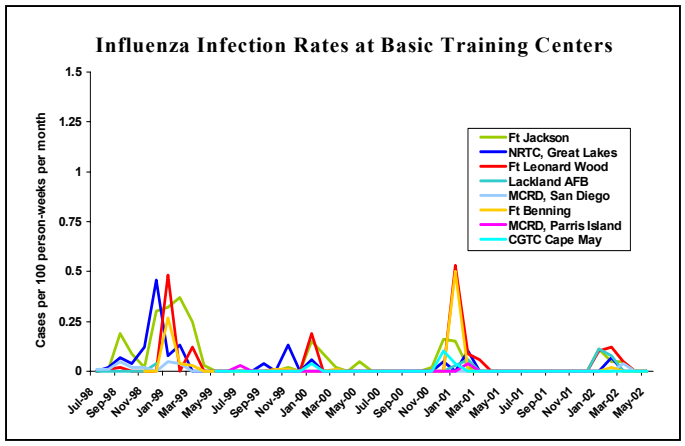
Current Progress – Of the 2422 FRI specimens collected between June 2001 and June 2002, 47 (1.9%) have tested positive for influenza. Of all the specimens collected from the start of this study, 428 (4.3%) out of 9860 have been positive for influenza, with 3.2% identified as type A and 1.1% as type B. Ill trainees who were not vaccinated against influenza were more than 5 times more likely to be influenza-positive (OR= 5.7, 95% CI, 4.5-7.1) than those who did receive the vaccine.

Geographic Trends – Our surveillance data demonstrate that morbidity among trainees caused by

Continued from page 2

infection with influenza A or B varies by training site location. Please see accompanying chart for influenza infection rates by basic training site location.

Temporal Trends – Influenza activity occurred only during the winter months in 2001-2002, during the typical "flu-season". Overall, rates were somewhat low in comparison with the 1998-1999 and 2000-2001 flu seasons.



Other FRI Study Pathogens

Of the 9860 throat cultures tested thus far under the FRI study, 21 (0.2%) have been positive for RSV and 95 (1.0%) have been positive for parainfluenza 1, 2, or 3. Of the 2422 specimens collected from June 2001 to June 2002, 3 (0.1%) have been positive for parainfluenza 1, 2, or 3; none of these samples were positive RSV.

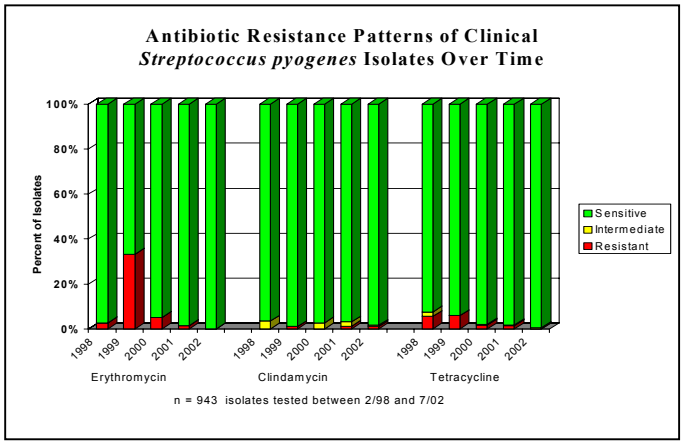
Streptococcus pyogenes Surveillance

Current Progress – *Streptococcus pyogenes* (Group A streptococcus) continues to be a threat to the health of military trainees. From the start of this surveillance in February 1998 through July 2002, 942 (389 in the last year) clinical isolates were collected from trainees at 8 military sites and tested for antibiotic resistance here at NHRC.

S. pyogenes Isolates Received per Site February 1998 to February 2002

SITE	ISOLATES RECEIVED
NRTC, Great Lakes	277
MCRD, Parris Island	424
MCRD, San Diego	12
Fort Jackson	4
Fort Knox	23
Fort Leonard Wood	53
Fort Sill	46
Lackland AFB	103
Total	942

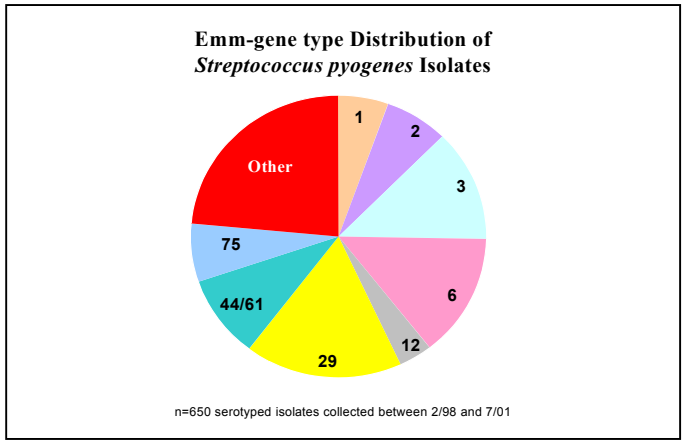
Antibiotic Resistance – Among the specimens tested, *S. pyogenes* maintains 100% susceptibility to the antibiotics penicillin, levofloxacin, and vancomycin. Fifty (5.3%) of the 943 isolates collected exhibited full or partial resistance to erythromycin, 43 (4.6%) to tetracycline, and 24 (2.5%) to clindamycin. Ten (1.1%) of the isolates were resistant to both erythromycin and tetracyline. Isolates from female trainees showed a similar proportion of erythromycin resistance as compared to male trainees (7.5% and 5.0%, respectively). Temporal trends in antibiotic resistance among *S. pyogenes* isolates collected to date demonstrate no discernible pattern, as shown in the following chart.



Emm-gene Types – As of September 2002, 650 *Streptococcus pyogenes* specimens had been emm-gene typed. Among these, the most common emm-gene types among military trainees were 29 (17.2%), 6 (14.0%), 3 (12.9%), 44/61 (9.4%), 75 (6.8%), 2 (6.9%), 1 (5.5%), and 12 (3.7%). These eight emm-gene types made up more than 76% of all the typed isolates.

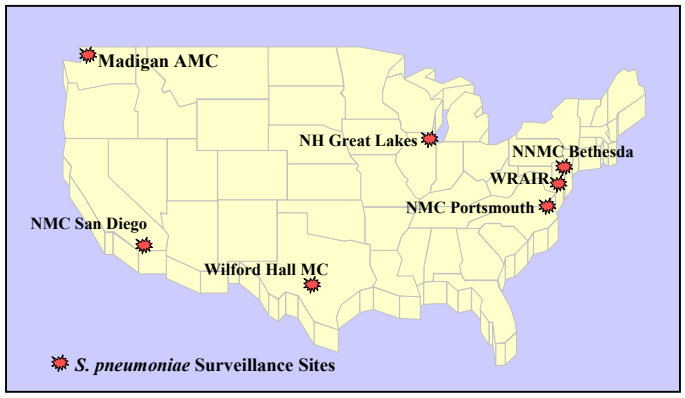
Continued from page 3

Resistance by Emm-gene type – Erythromycin resistance varied by emm-gene type. Type 75 exhibited the most erythromycin resistance of all emm-gene typed isolates, with 64% of type 75 isolates demonstrating full resistance. Seventy percent (31/44) of Type 75 isolates came from trainees at Lackland AFB, TX.



Geographic Trends – *S. pyogenes* isolates from military trainees continue to maintain high susceptibility to many commonly prescribed antibiotics including tetracycline, erythromycin and clindamycin, and 100% susceptibility to penicillin, levofloxacin, and vancomycin. However, we continue to observe an unequal geographic distribution of erythromycin resistance at the sites. Resistance to erythromycin differed between sites; the highest resistance rate was at Lackland AFB (29.1%). All of the erythromycin resistant isolates from Lackland AFB, were fully resistant.

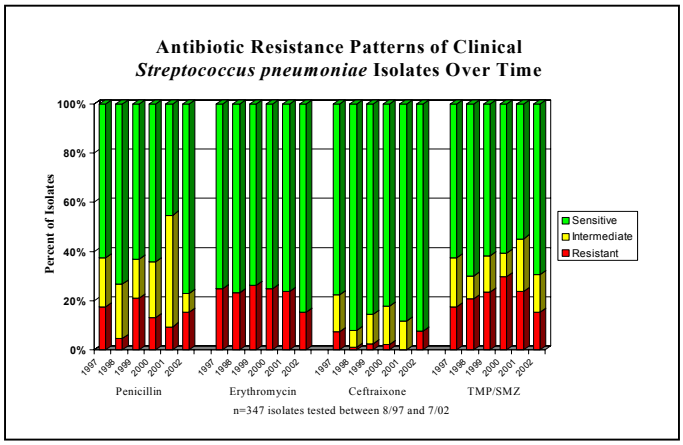
Streptococcus pneumoniae Surveillance



Current Progress – Invasive clinical isolates continue to be collected from 7 military medical centers across the nation. Three hundred and forty-seven isolates have been collected thus far, 42 within the last year.

Geographic Trends – Among sites with an adequate number of specimens, penicillin resistance was relatively stable, ranging from 33% at Madigan AMC to 47% at Walter Reed AIR.

Antibiotic Resistance – One-hundred and twenty-three (35.4%) of the 347 isolates collected demonstrated full or partial resistance to penicillin, and 79 isolates (22.8%) exhibited multiple resistance to three or more antibiotics. Penicillin resistance levels among males (37.4% resistant) and females (32.6% resistant) were similar. As shown in the graph below, temporal trends in antibiotic resistance among *S. pneumoniae* isolates collected to date do not demonstrate a discernible pattern.



Resistance by Serotype – Penicillin resistance differed by serotype, with types 19 (54.3%), 9 (50%), and 23 (37.5%) demonstrating the most resistance. These three serotypes accounted for more than 59% of all penicillin resistance among the serotyped isolates, though they only comprised 37% of the total number of serotyped isolates.

S. pneumoniae Isolates Received per Site
August 1997 to July 2002

SITE	ISOLATES RECEIVED
NRTC Great Lakes	4
NMC, San Diego	101
NMC, Portsmouth	2
NNMC, Bethesda	12
Walter Reed AIR	36
Wilford Hall MC	80
Madigan AMC	112
Total	347

Bordetella pertussis Surveillance

Current Progress – Surveillance of *Bordetella pertussis* is currently underway at four military recruit training sites: MCRD - San Diego, Ft. Benning, Ft. Leonard

Continued from page 4

Wood, and NRTC Great Lakes. Site personnel have enrolled 257 subjects to date. Of these, 147 specimen sets are complete and have been tested using 3 different methods. These specimens were collected from recruits meeting the case definition for pertussis, which is more than seven days of coughing with symptoms consistent with a respiratory infection. Preliminary results show that one (0.6%) of the specimens has tested positive by culture, 9 (6.1%) were positive by PCR, and 3 (2%) tested positive by serology (seroconversion).

***B. pertussis* Specimens Received per Site June 2000 to February 2002**

SITE	SPECIMENS RECEIVED
MCRD San Diego	156
Fort Leonard Wood	4
Fort Benning	3
NRTC Great Lakes	19
Total	257

Expanding Projects in Molecular Epidemiology

Sequencing of Adenovirus Vaccine-Breakthrough Samples

Through respiratory surveillance, NHRC has discovered that from 1996 to 1999 several individuals who received the adenovirus vaccine subsequently became ill with adenovirus 4 or 7 infection. Production of the vaccine was discontinued in 1996, but the DoD has recently contracted for resumed production, making knowledge of the antigenicity of circulating pathogenic strains crucial. Research by the CDC and the California Department of Health Services has shown significant variation in the epitope-coding portions of the hexon gene. NHRC, in collaboration with Brooks Air Force Base and the California Department of Health Services, is sequencing and phylogenetically analyzing the hexon gene of each breakthrough strain to determine if genetic drift contributed to these break-through infections.

Multiplex-PCR Subtyping of Adenovirus

NHRC is currently developing a multiplex-PCR assay to largely replace microneutralization as its current adenovirus typing method. This would allow us to become less dependent on the research that the California Department of Health Services, Berkeley, CA) generously provides us with.

Retrospective Molecular Surveillance for Human Metapneumovirus and Rhinovirus

A new respiratory virus, human metapneumovirus (HMPV), was discovered in June of 2001. The clinical symptoms of HMPV are similar to those of human respiratory syncytial virus and include mild respiratory problems, cough, bronchiolitis, and pneumonia, often presented with high fever, myalgia, and vomiting. Human rhinovirus, on the other hand, is a known cause of the common cold, but its relationship to FRI has yet to be fully elucidated. NHRC is performing PCR and RT-PCR on previously undiagnosed isolates from FRI surveillance to determine if human metapneumovirus or rhinovirus can be detected.

Triangulation Identification for Genetic Evaluation of Risks

Ibis Therapeutics is developing a high-throughput, single-pass process for the simultaneous detection of all infective threat agents, named Triangulation Identification for Genetic Evaluation of Risks (TIGER). The process uses PCR primers to amplify housekeeping genes that are conserved across species. With the species-specific molecular weight of amplified gene targets known for each pathogen, the samples can be identified with highly sensitive mass spectrometry. Using adenovirus samples located at NHRC, we have already implemented this technology in a successful proof of concept exercise. Additional respiratory pathogens will be utilized to further develop this methodology.

Epidemic Outbreak Surveillance

Researchers from the Massachusetts Institute of Technology, Johns Hopkins University, and the Naval Research Laboratory have begun a large project at Lackland AFB to implement an early detection and warning system for infectious disease outbreaks. This Epidemic Outbreak Surveillance (EOS) program will combine new diagnostic technology (e.g., microarrays) with bioinformatics to provide rapid diagnoses and public health information to patients, physicians, and public health officials. NHRC will provide archived respiratory pathogens, and help develop and validate these new tests.

Upcoming Protocols

Surveillance for viral respiratory pathogens among Venezuelan military and civilian personnel – NHRC may begin collecting viral cultures from both military and civilian populations in Venezuela. This surveillance will be similar to our current FRI study, and is scheduled to begin early next year.

Surveillance for viral respiratory pathogens in Singapore – NHRC may begin collecting viral cultures from the civilian population of Singapore. This surveillance will also be similar to our current FRI study.

Center News

Presentations or Posters at Recent Conferences

8th Annual Recruit and Trainee Healthcare Symposium, April 15-18, 2002, Towson, MD.

- Russell K. Surveillance for viral respiratory pathogens among US military basic trainees.

San Diego Biostatistics and Epidemiology Research Exchange, May 3, 2002, La Jolla, CA.

- Gunnill R, et al. Characteristics of military recruits participating in a double-blind placebo-controlled vaccine trial.
- Cameon C, et al. National Department of Defense surveillance for *Streptococcus pneumoniae*: antibiotic resistance and serotype distribution analyses.

V International Symposium on Pneumococcal Illness. May 5-9, 2002, Anchorage, AL.

- Russell L, et al. Efficacy of the 23-valent pneumococcal vaccine among young healthy adults: double-blind placebo controlled clinical trial among military trainees at increased risk of respiratory disease.

National Syndromic Surveillance Conference. September 23-24, 2002, New York City, NY.

- Hawksworth A, et al. Using autoregressive modeling to augment the existing Department of Defense (DoD) febrile respiratory illness surveillance system at military training centers.
- Marsden-Haug N, et al. A comparison of military surveillance systems for early detection of naturally occurring and bioterrerism-based epidemics of febrile respiratory illness.

42nd Interscience Conference on Antimicrobial Agents and Chemotherapy, September 27-30, 2002, San Diego, CA.

- Hawksworth, et al. Recurrent epidemics of adenoviral respiratory illness among US military recruits in the absence of oral vaccines.

Upcoming Conferences

- **International Symposium on Respiratory Viral Infections**, December 5-8, 2002, Casa de Campo, La Romana, Dominican Republic.
- **Prevention 2003**, February 19-23, 2003, San Diego, CA.
- **The Forty-Third Navy Occupational Health And Preventive Medicine Workshop**, May 8-16, 2003, San Diego, CA.

Recently Submitted for Publication

- **National Department of Defense Surveillance Study of Group A Streptococcal Isolates; Antibiotic Resistance, and *emm* Gene Types Among Eight Basic Training Military Sites.** Barrozo CP, et al. *At Clin Infect Dis, November 2002.*

Please contact the newsletter staff with any comments or suggestions regarding the information in this issue.

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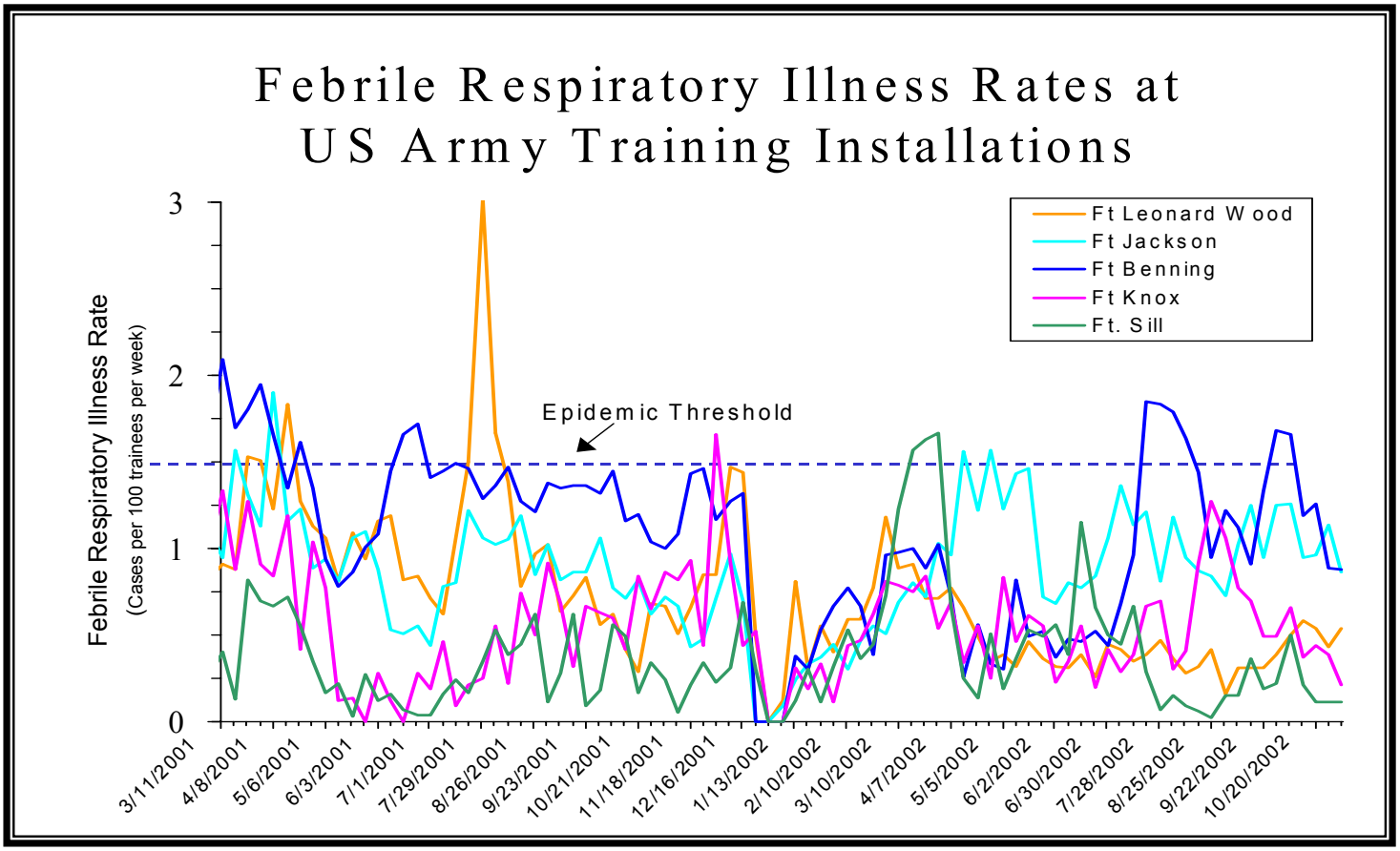
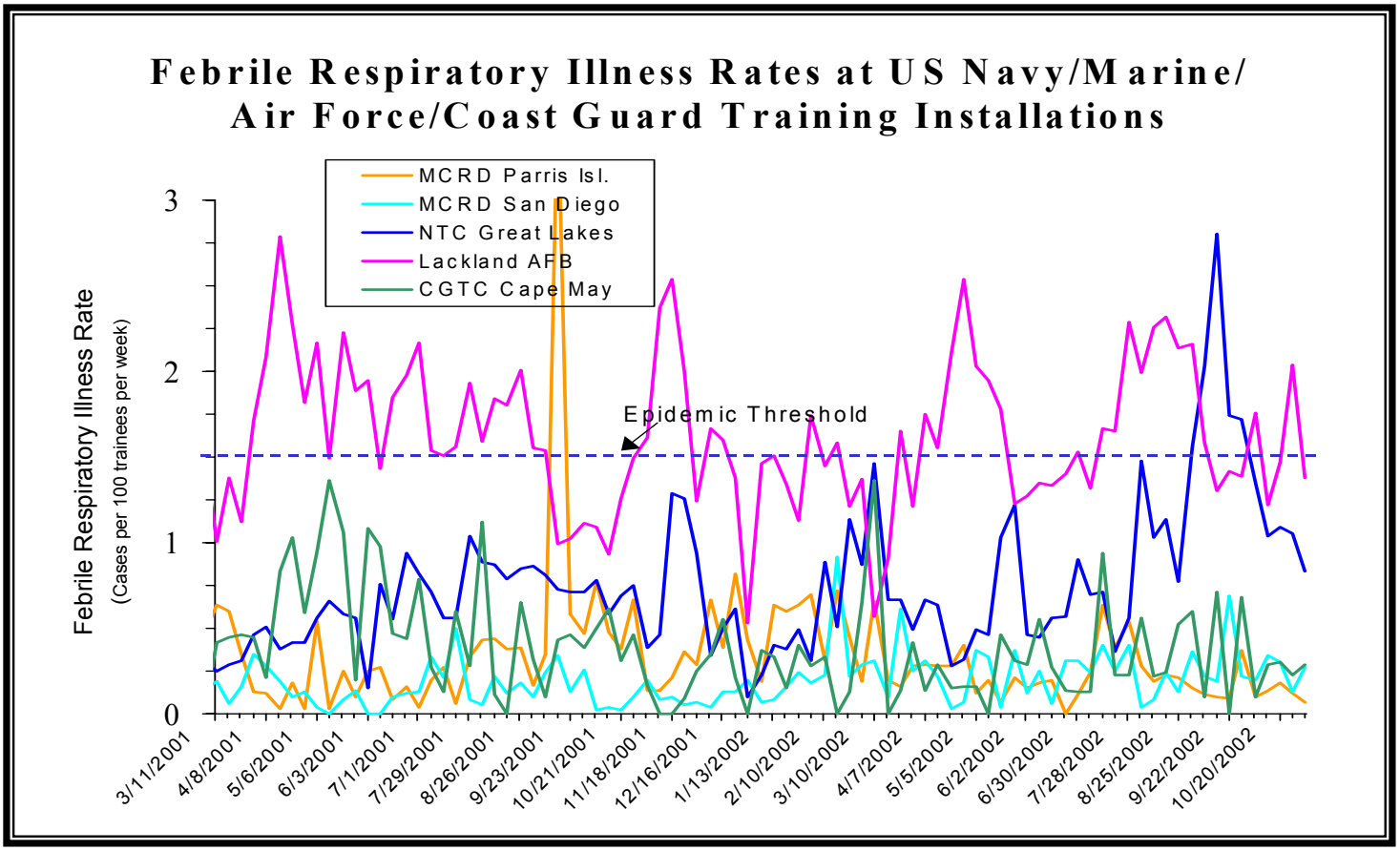
These studies would not be possible without the hard work and dedication to excellence of the staff at our collaborating institutions. Thank you!

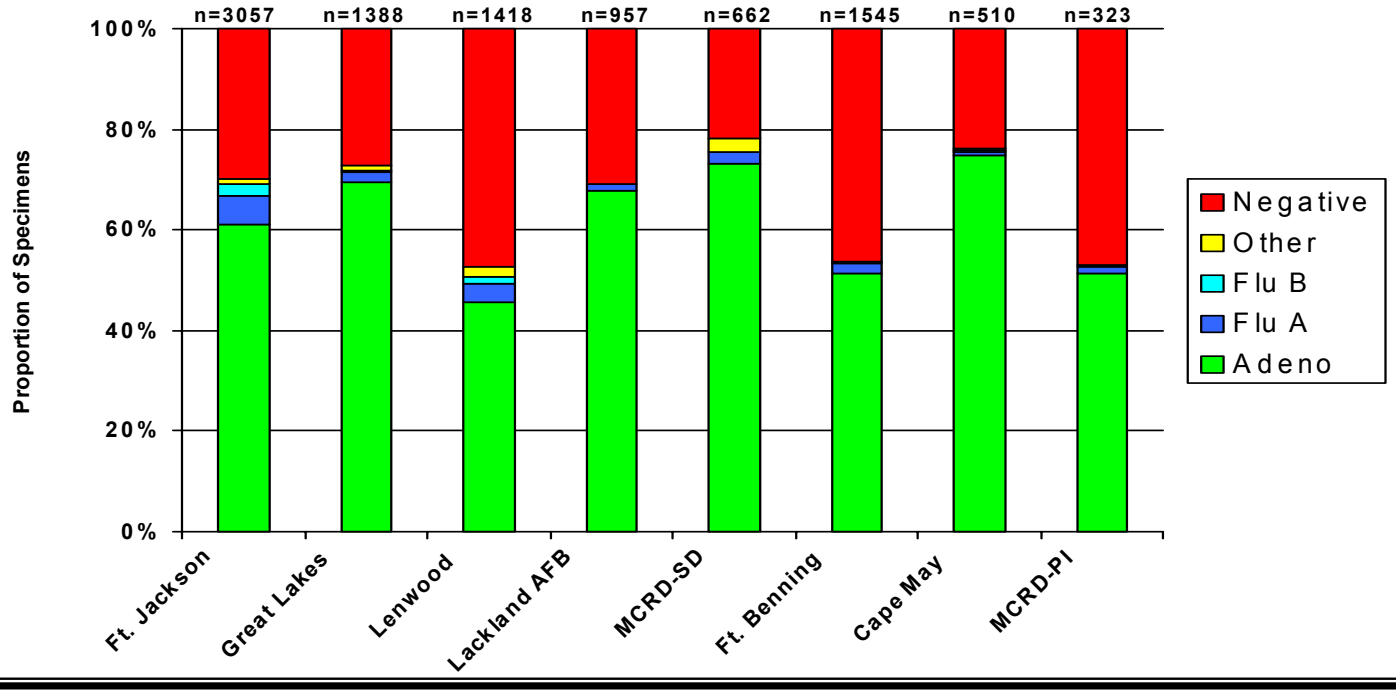

Febrile Respiratory Illness Rates at
US Army Training Installations
Febrile Respiratory Illness Rate
(Cases per 100 trainees per week)
Ft Leonard Wood
Ft Jackson
Ft Benning
Ft Knox
Ft. Sill
Epidemic Threshold
0
1
2
3
3/11/2001
4/8/2001
5/6/2001
6/3/2001
7/1/2001
7/29/2001
8/26/2001
9/23/2001
10/21/2001
11/18/2001
12/16/2001
1/13/2002
2/10/2002
3/10/2002
4/7/2002
5/5/2002
6/2/2002
6/30/2002
7/28/2002
8/25/2002
9/22/2002
10/20/2002


Febrile Respiratory Illness Rates at US Navy/Marine/
Air Force/Coast Guard Training Installations
Febrile Respiratory Illness Rate
(Cases per 100 trainees per week)
MCRD Parris Isl.
MCRD San Diego
NTC Great Lakes
Lackland AFB
CGTC Cape May
Epidemic Threshold
0
1
2
3
3/11/2001
4/8/2001
5/6/2001
6/3/2001
7/1/2001
7/29/2001
8/26/2001
9/23/2001
10/21/2001
11/18/2001
12/16/2001
1/13/2002
2/10/2002
3/10/2002
4/7/2002
5/5/2002
6/2/2002
6/30/2002
7/28/2002
8/25/2002
9/22/2002
10/20/2002

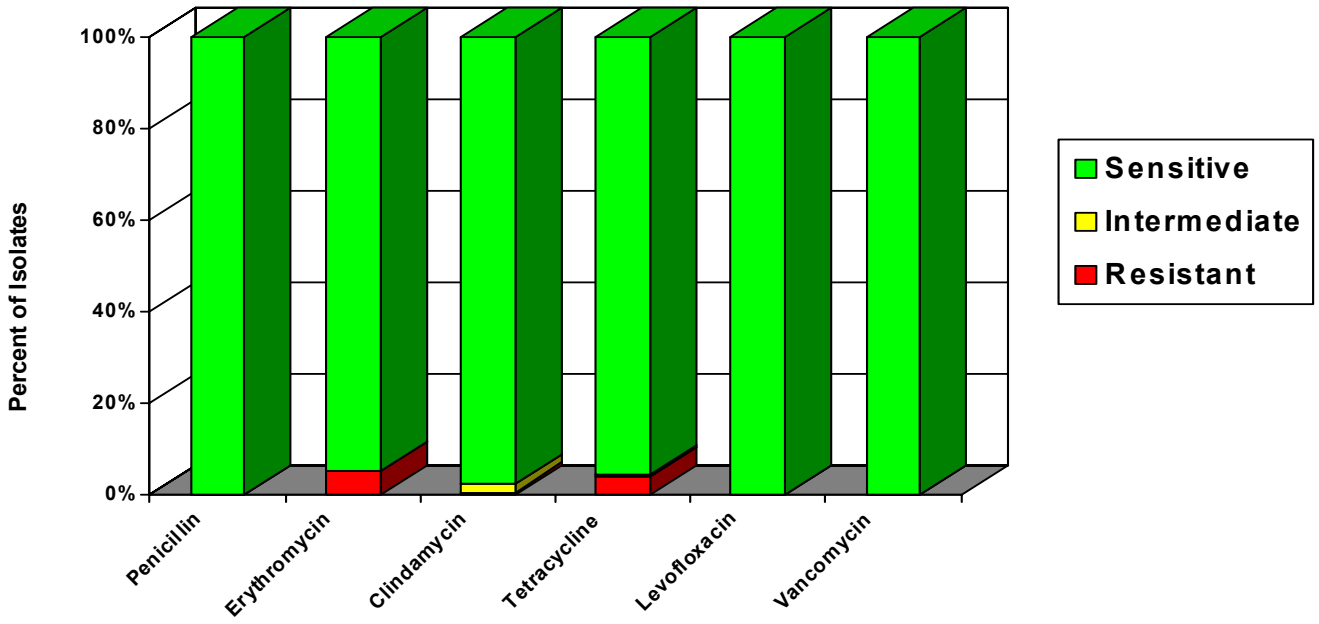
Distribution of Viral Test Results by Site

June 1998 - May 2002

n=9860



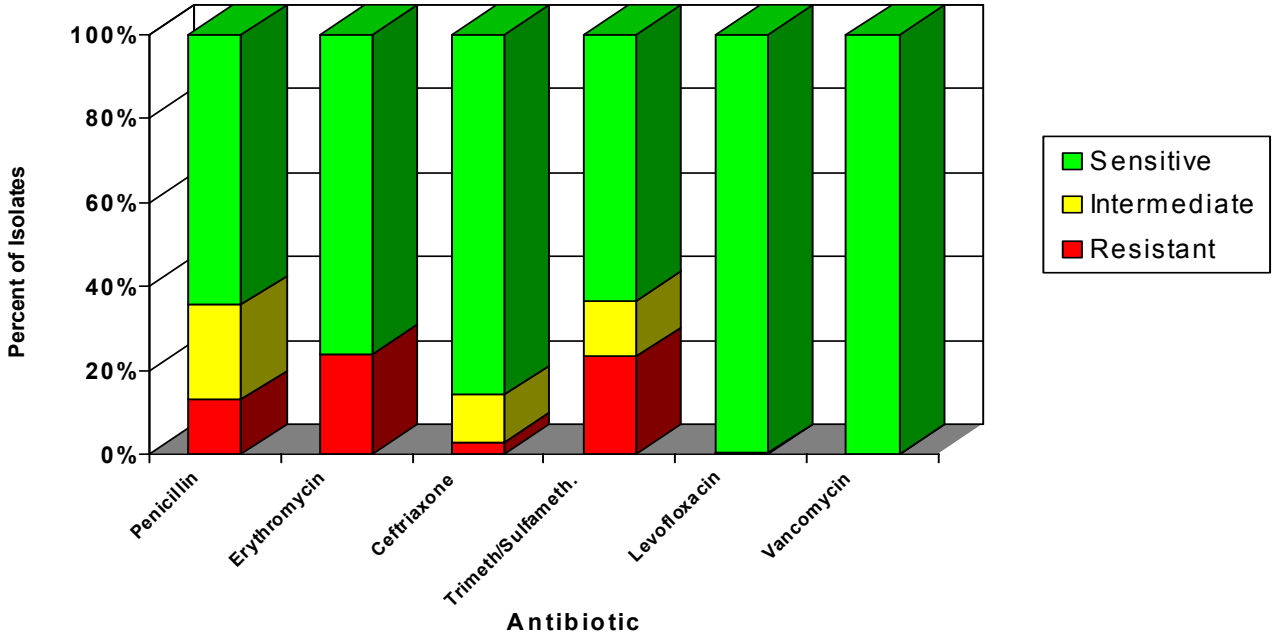
Antibiotic Resistance Patterns of Clinical *Streptococcus pyogenes* Isolates from Military Trainees



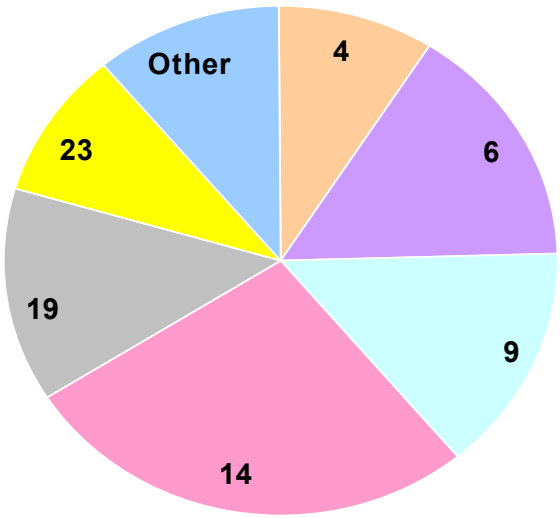
n=943 isolates tested between 2/98 and 7/02

Antibiotic Resistance Patterns of Sterile Site *Streptococcus pneumoniae* Isolates From Military Medical Facilities

n=347 isolates tested between 8/97 and 7/02



Serotype Distribution of Sterile Site *Streptococcus pneumoniae* Isolates



n=258 serotyped isolates collected between 8/97 and 7/02