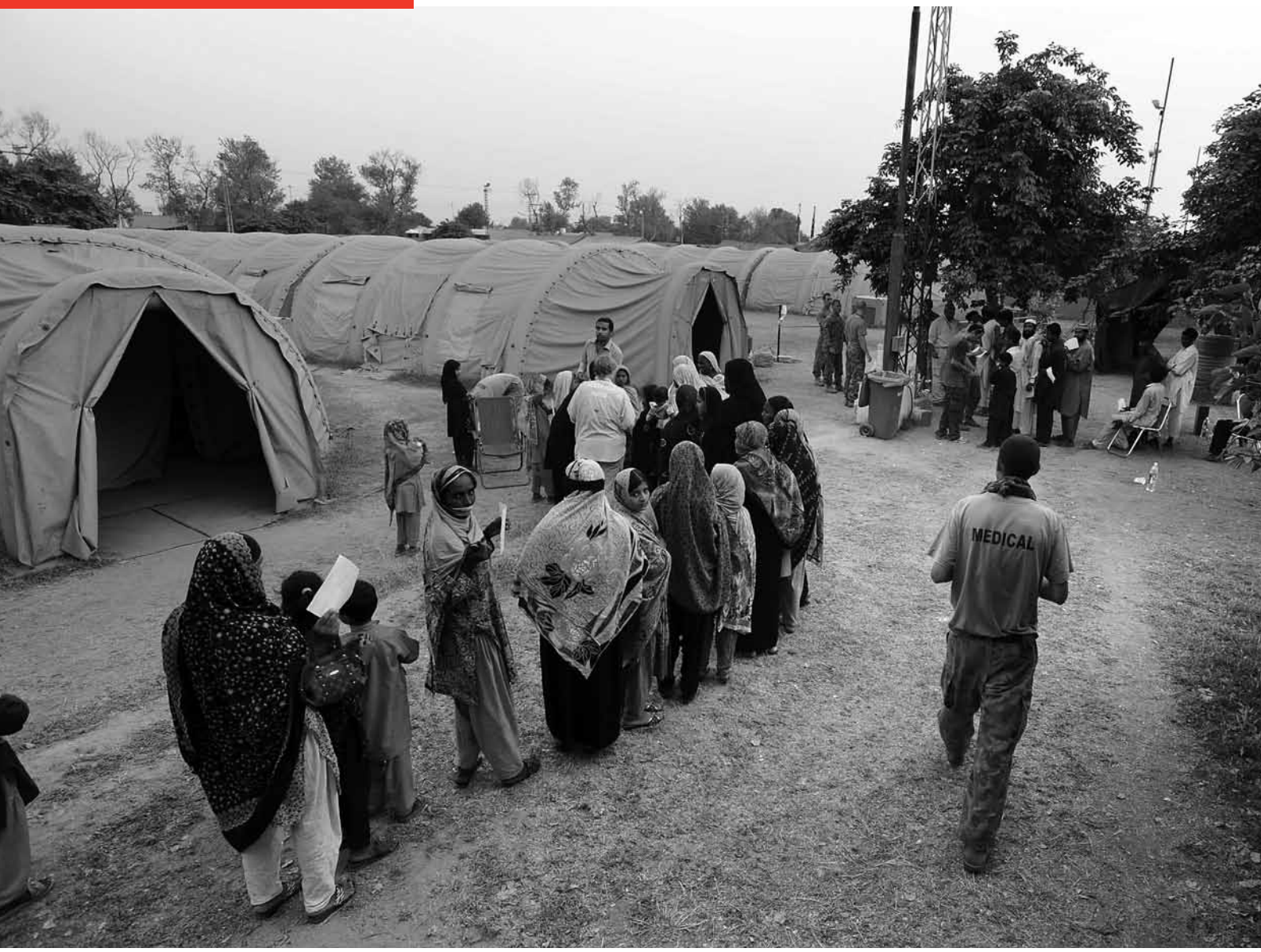
JMVH

Volume 21 Number 2 May 2013
Journal of Military and Veterans' Health



- Analysis of Presentations to the Sickbay of a RAN Warship
- Classification Of Physical Conditioning In Greek Army Officer Cadets
- Definition of Terrorism

The Journal of the Australasian Military Medicine Association

INNOVATIONS FROM THE BATTLEFIELD

AUSTRALASIAN MILITARY MEDICINE ASSOCIATION REPAT FOUNDATION INC. JOINT CONFERENCE 2013

ADELAIDE CONVENTION CENTRE
ADELAIDE, 1-3 NOVEMBER 2013

The Repat Foundation
Healthy Veterans, Healthy Communities

REGISTER NOW AND SUBMIT YOUR ABSTRACT

www.amma.asn.au/amma2013

Table of Contents

Editorial

Original Articles

Review Articles

Case Study

History

Cover photo: Courtesy of Department of Defence.

Journal of Military and Veterans' Health

Australasian Military Medicine Association

STATEMENT OF OBJECTIVES

The Australasian Military Association is an independent, professional scientific organisation of health professionals with the objectives of:

- Promoting the study of military medicine
- Bringing together those with an interest in military medicine
- Disseminating knowledge of military medicine
- Publishing and distributing a journal in military medicine
- Promoting research in military medicine

Membership of the Association is open to doctors, dentists, nurses, pharmacists, paramedics and anyone with a professional interest in any of the disciplines of military medicine. The Association is totally independent of the Australian Defence Force.

JMVH is published by the
Australasian Military Medicine Association

113 Harrington Street
Hobart Tas 7000
Australia
Ph: 61 3 62347844
Email: secretariat@amma.asn.au

ISSN No. 1835-1271

Inside this edition

As we enter May 2013, the challenges of providing military healthcare continue to intrigue our authors. With the new Defence White Paper and the expected withdrawal of our troops from Afghanistan over the next 12 months, the expected roles of our Defence forces, and the health support required to undertake those roles, continues to be a major matter for discussion. Similarly, the support to our veterans, including those from our more recent conflicts, will continue to tax available resources. Where will we be in 5 years' time is an intriguing question, which would benefit from some commentary from our readers.

While our submissions have picked up, we always are looking for more good military and veterans' health articles. As we move towards full Open Access, authors may like to consider the wider exposure that publication will bring to their articles, particularly as publication is peer-reviewed and without any authors fees or other costs.

In this issue, we have a range of excellent articles, including original articles on unexplained symptoms in veterans, sickbay presentations at sea and physical conditioning in the Greek Army. We also have a review article on terrorism, an interesting case study on Hepatitis C prophylaxis and another instalment in our history of the Australian Army Malaria Institute.

As we move through 2013, we have further themed issues and ask prospective authors to consider whether they may have suitable articles for these themed issues. Other military and veterans' health articles are always very welcome and we would encourage all our readers to consider writing on their areas of military or veterans' health interest.

Dr Andy Robertson, CSC

Editor-in-Chief

President's message

Welcome all to the lastest edition of the Journal, May finds us in the midst of preparation for our Annual Conference at the Adelaide Convention from 1-3 November. This promises to be bigger and better with the Repat Foundation Inc joining us as conference partners. Their involvement will no doubt broaden our scope of presentations and their contacts in the Adelaide community have meant that we will offer an exciting conference to the membership and delegates.

This year the Graeme Shirtley Oration will be delivered by Dr Brendan Nelson. Dr Nelson is the Director of the Australian War Memorial; prior to this, he was the Australian Ambassador to Belgium, Luxembourg, the European Union and NATO (2009–12). Many will remember Dr Nelson as a Federal member of parliament and a former president of the AMA. Our keynote speakers this year include Major General John Cantwell AO DSO, in 2010 he was the commander of all Australian forces in Afghanistan and the wider Middle East area of operations, for which he was awarded the Distinguished Service Cross for leadership in action. His book "Exit Wounds" is the compassionate and deeply human account of his combat experiences and resultant struggle with emotional trauma. Major General Cantwell is the Patron or Ambassador for several veterans' health organisations and is a prominent advocate for the care of veterans suffering emotional trauma resulting from their military service.

Our other keynote speaker will be Colonel Timothy Hodgetts CBE OStJ who was first appointed as Professor of Emergency Medicine in 1998 at the European Institute of Health and Medical Sciences. He was the inaugural Defence Professor of Emergency Medicine with the College of Emergency Medicine (2007-2010) and the Penman Foundation Professor of Surgery for 2011.

On the Journal front, it is pleasing to see the publication improving from edition to edition and the Editorial Board has some exciting plans for the future which our esteemed Editor will announce in due course.

I look forward to catching up with as many members as possible at our conference in November.

Greg Mahoney

President

Method Issues in Epidemiological Studies of Medically Unexplained Symptom-based Conditions in Veterans

Steven S. Coughlin[1], Rebecca B. McNeil[2], Dawn T. Provenzale[2,3], Erin K. Dursa[4], Catherine M. Thomas[2]

Abstract

Symptom-based conditions such as chronic fatigue syndrome (CFS) and medically unexplained multi-symptom illness (MSI) are fairly common in the general population and are also important veteran's health concerns due to their higher frequency among U.S. veterans who served during the 1990-1991 Gulf War. CFS, MSI, and other symptom-based conditions are often associated with considerable morbidity due to fatigue, chronic pain, neurologic symptoms, and other symptoms that can impair the quality of life. This article discusses several important issues of methodology that arise in population studies of CFS and MSI. These include the exclusion criteria that have been used in population studies to define CFS-like illness and unexplained MSI, the potential for false positive and false negative assessments of illness status, the potential for sex differences, and the poorly understood natural history of these symptom-based conditions across the life span. As an empirical example of these methodology issues, we examined existing data from a 2005 follow-up survey. We found that 64.9% (762 of 1,175) of female Gulf War veterans and 53.4% (2,530 of 4,739) of male Gulf War veterans had 1 or more exclusionary medical conditions. The prevalence among veterans with one or more exclusionary medical conditions increased markedly by age among females and those with a low income.

Key Words: chronic fatigue syndrome; medically explained multi-symptom illness; epidemiologic methods; menopause; Gulf War; survey; veterans

Symptom-based conditions are commonly seen in medical, neurologic, and psychiatric practice and are frequently the focus of epidemiologic studies, including those conducted to monitor the health and well-being of military and veteran populations. There has been extended discussion in the epidemiologic literature on how best to assess symptom-based conditions that are well-recognised in medicine and psychiatry—and for which diagnostic tests, procedures, or other clinical assessments are available—including those that are idiopathic or for which the causal mechanisms are only partly understood. Symptom-based conditions that are the focus of acute outbreak investigations have also been discussed in the literature on field epidemiology and emergency response. However, much less attention has been given to the epidemiologic assessment of health conditions that have no universally accepted medical diagnosis and cannot be further characterised by laboratory or diagnostic tests, particularly assessments that are included in longitudinal studies conducted over long periods of time.

We identified several important methodologic issues that arise in epidemiological studies of medically unexplained multi-symptom illness (MSI) in the course of planning two related national surveys of U.S. veterans (a follow-up survey of an established panel of veterans and a new survey and biorepository initiative), which are part of the Department of Veterans Affairs (VHA) coordinated efforts to carefully monitor the health and well-being of veterans who served during the 1990-1991 Gulf War.

Background

Results from epidemiologic studies indicate that U.S.-veterans who served in the 1990-1991 Gulf War are more likely to have medically unexplained multisymptom illness (MSI) than veterans who served during the same era but were not deployed to the Gulf[1,2]. Symptom-based conditions such as unexplained MSI and chronic fatigue syndrome-like (CFS-like) illness are health conditions of exclusion in that established medical diagnoses that may account for fatigue, chronic pain, or other debilitating symptoms first have to be excluded in order to identify possible cases. Psychiatric illnesses such as major depressive disorder (MDD) and post-traumatic stress disorder (PTSD) are an exception to this rule; they can co-occur with unexplained MSI

and CFS-like illnesses even though MDD and PTSD have been associated with chronic pain and fatigue. Epidemiologic studies have shown that deployment to the Gulf during the 1990-1991 war is associated with unexplained MSI and CFS-like illness even after adjustment for PTSD and MDD.[3,4]

Definitions of symptom- based conditions have varied across studies of Gulf War era veterans. For example, in the Longitudinal Health Study of Persian Gulf War Era Veterans[5], unexplained MSI was assessed using self-reported information about physical symptoms and illnesses (fatigue, muscle or joint pain, headaches, memory problems, digestive problems, respiratory problems, and skin problems) that persisted for 6 months or longer and were not adequately explained by an established, conventional medical or mental disorder diagnosis. Such unexplained physical symptoms and illnesses, which are often not labeled, are sometimes diagnosed as chronic fatigue syndrome, fibromyalgia, irritable bowel syndrome, or multiple chemical sensitivity. This study also used a modified version of the 1994 Centers for Disease Control and Prevention case definition of CFS[6] which takes into account differences in time frame[5]. CFS-like illness consisted of persistent problems in the past 12 months with fatigue lasting > 24 hours after exertion and persistent problems with at least three of seven symptoms (headaches, sore throat, tender lymph nodes, muscle aches or cramps, joint aches or pain, awakening feeling tired or worn out after a full night of sleep, and difficulty concentrating or reasoning or memory loss) and none of the following medical conditions: arthritis, skin cancer, other cancer, cirrhosis of the liver, hepatitis, diabetes, other endocrine disorder, repeated seizures or convulsions or blackouts, neuralgia or neuritis, disease of genital organs, coronary heart disease, stroke or cerebral vascular accident, tachycardia or rapid heart, asthma, emphysema or chronic bronchitis, and repeated bladder infections[5].

Another example of a modified definition was used in an epidemiologic study of illnesses among Gulf War veterans in Kansas[7]. They used a modified version of the CDC case definition of chronic MSI[8] that excluded veterans who had one or more medical conditions (cancer other than non-melanoma skin cancer, diabetes, heart disease other than high blood pressure, chronic infectious disease, liver disease, lupus, multiple sclerosis, stroke, bipolar disorder, schizophrenia) or who had been hospitalised since the Gulf War for depression, post-traumatic stress disorder, or alcohol or drug dependence.[7] There are several other examples of diverse exclusion criteria used to assess symptom-based conditions in epidemiologic studies of veteran and non-veteran populations. In some studies of symptom-based conditions, physical exams and biomedical assessments have been performed.[8-10] Efforts have been made (and are continuing to be made) to arrive at consensus definitions of these conditions that are suitable for epidemiologic research, clinical trials, or medical practice.[6,11]

False Negative And False Positive Assessments Of Multi-Symptom Illness

It has now been over 20 years since the 1990-1991 Gulf War, and there is ongoing interest in re-contacting established panels of Gulf War and Gulf Era veterans to conduct additional follow-up surveys, enhance existing studies by establishing biological repositories, and facilitate translational clinical research aimed at providing effective, evidence-based treatment, including complementary and alternative medicine therapy. In order to assess the frequency of symptom-based conditions in repeat follow-up surveys, it is necessary to consider both false positive and false negative assessments of these conditions at two or more time points.

For example, comparisons of symptom-based conditions in surveys conducted in 2005 and 2012 would need to consider the possibility of misclassification of symptom-based conditions at one or both time points, as well as the comparability of exclusion criteria. It is conceivable that these symptom-based conditions may wax and wane over time, similar to autoimmune conditions such as multiple sclerosis. This natural variation could contribute to misclassifications in assessment of symptom-based conditions based upon self-reported data or clinical examinations conducted at any time point. Results obtained from such studies conducted a few years following the 1990-1991 Gulf War suggested a potential for misclassification or resolution of self-reported symptoms[10,12]. It is possible that these symptom-based conditions may go into remission, resolve entirely, or, conversely, worsen over time. There is also very little known about the possible impact of treatment for PTSD, alcohol dependence, or other deployment-related health conditions on the course of symptom-based conditions.

Exclusion Criteria For Multi-Symptom Illness and Age

The use of exclusion criteria for medical illnesses in assessing symptom-based conditions is scientifically defensible given the fact that these health concerns are conditions of exclusion. However, almost half of men and women in the general U.S. population have one or more chronic illnesses (for example,

diabetes, cardiovascular disease, rheumatoid arthritis, chronic obstructive pulmonary disease, or neurodegenerative illness) and the percentage rises steeply with advancing age.[13,14] There has been little or no discussion in the literature about the impact of chronic illness or co-morbid chronic illnesses on the assessment of the population burden of symptom-based conditions in aging cohorts of veteran or non-veteran populations.

A further issue is that the population prevalence of some age-related chronic health conditions that can be associated with fatigue or chronic pain, such as obesity and osteoporosis-related fractures, have not traditionally been included as exclusion criteria for symptom-based conditions. Several authors have noted the high and increasing prevalence of overweight and obesity in veteran populations and in the general U.S. population.[15-17] We are unaware of any published information about whether the development of obesity worsens symptom-based conditions among 1990-1991 Gulf War era veterans. However, there have been studies of morbid obesity and metabolic syndrome among women in the general population who suffer from CFS.[18-19] Obesity, which is an established risk factor for several chronic diseases including coronary heart disease, diabetes, arthritis, and some forms of cancer, is associated with higher levels of inflammatory markers such as interleukin-6 and tumour necrosis factor-alpha.[20]

Multi-Symptom Illness And Gender

The published literature on the health of U.S. veterans indicates that symptom-based conditions are more common among men and women who had been deployed to the Gulf during the 1990-1991 conflict, as compared with men and women who had served during the same era but who had not been deployed to the Gulf.[1,2,5,21] Sex-differences have also been noted, with women at higher odds of reporting symptom-based conditions than men.[22] We are unaware of any published information about the relative impact of exclusion criteria for medically unexplained symptom-based conditions in women versus men, including those who served in the 1990-1991 Gulf War. However, because women veterans who served during the 1990-1991 Gulf War era are only now approaching the age at which they may be peri-menopausal or post-menopausal, the possible impact of menopausal status on symptom-based conditions in veteran populations is unknown. This is an important gap in our current understanding of the natural history of medically unexplained symptom-based conditions, particularly since some scientific theories about the causation of CFS and unexplained MSI focus on neuro-immune mechanisms.

Menopause has effects on a number of organ systems including the cardiovascular, skeletal, central nervous, and genitourinary systems.[23] Clinical research studies have shown that women who are peri-menopausal and post-menopausal also undergo important changes in immune function.[24] Early menopause is a risk factor for rheumatoid arthritis, and post-menopausal status is associated with greater tissue damage and disability in rheumatoid arthritis.[23] Studies have shown that following menopause, women can experience an increase in pro-inflammatory serum markers such as interleukin-1, interleukin-6, and tissue necrosis factor-alpha.[24] In non-veteran populations, there has been very little research on menopausal status and CFS. A recent case-control study of gynaecological history and CFS among civilian women in Wichita, Kansas found that a greater proportion of women with CFS than controls reported pelvic pain unrelated to menstruation (22.2% vs. 1.7%, p=.004), and periods of amenorrhea (53.9% vs. 46.2%, p=.06) (24). Menopause occurred about 4.4 years earlier in the CFS group (41.7 years vs. 46.1 years, respectively, p=.11), even though the mean ages of the cases and controls were very similar.[25]

Empirical Example

In order to illustrate these issues, we examined existing data from a 2005 follow-up health survey among population-based samples of 15,000 Gulf War Veterans and 15,000 Gulf Era Veterans who served in the military during the same era but who were not deployed to the Persian Gulf.[5] Our goal was to obtain the frequency of exclusionary medical conditions in these samples according to broad age categories in order to estimate what the overall frequency of excludeable medical conditions might be as the cohort of veterans ages. This analysis does not take into account possible cohort effects that might occur due to temporal changes in the prevalence of chronic disease risk factors, but it does provide useful information for study planning purposes and for thoughts on methodology issues.

Study population. The sampling frame consisted of 15,000 Gulf War Veterans and 15,000 Gulf Era Veterans selected for an earlier 1995 survey. Kang et al.[26] sampled Gulf War Veterans from 693,826 U.S. troops who were identified by the Department of Defense Manpower Data Center in Monterey, California as being deployed to the Persian Gulf area during the 1991 Gulf War. They sampled Gulf Era Veterans from 800,680 persons who represented about one half of all troops who were in the military between September 1990 and May 1991 but who did not serve in the Persian Gulf theatres of operations.

Branch of service (Army, Navy, Air Force, and Marine Corps) and unit component (active, reserve, and National Guard) were represented in both groups. Kang et al.[26] applied a stratified random sampling method to ensure that women and those who served in the reserve or National Guard were adequately represented. Approximately 20% of the sample were women. The survey data collection methods followed a modified Dillman method[27]; additional details may be found in previous publications.[5] About 26.8% of the Gulf War and Gulf Era Veterans in this sample (n=9,970) had unexplained MSI; the percentage with MSI was higher among those who had been deployed.[5] About 7.1% of the Gulf War and Gulf Era Veterans in this sample had a CFS-like illness; the percentage with CFS-like illness was higher among those who had been deployed.[5]

Current analysis. For our empirical example, we used the same criteria for CFS-like illness that were used in previous analyses of these data[4,5] except that we did not exclude those who reported they had ever been told by a doctor that they had skin cancer (since most cases of skin cancer in this population would not be melanoma). As noted above, these criteria consisted of a modified version of the 1994 Centers for Disease Control and Prevention case definition of CFS illness[6] which takes into account differences in time frame.[5] We also used the criteria for unexplained MSI that were used in published articles from the Longitudinal Health Study of Persian Gulf War Era Veterans.[5] Explanatory information in the survey questionnaire informed respondents that: "The following questions ask about unexplained multisymptom illnesses, that is having several different symptoms together that persist for 6 months or longer and are not adequately explained by conventional medical or psychiatric diagnoses…" Respondents were asked: "Since January 1991, have you ever experienced unexplained multisymptom illness that lasted 6 months or longer?" Thus, it was possible for respondents to self-report that they had unexplained MSI and to also report (earlier in the questionnaire) that they had been told by a doctor that they had 1 or more medical conditions considered to be exclusionary in other case definitions.[6]

Variables used in this example. The variables used included age in 2005 (categorized as <40, 40 to 49, 50 to 59, 60+ years), education (<high school, high school or equivalent, some college, associate degree, bachelor's degree, or graduate degree), income (<$20,000, $20,000-34,999, $35,000-49,999, $50,000-74,999, $75,000-99,999, >$100,000), deployment status (Gulf War, Gulf Era), and unexplained MSI (yes/no). We created a dichotomous variable coded as '1' if the respondent reported being told by a doctor that they had 1 or more of the exclusionary medical conditions, and '0' otherwise. Using SAS, we performed cross-tabulations of these variables as well as a logistic regression analysis in which the dependent variable indicated whether or not the respondent reported being told by a doctor that they had 1 or more of the exclusionary medical conditions (1=yes, 0=no).

Table 1. Adjusted odds ratios from logistic regression modeling of having 1 or more exclusionary medical conditions.

Covariate	Adjusted odds ratio (95% CI)
Age in 2005 (years)**	
<40	1.00*
40 to 49	1.47 (1.32-1.63)
50 to 59	2.86 (2.55-3.22)
60 +	4.31 (3.62-5.12)
Sex**	
Male	1.00*
Female	1.93 (1.73-2.15)
Deployment status**	
Gulf Era	1.00*
Gulf War	1.32 (1.21-1.44)
Education**	
< High school	1.00*
High school degree	1.05 (0.68-1.61)
Some college, no degree	1.26 (0.82-1.92)
Associate degree	1.13 (0.73-1.75)
Bachelors degree	0.96 (0.60-1.43)
Masters, doctorate, or professional degree	1.11 (0.71-1.72)
Annual household income**	
<$20,000	1.00*
$20,000 to $34,999	0.59 (0.49-0.72)
$35,000 to $49,999	0.48 (0.40-0.58)
$50,000 to $74,999	0.42 (0.35-0.50)
$75,000 to $99,999	0.39 (0.32-0.48)
>$100,000	0.36 (0.29-0.44)

*Referent category.

**p<.0001 from Wald chi-square test.

Results. The demographic and military characteristics of this sample have previously been detailed[4,5,15] The mean ages of the Gulf War (n = 6,111) and Gulf Era (n = 3,859) veterans who participated in 2004-2005 were 45.5 and 47.6 years, respectively. About 20.1 and 21.8 percent of respondents were women.

We found that the crude prevalence of unexplained MSI was 36.2% and 11.6% in the deployed and nondeployed groups, respectively, with a crude odds ratio of 4.33 (95% CI 3.87, 4.86), and an odds ratio of 4.25 (95% CI 3.78, 4.79) when adjusted for sex, education, and income. When persons who self-

reported that they had been told by a doctor that they had 1 or more exclusionary medical conditions were considered not to have unexplained MSI, the crude prevalence of unexplained MSI was 9.7% and 2.6% in the deployed (n=5,788) and nondeployed groups (n=3,714), respectively. However, the crude odds ratio of unexplained MSI comparing deployed to non-deployed increased to 5.08 (95% CI 4.05, 6.38) and 4.76 (95% CI 3.78, 6.00) when adjusted for sex, education, and income. In other words, when persons with a medical history of 1 or more of the exclusionary medical conditions are considered not to have unexplained MSI (but are retained in the denominator), the observed prevalence of MSI decreases considerably but the crude odds ratio of MSI (comparing deployed and non-deployed groups) increases. This is consistent with increased specificity of the case definition and a corresponding decrease in misclassification.

About 64.9% (762 of 1,175) of female Gulf War veterans and 53.4% (2,530 of 4,739) of male Gulf War veterans had 1 or more exclusionary medical conditions. Among male Gulf War veterans, the percentage with 1 or more exclusionary medical conditions increased markedly by age category (<40, 40 to 49, 50 to 59, 60+ years): 41.8%, 50.9%, 65.5%, and 73.1%, respectively. Among female Gulf War veterans, the percentage with 1 or more exclusionary medical conditions also increased by age category (<40, 40 to 49, 50 to 59, 60+ years): 56.4.8%, 63.4%, 78.8%, and 89.6%, respectively. Non-linear relationships were observed between educational attainment and the percentage with 1 or more exclusionary medical conditions in both male and female Gulf War veterans. About 65.2% of Gulf War veterans with less than a high school education had one or more exclusionary medical conditions compared with 55.6% of those with a high school education, college degree, or graduate degree. Income was strongly associated with the percentage with 1 or more exclusionary medical conditions, except among the relatively small number of women who were 60+ years of age. Among male Gulf War veterans with a reported income of <$20,000, 72.8% (386 of 530) had 1 or more exclusionary medical conditions compared with 49.5% (358 of 723) of those with an income of >$100,000 per year.

The overall pattern was similar for Gulf Era veterans, except that the frequency with 1 or more exclusionary medical conditions tended to be lower than in the Gulf War veterans. About 61.9% (508 of 821) female Gulf Era veterans and 47.7% (1,393 of 2,923) of male Gulf Era veterans had 1 or more exclusionary medical conditions.

In multivariate analysis using logistic regression (Table 1), advancing age, female gender, and deployment status were positively associated with having 1 or more exclusionary medical conditions ($p<.001$ in each instance). Annual household income was strongly and inversely associated with having 1 or more exclusionary medical conditions ($p<.0001$).

Discussion

To our knowledge, no epidemiological studies have examined CFS-like illness and unexplained MSI over 2 or more decades of life among U.S. veterans or members of the general population, although the VHA has efforts underway to address this important gap in scientific knowledge. A recent report by Li et al.[28] examined changes in health status in U.S. veterans who served during the 1990-1991 Gulf War era over a 10-year period (1995-2005). At 10-year follow-up, deployed veterans were more likely to report persistent poor health, repeated clinic visits, recurrent hospitalisations, CFS-like illness, and PTSD, than non-deployed veterans. In addition, deployed veterans were more likely to experience new onset of adverse health and certain chronic diseases than were non-deployed veterans, after adjustment for age and other confounding factors.[28]

As highlighted in our report, criteria for medically unexplained symptom-based conditions such as CFS and CFS-like illness require that persons with a history of certain medical conditions (e.g., cancer, heart disease, diabetes) be excluded. This is an important consideration in longitudinal studies of the natural history of CFS and unexplained MSI in both veteran populations and samples of the general U.S. population, particularly since the prevalence of chronic conditions such as diabetes and obesity have increased in recent years and both men and women are at increased risk of several chronic medical and neurodegenerative conditions as they get older.[15,16] A very high percentage of older men and women who participated in the 2005 survey,[5] particularly the older women, self-reported being told by a doctor that they had 1 or more of the exclusionary medical conditions, and thus would not be considered to have unexplained MSI or CFS according to "strict" criteria such as those used by Steele et al.[7] in their study in Kansas. It is also important to note that "strict" criteria for unexplained MSI exclude a very high percentage of U.S. veterans who served in the 1990-1991 Gulf War or during the same era who had low income or less educational attainment. This may contribute to a perception that unexplained MSI is a condition of the relatively affluent and well-educated. Policy decisions about optimal case definitions for symptom-based conditions should

take into account both scientific considerations (e.g., approaches for increasing specificity and decreasing misclassification) and the potential impact on vulnerable groups of veterans.

Although our empirical example uses data from a cross-sectional survey conducted in 2005 to project the age-specific frequency of exclusionary medical conditions in the future, our estimates are likely to be conservative because:

1. we did not take into account a self-reported history of alcohol dependence, serious psychiatric illness, or HIV/AIDS, and
2. the population frequencies of obesity, diabetes, and certain other chronic medical conditions which have increased in the U.S. over the past several years.

Nonetheless, these data provide a quantitative assessment of how definitions and exclusion criteria for symptom-based conditions can potentially introduce statistical bias, especially in an ageing population. Continued discussion and empirical analyses are likely to contribute to new or updated consensus definitions of symptom-based conditions that are suitable for longitudinal epidemiologic research.

Acknowledgments

The authors are indebted to Dr. Han Kang and Dr. Clare Mahan who served as Principal Investigators in the 2005 survey used in our analysis as an empirical example.

Authors' Affiliations:
1 Department of Epidemiology, Rollins School of Public Health, Emory University, Atlanta, GA
2 Durham Epidemiologic Research and Information Center, Veterans Affairs Medical Center, Durham, NC
3 Division of Gastroenterology, Duke University Medical Center, Durham, NC
4 Epidemiology Program, Post-deployment Health Group, Office of Public Health, Department of Veterans Affairs, Washington, DC
Corresponding author: Dr. Erin K. Dursa
Email: erin.dursa2@va.gov

References

1. Committee on Gulf War and Health: Health Effects of Serving in the Gulf War, Update 2009; Board on the Health of Selected Populations. Washington, DC: Institute of Medicine, 2010.
2. Kelsall HL, McKenzie DP, Sim MR, Leder K, Forbes AB, Dwyer T. Physical, Psychological, and Functional Comorbidities of Multisymptom Illness in Australian Male Veterans of the 1991 Gulf War. Am J Epidemol. 2009; 170:1048-56
3. Wolfe J, Proctor SP, Erickson DJ, et al. Relationship of psychiatric status to Gulf War veterans' health problems. Psychosom Med 1999;61:532-40.
4. Coughlin SS, Kang HK, Mahan CM. Alcohol use and selected health conditions of 1991 Gulf War veterans: survey results, 2003-2005. Prev Chronic Dis 2011;8:A52 http://www.cdc.gov/pcd/issues/2011/may/10_0164.htm. Accessed March, 9 2012.
5. Kang HK, Li B, Mahan CM, et al. Health of US veterans of 1991 Gulf War: a follow-up survey in 10 years. J Occup Environ Med 2009;51:401-10.
6. Fukuda K, Straus SE, Hickie I, et al. International Chronic Fatigue Syndrome Study Group: the chronic fatigue syndrome: a comprehensive approach and its definition and study. Ann Intern Med 1994;121:953-9.
7 . Steele L, Sastre A, Gerkovich MM, Cook MR. Complex factors in the etiology of Gulf War Illness: wartime exposures and risk factors in veteran subgroups. Environ Health Persp 2012;120:112-8.
8 . Fukuda K, Nisenbaum R, Stewart G, et al. Chronic multisymptom illness affecting Air Force veterans of the Gulf War. JAMA 1998;280(11):981-8.
9 . Eisen SA, Kang HK, Murphy FM, et al. Gulf War veterans' health: medical evaluation of a U.S. cohort. Ann Intern Med 2005;142:881-90.
10 .Spencer PS, McCauley LA, Lapidus JA, et al. Self-reported exposures and their association with unexplained illness in a population-based case-control study of Gulf War veterans. J Occup Environ Med 2001 ;43(12):1041-56.
11. Carruthers BM, van de Sande MI, De Meirleir KL, et al. Myalgic encephalomyelitis: international consensus criteria. J Intern Med 2011;270:327-38.
12. McCauley LA, Jooes SK, Lasarev MR, et al. Gulf War unexplained illnesses; persistence and unexplained nature of self-reported symptoms. Environ Res Section A 1999;81:215-23.

13. Hoffman C, Rice D, Sung HY. Persons with chronic conditions. Their prevalence and costs. JAMA 1996; 276:1473-9.14 . Anderson G, Horvath J. The growing burden of chronic disease in America. Public Health Rep 2004;119(3):263-70.

15 Coughlin SS, Kang HK, Mahan CM. Selected health conditions among overweight, obese, and nonobese veterans of the 1991 Gulf War: results from a survey conducted in 2003-2005. Open Epidemiol J 2011;4:140-6.

16. Koepsell TD, Littman AJ, Forsberg CW. Obesity, overweight, and their life course trajectories in veterans and non-veterans. Obesity 2012;20(2):434-9.

17. Almond N, Kahwati L, Kinsinger L, Porterfield D. Prevalence of overweight and obesity among U.S. military veterans. Mil Med 2008;173(6):544-549.

18. Maloney EM, Boneva RS, Lin JM, Reeves WC. Chronic fatigue syndrome is associated with metabolic syndrome: results from a case-control study in Georgia. Metabolism 2010:59:1351-7.

19. Aslakson E, Vollmer-Conna U, Reeves WC, et al. Replication of an empirical approach to delineate the heterogeneity of chronic unexplained fatigue. Popul Health Metr 2009;17.

20. Wildman RP, Kaplan R, Manson JE, et al. Body size phenotypes and inflammation in the Women's Health Initiative Observational Study. Obesity 2011;19:1482-91.

21. Blanchard MS, Eisen SA, Alpern R, et al. Chronic multisymptom illness complex in Gulf War I veterans 10 years later. Am J Epidemiol 2006;163:66-75.

22. Wolfe J, Proctor S, Erickson D, Hu H. Risk factors for Multisymptom illness in US Army Veterans of the Gulf War. JOEM 2002;44:271-81.

23. Sammaritano LR. Menopause in patients with autoimmune diseases. Autoimmune Rev 2012;11:A430-6.

24. Gameiro CM, Romao F, Castelo-Branco C. Menopause and aging: changes in the immune system—a review. Maturitas 2010;67:316-20.

25. Boneva RS, Maloney EM, Lin J-M, et al. Gynecological history in chronic fatigue syndrome: a population-based case-control study. J Women's Health 2011;20:21-8 .

26. Kang HK, Mahan CM, Kee KY, et al. Illnesses among United States veterans of the Gulf War: a population-based survey of 30,000 veterans. J Occup Environ Med 2000;42:491-501.

27. Dillman DA. Mail and Internet Surveys, 2nd ed. Hoboken, NJ: John Wiley & Sons, Inc., 2007.

28. Li B, Mahan CM, Kang HK, et al. Longitudinal health study of US 1991 Gulf War veterans: changes in health status at 10-year follow-up. Am J Epidemiol 2011;174:761-8.

Analysis of Presentations to the Sickbay of a RAN Warship During An Overseas Deployment

CMDR Ross Mills, RAN (MBChB, FRACGP, FAFOEM, FACAsM) and LSMED William Taylor, RAN (BSc Sport and Exercise).

Abstract

Objectives: To describe the pattern of sickbay utilisation and lost time aboard a RAN Major Fleet Unit during a prolonged overseas deployment.

Methods: A prospective cohort analysis was performed on the ships company of an Australian warship (n = 226) over a 100 day overseas deployment to multiple ports, to analyse sickbay utilisation (number of consultations) and lost time (number of days on reduced duties). Statistical analysis was performed using regression analysis, ANOVA and partitioning methods.

Results: Female gender and smoking were associated with increased sickbay utilisation and lost time. Males with MEC class 1 or non-smoking males with MEC 201 or worse were protective of sickbay utilisation. Being a non-smoking male over 22 years was protective of lost time. Gender, rank, MEC, age and smoker were all found to be discriminators. BMI was not related to either sickbay utilisation or lost time.

Conclusions: Being a male with no medical restrictions at the time of deployment was protective of sickbay utilisation, and being a non-smoking male older than 22 was associated with reduced morbidity. No association was found between BMI and either sickbay utilisation or morbidity.

Key Words: prospective cohort, sickbay utilisation, lost time, gender, smoking, age, BMI.

Introduction

It is recognised that work attendance and absenteeism is governed by more than purely medical factors,[1] and thus medical planners need to consider more than just injury / illness incidence when constructing health support plans. The literature also suggests that females and smokers have an increased incidence of being medically certified for reduced duties[2,3] with higher lost productive time.[2,4,5]

Smoking is currently banned indoors in Royal Australian Navy (RAN) ships and establishments, and from 01 Jan 2012, excise-free tobacco is no longer available on RAN vessels.

There are also multiple studies which suggest that increasing age may be associated with increased absenteeism.[6,7,8,9,10,11]. There is some evidence which conflicts with this, with age either being protective,[12] or unrelated[13] to absenteeism. That increasing age is cited by both Consensus Review[9] and Government Policy[10] suggests that there is a widely held belief that increasing age is indeed associated with absenteeism.

In 2004 a retrospective analysis of the ship's medical journals of RAN major fleet units was conducted between the years of 1991 and 2003.[14] The authors found an evacuation (Casevac) rate of approximately 1.2 evacuations per ship year, compared with USN figures of 2 Casevac per ship year. Morbidity rates were calculated as being 0.54 per 1000 man days (95% confidence limits, CL, 0.11 – 0.98) during Persian Gulf deployments and 0.52 (0.23 – 0.82) during refits. Morbidity consisted of summing the days of admissions to sick list onboard, admissions to hospital and sick while on leave.

There was a poor correlation between the total sick per 6 months and the number of man sea days (R^2 0.271).

These authors also estimated that (depending upon the size of the ship's complement) there would be 1 or 2 medical or surgical emergencies per ship year. These figures were not adjusted for the number of ship's company, although it is implied that this number was approximately 200. The presenting diagnoses and telephone calls to the Fleet Medical Officer are discussed, but not sickbay utilisation or lost time rates. The demographics of the ship's complement (including gender) were not analysed.

Royal Navy and South African Navy studies have found that female attendance rates at ship board

medical facilities are at least twice the attendance rates of men.[15,16]

The purpose of this study is to analyse current sickbay utilisation rates and patterns and morbidity rates aboard a deployed RAN major fleet unit, and to compare these with historic rates.

Background

HMAS Newcastle (FFG 06) is an Australian built US-designed Oliver Hazard Perry class escort frigate, operated by the RAN. Although usually manned by approximately 180 personnel, her crew was supplemented (to 226) for a four month deployment between April and August 2010. HMAS Newcastle has a Level One medical facility, having deployed with an embarked medical officer (the lead author).

On 17 April 2010 HMAS Newcastle departed Sydney for a deployment including Guam, Japan (Yokosuka), Canada (Esquimalt) and USA (Hawaii). This deployment included diverting to the Aleutian Islands (Alaska) to Casevac one of its complement, attending the Canadian Naval Centenary Fleet Review (Esquimalt), and participating in exercise RIMPAC (Hawaii).

This paper describes the presentations to HMAS Newcastle's sickbay during the first 100 days of its deployment, analysing patterns of presentations and morbidity. Morbidity is assessed as sickbay utilisation (presentations) and lost time (days certified with a medical chit).

Statistical Analysis

This paper attempts to identify relationships between the demographic variables age, gender, smoker, Body Mass Index (BMI) and rank, and sickbay utilisation and morbidity (with the measure of this being provided by a reduced duties medical chit).

Statistical analysis was performed using JMP statistical software package version 7.0.2 (SAS Institute).

In this paper the predictor variables are age, gender, BMI and Medical Employment Classification (MEC). At the time of this study the RAN was using a MEC numerical classification[17] to describe an individual's employability (and deployability) restrictions (Table 1).

Table 1. MEC Classification System

MEC Class	MEC Subclass	Employment Restrictions
1		NIL
2		Fit for seagoing duties with restrictions
	201	Restricted range of duties, no specific support required
	202	With pharmaceutical or other medical support
	203	With Advanced Medical Assistant or Nursing Officer support
	204	With Clinical Manager, specialist Military Nursing Officer, or Advanced Practice Military Nursing Officer support.
	205	With Medical Officer support
3		Temporarily unfit for seagoing duties
4		Unfit for RAN

BMI is a mathematical relationship between height and weight (height^2/ weight), and is used by the RAN as a tool for defining the MEC class of an individual.

The response variables are:

1. Number of days provided with a medical chit (being the total of days excused duties and selected duties). This is a marker of lost time and morbidity.
2. Number of presentations to the sickbay. This is a marker of sickbay utilisation.

The following analytic techniques have been used:

1. Descriptive analysis, describing patterns within the data without determining their statistical significance.
2. ANOVA.
3. Regression analysis including bivariate analysis.
4. Partitioning analysis based on population diversity.

Analysis of Variance (ANOVA) analyses the relationship between a continuous response variable and a categorical predictor variable. It focuses on the differences between the means of the different groups, measuring the change in the mean of the response variable when there is a change between the levels of the predictor variable. This form of analysis assumes that all observations are independent, and that the noise component of the statistical model is normally distributed. The Regression Correlation Coefficient (R^2) was used as a measure of the proportion of

the variability of the Response Variable explained by the Predictor Variables. R^2 is a measure of the degree of linear association between the Response and Predictor Variables being studied. The bigger the R^2 value (that is, the R^2 value being closer to 1.0), the more the variation in the Response Variable is explained on the basis of the Response Variable being tested.

Regression analysis (including Bivariate analysis) is a statistical method of modeling relationships between continuous variables calculating linear relationships (Linear Regression). This technique is used to identify the presence of statistically significant relationships between the Predictor Variables and the Response Variable(s).

This form of analysis assumes that:

- The relationship between the variables is linear
- The statistical noise is normally distributed
- The Response Variables are independent of each other
- There is constant variance of the Response Variable (the statistical noise has the same variance for all observations)

Partitioning analyses data on the basis of its population diversity, giving the probability that the second variable encountered comes from the same class as the first. Decision trees are based upon identifying splits which cause the maximum reduction in the population variability. The first splits are the best, with subsequent splits becoming less meaningful as the group size drops. Splitting was performed to four levels in this analysis.

Descriptive Analysis

Number of Consultations

The departure (Eastern Australian) time zone has been used for the dates in this analysis (to avoid adjusting for having crossed the International Date Line). The ship's company was dynamic during this deployment. For the purposes of the calculations in the analysis the ship's company has been deemed to be 226. (HMAS Newcastle left Sydney with 228 personnel, carrying 228 at its peak and 209 at day 100.) Twenty-two percent of the ship's company did not present to the sickbay for treatment. The data includes attendance at sick parade, vaccinations and medical examinations. The distribution of presentations is depicted in Figure 1.

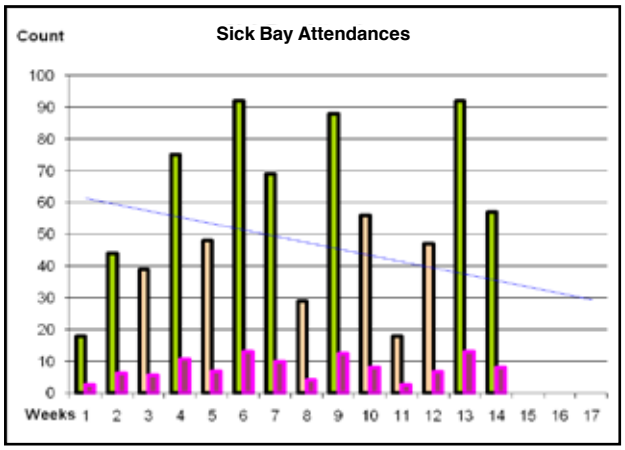


Figure 1. Weekly Sickbay Attendances

Key:

Green = a week including 2 or fewer days alongside
Orange = a week including 3 or more days alongside
Pink = daily average
Blue = trend line

There are three weeks with high presentations, being weeks 6 (a ship-wide viral illness), 9 (a ship- wide viral illness combined with sailing after a prolonged period alongside in Pearl Harbour) and 13 (where a flu vaccination programme was conducted).

A pattern was also identified between the number of sickbay presentations and whether HMAS Newcastle was alongside a port (in a foreign country), just about to depart from a port, or sailing (Figure 2).

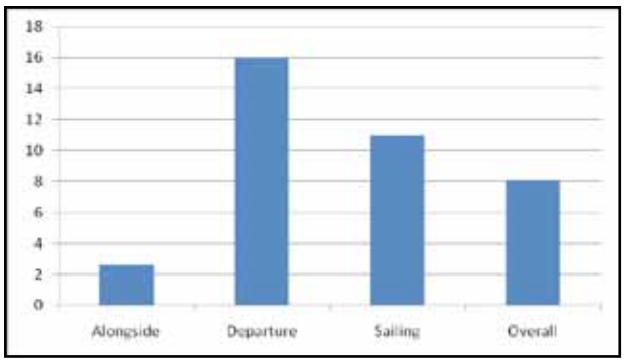


Figure 2. Sickbay Presentations by Ship Status

These presentations show the frequency of the most common diagnoses (based on the EpiTrack Descriptor) in Table 2.

Table 2. Presentation by Frequency

EpiTrack Descriptor	Count	%
Upper Respiratory Tract Conditions (including URTI)	101	11
Other Musculo-Skeletal Diseases (excluding knees and backs)	89	7.4
Disorders of the Ear, Nose and Throat	81	8.7
Other Dermatological Conditions	78	8.3
Medical Examinations: Routine, Periodic etc	48	5.1
Diseases of the Digestive System (excluding viral, bacterial or parasitic conditions)	47	5.0
Vaccinations, Inoculations and Prophylactic Injections	46	4.9
Counselling, Specimen Collection and Special Screening	33	3.5
Disorders of the Knee	32	3.4
Miscellaneous Administration: Routine Medicals	27	2.9
Disorders of the Back	26	2.8
Injuries Not Due to TAs, Training, Sport of Hostile Action	23	2.5
Lower Respiratory Tract Conditions (including Asthma)	22	2.4
Other	282	30
Total	935	-

A total of five cases required returning to Australia for medical reasons. None of the conditions for which these patients were casevaced were reflected in their pre-existing MEC.

There were an additional four patients with diagnoses requiring management away from HMAS Newcastle, but who were able to rejoin the ship at the completion of their treatment.

The variables, the total number of days certified "Excused Duties" (33 days) and "Modified Duties" (24 days) were added to produce the predictor variable "Number of Days with Chit". These variables were not analysed separately due to their small sample size. The total number of days "With Chit" comprised only 0.25% of the available man-days in this study (100 days with a ship's company of 226). Using O'Connor and Parrish's[14] definition of morbidity, this is equivalent to a morbidity rate of 1.4 days per 1000.

The ship's company was 85.8% male. By comparison, the population which did not present to the sickbay (22%) was 88.3 % male.

The age range was between 17 and 54, with a mean of 28.0 and median of 26. Age was not distributed normally, as depicted in Figure 3 (Normal Distribution and Smooth Curve). The dark colour represents male gender. The population which did not attend the sickbay ("Non-attender Group") had a mean age of 28.5 and median of 26.

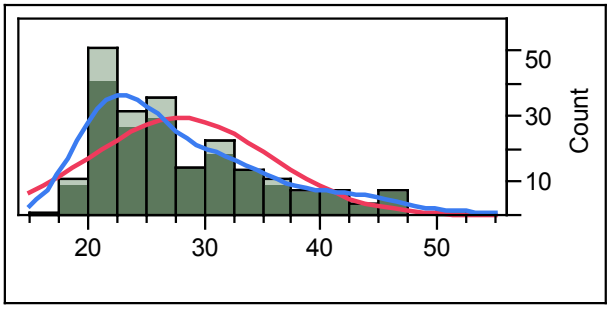


Figure 3. Distribution of Age

BMI ranged from 18.0 to 37.0, with a mean of 26.1 and median of 26.0. As depicted in Figure 4, BMI is close to being normally distributed (Normal Distribution and Smooth Curve). The Non-attender group was also close to being normally distributed, with a mean of 26.4 and median of 26.1.

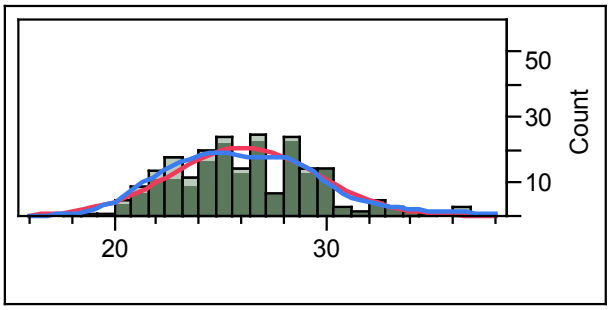


Figure 4. Distribution of BMI

The rank of Able Seaman (AB) comprised 43% of the ship's company and Seaman (SMN) 8.4%. By comparison, in the group of non-attenders these

lower ranks were slightly under-represented (AB 42% and SMN 6.7%).

Twenty two percent (22.2%) of the ship's company were MEC 201 or lower (Table 3).

Table 3. Distribution of MEC

MEC	1	201	202	203	204	205	3	4
Count	178	6	23	15	4	0	0	0

Regression Analysis

Oneway analysis of the number of consultations by gender found females to have more consultations than males, with the means being significantly different (Tukey-Kramer HSD). This is depicted in Table 4, with the Confidence Limits (CL) being 95%. Females account for 19.8% of the consultations or 1.39 the rate of males.

Table 4. Oneway Analysis of the Number of Consultations by Gender

Variable	Count	Mean	SD	Std Error	Lower CL	Upper CL
F	32	5.06	4.28	0.757	3.52	6.61
M	194	3.29	3.65	0.262	2.77	3.81

Bivariate analysis of the number of consultations found no relationship by age (R^2 0.001), BMI (R^2 0.003), or MEC (R^2 all 0.000). Comparisons for all pairs of rank (using Tukey-Kramer HSD) found no association. Oneway analysis of the number of consultations by smoker / non-smoker found no significant difference between the groups.

Stepwise regression calculated $R^2 = 0.090$ for gender, rank, smoker, age, MEC and BMI against the response variable number of consultations. This is interpreted as the predictor variables listed, accounting for 9% of the variation in the response variable number of consultations.

Oneway analysis of the number of days of chit by gender also found females to have more consultations than males, with the means being significantly different (Tukey-Kramer HSD). This is depicted in Table 5. Females account for 34 days on chit (30.0%), or 2.11 times the rate of males.

Table 5. Oneway Analysis of Number of Days of Chit by Gender

Variable	Count	Mean	SD	Std Error	Lower CL	Upper CL
F	32	1.06	1.95	0.345	0.3359	1.77
M	194	0.41	1.12	0.08	0.25	0.57

Bivariate analysis of the number of days of chit found no relationship by age (R^2 0.003), BMI (R^2 0.003), or MEC (R^2 all 0.000). Comparisons for all pairs of rank (using Tukey-Kramer HSD) found no association, with all CLs including one.

Stepwise regression calculated $R^2 = 0.125$ for gender, rank, smoker, age, MEC and BMI against the response variable number of consultations. This is interpreted as the predictor variables listed, accounting for 12.5% of the variation in the response variable number of consultations.

Partitioning

The variables identified as being the best discriminators in rank order for number of consultations were:

1. Gender
2. Rank (AB / SMN)
3. MEC (< 201)
4. Smoker

This form of analysis identified the two groups with the least number of consultations to be males of MEC 1 (n = 154, mean 3.01) and non-smoking Males with MEC 201 or higher (n = 25, mean 3.00). The group with the highest number of consultations was female with the rank of SMN / AB (n =19, mean 6.68).

The variables identified as being the best discriminators in rank order for number of days on chit were:

1. Gender
2. Age (22 years)
3. Smoker
4. MEC (< 201)

This form of analysis identified the group with the

smallest number of days on chit to be non-smoking males over the age of 22 (n = 96, mean 0.198). The groups with the largest number of days on chit were females (n = 32, mean 1.06) and male smokers with MEC 201 or lower (n = 5, mean 1.4).

Discussion

This deployment had a very high rate of Casevacs, being 18 per ship year compared with the 1 – 2 per ship year predicted by O'Connor and Parrish. The morbidity rate was also found to be significantly higher, being 1.4 days per 1000 compared with 0.54 man sea days per 1000 found by O'Connor and Parrish. The difference in sample size and study design between this paper and,[14] combined with the lack of demographic data of the study population in,[14] make meaningful comparison between these two study groups difficult. Potential explanations for the difference in medevac and morbidity rates between these two studies include:

- There being a more conservative medical practice aboard HMAS Newcastle (in the face of geographic isolation, the lack of ancillary diagnostic aids and a risk averse culture),
- There having been a change in the cultural approach to illness since 2004, and
- Personnel posting to sea with pre-existing conditions not reflected in their MEC (which resulted in half of the Casevacs).

There was one medical emergency (which required the patient to be Casevaced to Alaska for definitive treatment) during this deployment, consistent with the predictions of O'Connor and Parrish.

Neither age nor BMI were found to be associated with an increase in sickbay utilisation or morbidity. That two of the three fractures Casevaced subsequently required surgery suggests that overly conservative medical practice was insufficient on its own to explain the high Casevac and morbidity rates.

The small population size (n = 226) and the small number of days with chit (0.25%) markedly weakened the validity of this study. The only predictor variable with a statistically significant association with the response variables was gender. Female gender is associated with both increased sickbay utilisation and with a modified or excused duties medical chit.

The failure of this study to demonstrate statistically significant associations with the predictor v ariables of age, rank, BMI and smoking may also be a consequence of the other predictor variables not being normally distributed and thus not meeting the underlying assumptions of the regression model.

Partitioning did however demonstrate possible associations between the predictor variables of age (17 to 21 year old males), smoking, MEC, and rank. BMI was the only predictor variable not to show any degree of association with the response variables.

Although partitioning is unable to identify statistically significant associations, it is a useful tool for identifying clusters, and thus for hypothesis generation. This form of analysis found that being a male with MEC 1, or a male with MEC worse than 1 but a non-smoker, are protective of sickbay consultations, and a female of lower rank (AB or SMN) was associated with increased sickbay utilisation. Being a non-smoking male older than 22 was protective for lost time, and being a female was associated with increased lost time.

The findings of partitioning analysis for gender are consistent with the findings of regression analysis.

The findings of increased utilisation of the sickbay by females (and increased time on a medical chit) are potentially explainable at a variety of levels, including cultural, different disease/illness patterns. Females populate all of the different occupational classes onboard HMAS Newcastle, and thus this finding probably does not reflect a gender difference in work duties or the availability of selected duties. This finding is consistent with other literature which has also found increased absences due to sickness in females.

Conclusions

This study has found high rates of both morbidity and Casevacs during the first 100 days of HMAS Newcastle's deployment. Compared with males, females had increased rates of sickbay utilisation and morbidity. Being a MEC 1 male was protective of sickbay utilisation and being a non-smoking male older than 22 was associated with reduced morbidity.

The small size of the study population and the unique nature of this deployment (being a non-warlike cruise around the pacific rim) limit the general application of the findings of this study. Tests of statistical difference were not used when comparing sub-populations within this study, as the differentiation between "similar" and "statistically the same / different" were moot given the small population sizes. The failure of regression techniques to demonstrate statistically significant relationships is a reminder, not only of the limitations arising from small sample sizes, but also from potential failures of the study population(s) to meet the underlying regression assumptions.

The use of partitioning as a statistical tool attempts to overcome the major weakness of this study (small

population size) by identifying potential relationships buried within the data. This form of analysis is however only hypothesis generating. Further study (achieved through either larger study populations or applying metanalysis techniques) is needed to clarify these potential relationships within the RAN operational context.

This study also highlights some of the difficulties associated with the medical data currently available for analysis. Epitrack is a system of classification based around causality rather than pathology. It thus has limited application for analysing medical trends or illness/injury patterns.

The primary focus of this paper was sick bay utilisation to allow workload planning for future RAN deployments. To facilitate this, consolidated sick bay data (including vaccinations and medical) was used. The specifics of certain diseases/illness were not included in this paper to ensure that no personnel could be identified. This has resulted in dirty data, which combined with the limitations of the Epitrack classification system, further limits the ability to undertake meaningful analysis.

Future studies will need to consider how information is categorised, sample size and which statistical techniques are used in the analysis.

Study Implications

These study findings demonstrate that an embarked Medical Officer will be faced by a wide variety of health conditions encompassing multiple disciplines and sub-specialties. The pattern of presentations also has implications for health resource planning for mixed gender seagoing deployments, for ensuring that the MEC of personnel truly reflects their medical status, for the construction of medical watch bills when alongside (in foreign ports), and for the structuring of ship's sick parade timings when departing foreign ports.

Operational commanders should therefore expect a level of personnel attrition (including both temporary and permanent) during sea- going deployments of longer duration, and approximately one medical or surgical emergency every 6 month deployment.

No evidence was found for any relationship between BMI and the health indicators sickbay utilisation or lost time.

Further research needs to be conducted to determine the general application of these findings. The small sample numbers prevented any meaningful analysis of a potential relationship between job stress and morbidity. This potential association may however be able to be analysed in a bigger sample, where there is sufficient statistical power to compare morbidity rates between either departments or employment categories.

Disclaimer: The opinions expressed in this paper are those of the primary author alone and do not reflect those of the RAN.

Acknowledgements

The support of the Clinical Manager of HMAS Newcastle (POMED M Rowbottom) is gratefully acknowledged.

Authors' affiliation: Royal Australian Navy
Corresponding author: CMDR Ross Mills, CP4-6-009
PO Box 7913 BC, Canberra, Australian Capital Territory 2610
Email: ross.mills2@defence.gov.au

References

1. Australasian Faculty of Occupational and Environmental Medicine: Workplace Attendance and Absenteeism. A consensus statement.
2. Mills R, Predicting Failure to Return to Work; Internal Medicine Journal. 2012; 42: 924-27
3. Atlas SJ, Chang Y, Kammann E, Keller RB Long-term disability and return to work among patients who have a herniated lumbar disc: the effect of disability compensation.; J Bone & Joint Surgery. 2000; 82: 4
4. Stewart W F, Ricci J A, Chee E, and Moranstein D. Lost Productive Work Time Costs From Health Conditions in the United States: Results From the American Productivity Audit. 2003. JOEM 45 12: 1234-46
5. Boyce R W, Perko M A, Jones G R, Hiatt A H, and Boone E L. Physical fitness, absenteeism and workers' compensation in smoking and non-smoking police officers. Occ Med 2006; 56: 353-356
6. Terry L Blackwell, Stephen J Leierer, Stephanie Haupt, Angeliki Kampitsis. Predictors of vocational rehabilitation return-to-work outcomes in workers' compensation Rehabilitation Counseling Bulletin. Washington: 2003; 46: 108-14
7. Johansson JA. Work-related and non-work-related musculoskeletal symptoms. Applied Ergonomics 1994 25(4) 248-51
8. Kovacs F M, Abraira V, Zamora J, and Fernandez C The Spanish Back Pain Research Network, "The Transition from Acute to Subacute and Chronic Low Back Pain", SPINE, 2005; 30: 1786-1792.

9. Work related traumatic fatalities in Australia 1989 to 1992. Author: Work-related Traumatic Fatalities Study Team. 12 February 1999
10. Assessing Fitness to Drive. Austroads. 2003
11. Ijzelenberg W, and Burdorf A, 'Risk Factors for Musculoskeletal Symptoms and Ensuring Health Care Use and Sick Leave', SPINE 2005; 30: 1550-1556
12. Voss M, Floderus B, & Diderichsen F, 'How Do Job Characteristics, Family Situation, Domestic Work, and Lifestyle Factors Relate to Sickness Absence? A Study Based on Sweden Post', JOEM, 2004; 46: 1134-1143.
13. Wasiak R, Verma S, Pransky G, and Webster B. Risk factors for recurrent episodes of care and work disability: Case of low back pain JOEM. 2004; 46: 68
14. O'Connor, M and Parrish M. 'Morbidity and mortality on RAN ships.' ADF Health. 5 Sep 2004. pp 51 – 58.
15. Dorrell, M.G. 'Sick bay attendances by Royal Navy Women at sea.' J. R. Nav Med Serv. 1994 80 (2) pp 83-5.
16. Van Wijk, C. 'Never, Never Sick At Sea: Gender Differences in Health Care Utilisation on Board South African Naval Vessels.' Journal of Gender Studies. 14 (3) November 2005. pp 251-260.
17. Defence Instructions (General) PERS 16 – 15. Australian Defence Force Medical Employment Classification System.

Delineation And Classification Of Physical Conditioning In Greek Army Officer Cadets

Assist.Prof Konstantinos Havenetidis and Assoc Prof Thrasivoulos Paxinos

Abstract

Background: Even though considerable research has been conducted on the military fitness of reserves (soldiers, low-rank officers), few studies exist where career officers form the sample group.

Purpose: We assessed the physical fitness profiles of Greek Army Officer Cadets (GAOCs) and these data were compared with norms from age and sex-matched peers.

Material and Methods: 68 GAOCs from the Hellenic Army Academy participated in a series of laboratory tests (multistage shuttle run, handgrip dynamometry, counter movement jump, repeated bench press, sit and reach and body fat determination) and field tests (mile run, push ups, sit ups, pull ups and body mass index).

Results: Scores across all tests (mean ± SD) were on the 60th±20, 43th±13, 62th±27, 50th ±22, and 53th±18 percentile (males) versus the 60th±16, 63th±18, 63th±17, 65th±18, and 33th±14 percentile (females) for cardiorespiratory endurance, muscular strength, muscular endurance, flexibility and body composition respectively. Additionally, a mean 9% percentile increase across all GAOCs was found in field tests compared to laboratory tests (75th±13 versus 45th±22, 64th±16 versus 60th±24 and 33th±18 versus 53th±9 for the respective fitness abilities of cardiorespiratory endurance, muscular endurance and body composition.

Conclusion: Percentile scores for all fitness components place GAOCs above the average level (with the exception of body composition values for females) when compared with health norms for similar sample groups. However, a better standardised and/or more occupationally relevant fitness tests are needed in order to improve the accuracy of the physicalfitnessassessment.

Key words: Army personnel, Physical fitness, Work Capacity Evaluation, Diagnostic Techniques and Procedures, norms.

Conflict of Interest: There were no financial or personal conflicts of interest for this study. The results of the present study do not constitute endorsement of any product by the author or the Journal.

Introduction

The Hellenic Army Academy (HAA) is one of the oldest tertiary institutions in Greece. For all those who want to pursue a military career in Greece, the HAA is the only institution which qualifies them to be high ranking combat and non combat officers. Admission to military education depends on individual students' performance in national examinations at the end of their secondary education, in addition to their performance in the following physical fitness tests where they get a "pass" or a "fail" mark: a) 100m run b) long jump c) shot-put d) high jump and e) 1000m run. Candidates who fail to pass the physical conditioning tests are not admitted into the HAA but (not allowed to be admitted into the HAA) are replaced by others from a substitute list. Candidates who successfully complete these physical fitness tests,are considered to meet the HAA standards for entry into the Basic Combat Training.

Basic Combat Training is a mandatory standardised physical conditioning period that all potential Greek Army Officer Cadets (GAOCs) must attend according to the structure and missions of the HAA in Greece as described previously[1]. Additionally, passing a battery of physical fitness tests immediately following Basic Combat Training is also essential, as improving scores in these test items will improve the GAOCs' physical conditioning level, a prerequisite for performing future military occupational tasks.[2,3,4] These tests are part of a semestrial physical fitness examination and include the maximum number of push ups in one minute, the maximum number of sit ups in one minute, the maximum number of pull ups and (an) a one mile run.

During the first academic year at the HAA, the physical fitness conditioning programmes do not comprise skill-based military tasks such as passing military obstacle courses; therefore, the tests initially used in semestrial physical fitness examinations aim to keep GAOCs at a high general physical conditioning level. Despite the periodical (twice a year) character of the semestrial physical fitness examination, no comprehensive reports of GAOCs' fitness profile in relation to other populations (matched for age and sex) currently exist. Furthermore, no additional tests have been used in order to increase the low validity of field testing[5] and give more insight on GAOCs' specific exercise abilities. The semestrial physical fitness examination is considered a reliable model, where all GAOCs perform various tests under exactly the same conditions. It therefore provides an ideal opportunity to gather physical conditioning information about future Army officers. This data may also serve to validate GAOCs' physical conditioning evaluation practices within the HAA and eventually provide direction to design more efficient exercise programmes. The purpose of the present study was to delineate the fitness profile of GAOCs and classify them according to the health data norms which are derived from matched sex and age sample groups.

Material and Methods

A total number of 68 GAOCs (50 males and 18 females; plebes) were recruited from the HAA and who volunteered to participate in the present study, after being informed of the nature and risks of the experiments and after signing subject consent forms. Their mean ± SD values for age, body mass and body height were 19.8±1.0 years, 71±10.3 kg and 171±8 8.2 cm respectively. All procedures were approved by the Research Committee of the Greek Ministry of Defence. Ethical approval was obtained by the HAA Ethics Committee. This research has been conducted in compliance with all applicable international regulations governing the protection of human subjects in research.

The present study was conducted after the completion of the 7-week period of Basic Combat Training. That period consisted of an average of 15 hours running (intense and low paced), 2 hours sprinting (combat manoeuvres and live fire exercises), 35 hours marching (hiking with pack and equipment), 37 hours strength training (callisthenic and partner assisted exercises) and 43 hours military activities (prolonged standing in formation-bayonet training). Following a 48 hour rest from Basic Combat Training period, subjects participated in two series of tests (laboratory and field tests) interspersed by a 48 hour recovery.

The military abilities tested (endurance, strength and mobility) were selected according to the U.S. Army Physical Fitness School.[6,7,8]. Endurance refers to cardiorespiratory endurance, whereas the strength concept incorporates muscular strength/power and muscular endurance.

Mobility includes balance, flexibility, coordination, speed, and agility. In the present study, throughout the first and second series of testing, most of these components have been evaluated. (The only exception was mobility testing, which does not permit assessment in a single test because it comprises various skills, therefore, only flexibility was selected for evaluation). The only exception was mobility testing, which did not permit assessment in a single test because it measures various skills. Only flexibility was therefore selected for evaluation. Body composition was also assessed since there is evidence that it may be related to performance in various military tasks.[9]

Cardiorespiratory endurance was evaluated via a multistage shuttle run test (MSRT); strength via the handgrip dynamometer test (HDT) and power via the counter movement jump test (CMJT); muscular endurance via repeated paced efforts on a bench press (BPET); flexibility via the sit and reach test (SRT); and body composition via body fat determination (BF). Although many factors influence the testing sequence, in this study the more fatiguing tests were performed last. Consequently, tests were performed in the following order: BF, HDT, SRT, CMJT, BPET and MSRT. The battery of laboratory tests used to evaluate these qualities were selected due to their simplicity, swiftness, validity and reliability. Additionally, these tests are associated with the largest database of normative data. Since most of the military data worldwide are considered classified, not widely published and with limited access, percentile scores for GAOCs were determined according to various norms for healthy subjects in the respective age category.[5,10,11]

First series of testing - laboratory tests – MSRT, HDT (Average of both dominant and non-dominant hand), CMJT, BPET, SRT and BF.

Multistage shuttle run test

The test was conducted on a flat, clearly marked 20m stretch on an indoor running track. Subjects were required to run over and back on the 20m stretch, touching the line at either end with one foot, as a signal sounded from a pre-recorded tape (Multi-Stage Fitness Test, National Coaching Foundation, Leeds, UK). The signal from the tape was incremental and corresponded to a specific speed. The initial

running speed of 8.0km/h was increased to 9.0km/h after 1 minute and was subsequently increased by 0.5km/h each minute thereafter. The test was terminated when a subject voluntarily dropped out or did not make the line on two consecutive laps. Both lines were monitored closely by two spotters at either end. The final successfully completed lap was recorded as the finishing point. Subjects were instructed to complete as many laps as possible. The final successfully completed lap was expressed in metres per second, which was recorded and then converted to a VO^2max value.[12] Both the tape and tape recorder were calibrated before each test. Body mass and height were measured to the nearest 0.1kg and 0.5 cm respectively, using a balance beam scale (Seca 710, Hamburg, Germany) equipped with a stadiometer. During each measurement subjects were standing barefoot wearing minimal clothing. BMI was also calculated as body mass in kilograms divided by height in metres squared.

Handgrip dynamometry test

From the standing position and with the upper arm in vertical position, subjects placed their forearm at any angle between 90° and 180° (right angle to straight) of the upper arm. Their wrist and forearm was at midprone position. Then they exerted maximally and quickly the dynamometer (Takei 5001, Nigata, Japan) in each hand with at least 30s recovery between trials for the same hand. Three trials were allowed for each hand and the examiners recorded as the maximum score the sum of the best right and left grip strength measurement.

Counter movement jump test

Subjects performed three maximal vertical jumps on a portable Bosco force plate with dimensions 170x73cm (Musclelab, Ergotest Innovation, Italy) from an upright standing position with a preliminary counter movement of legs and arms. The flight time values obtained from the force-time curves were used to calculate the height of rise of the centre of gravity for the best trial.

Bench press endurance test

Subjects lay supine on a wide bench with the knees bent and the soles of the feet on the bench. The spotters handed the 36.3kg (males) or the 15.9kg (females) barbell into the subjects' hands (thumbs medial) spaced about shoulder width apart and at chest level. Upon the signal of the metronome (60 beats/min) the subjects raised the weight to a straightened-arms position directly above the chest and then returned the barbell to the preparatory position. The test was terminated when the subjects were unable to follow the pace of the metronome (30 lifts/min) or to reach full extension of the elbows. The score recorded was the maximum number of successful repetitions.

Sit and reach test (modified)

After a standardised 5 min warm up, shoeless subjects sat on the floor with their back, hips and head against a wall. Then they placed soles and heels against the sit and reach box (Acuflex I, Power Systems, USA) and fully extended their legs about shoulder width (20-30cm) apart. The starting (zero) position was determined when subjects reached forward as far as possible along the measuring device and slid the indicator without having their head and back leave the wall. After the recording was made, subjects reached again three times along the device, with each trial being held for at least 2 seconds. The best score across the three trials was recorded.

Body fat percentage determination

Body fat percentage (BF) was assessed using a hand-held bioelectrical impedance device (Omron BF300, Kyoto, Japan). Prior to testing all subjects were instructed to adhere to the following bioelectrical impedance guidelines[13]: i) empty bladder within 30 min of the start of the test, ii) no diuretic medications within 7 days before the test, iii) no exercise within 12 hours before the test, iv) no food or drink within 4 hours before the test and v) no alcohol within 48 hours of the start of the test. Measurements were made at a specific time period (06:00-09:00 am) in a comfortable and standard ambient temperature (22°C).

Second series of testing - field tests - push ups, sit ups, pull ups (only males) and a (an) one mile run. For these tests (except for the one mile run) the maximum number of repetitions during a one minute period was recorded for each individual separately. The one mile run took place on an outdoor synthetic track (tartan) 400m long and it was performed by 12 subjects each time. Three experienced track coaches recorded the time using handheld digital stopwatches (Accusplit 625, Linemore, USA). Push ups, sit ups and pull ups were performed using standardised procedures.[10] Subjects were allowed 30 minutes to recover between each series of test trials. They also followed the same eating, sleeping and activity schedules throughout the study. The reliability of all laboratory measurements was assessed by making repeated trials on successive days on a random subsample (n=10) of subjects (average intra-class correlation coefficient ranged from 0.90 to 0.97; $p<0.01$). Means and standard deviations (SD) of variables and fitness scores

were computed. A paired t-test was used to detect differences in percentile scores between laboratory and field tests. Probability values from level 0.001 to level 0.05 were taken to indicate statistical significance. All statistical analyses were conducted using the Statistical Package for Social Sciences (SPSS, Chicago, Illinois, USA) version 13.0.

Results

Mean ± SD values for the first and second series of testing as well as fitness assessment based on health norms are presented in Table 1.

The statistical analysis showed that there were significant differences in percentile scores ($p<0.001$ and $p<0.01$) between laboratory and field tests both in males and females.

Table 1. Mean ± SD values across all fitness tests with percentile scores in parentheses

Laboratory tests			Field tests		
Test	Females	Males	Test	Females	Males
MSRT VO2max (ml/kg/min)	42.2±6.1 (40%)***	49.6±5.3 (50%)†††	One mile run (min)	473±36 (80%)	371±33 (70%)
HDT (kg)	50.5±10.0 (55%)***	95.6±9.0 (40%)	Pull ups (reps)	--	12.0±7.0 (60%)
CMJT (cm)	28.4±4.36 (70%)***	35.0±5.1 (45%)††	Sit ups (reps)	40.0±3.0 (45%)	45.0±3.0 (50%)
BPET (reps)	25±8.0 (60%)***	25.0±10.0 (60%)†††	Push ups (reps)	40.0±2.0 (85%)	45.0±4.0 (75%)
BF (%)	23.0±3.7 (45%)***	13.8±5.3 (60%)†††	BMI (kg/m2)	24.3±2.5 (20%)	23.9±2.7 (45%)
SRT (cm)	43.6±7.8 (65%)	35.0±9.1 (50%)			
***p<0.001 significantly different from field test (males) †††p<0.001 and ††p<0.01 significantly different from field test (females)					

As shown in Table 1 during the first series of testing both female and male subjects were above the average category (50th-65th percentile) in four out of six laboratory tests respectively.

The mean percentile score for females and males were 56±18 and 44±22 respectively. During the second series of testing, female and male subjects were equal or above the average category (50th-65th percentile) in two out of four females and four out of five males. The field tests had mean percentile scores of 58±14 and 60±27 respectively . The mean percentile score for laboratory and field tests were 50±18 and 59±24 for females and males respectively. Percentile values for female and male subjects across laboratory and field tests for cardiorespiratory endurance, muscular strength, muscular endurance, flexibility and body composition are illustrated in Figure 1.

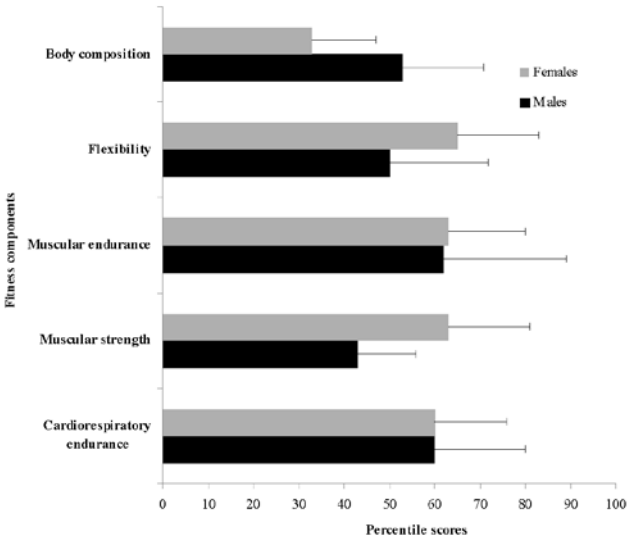


Figure 1. Average percentile scores across all fitness tests for the major components of military fitness

Discussion

This is the first study published where the physical fitness profile of a Greek military population has been assessed according to physical fitness norms. The present data suggest that subjects were classified according to health norms in the "average" category and especially from the 50th percentile (cardiorespiratory endurance) up to the 65th percentile (muscular endurance) across the four major military fitness components. Differences in fitness tests, military populations and age ranges between the present study and previous studies[14,15,16] complicate the comparison of test figures. Nevertheless, the subjects presented similar fitness values compared to previous studies,[7,18] where sample groups consisted solely of Army Officer Cadets.

In a study conducted in the U.S.,[17] Army Officer Cadets presented exactly the same VO^2max values as those in the present study (49.6 ml/kg/min), whereas females were characterized by slightly lower values (40.8 ml/kg/min). In another study (in the United Kingdom)[18] where the same testing methodology was used for aerobic capacity determination (time in the multistage shuttle run test), (subjects in the current study have considerably lower values compared to British Officer Cadets) subjects in the current study were found to have considerably lower values than those found for the British Officer Cadets [500 sec versus 720 sec (males) and 380 versus 515 sec (females)]. The British Officer Cadets however presented fewer pull up repetitions (8.3±4.3) when compared with their Greek counterparts (12.0±7.0).[14]

In terms of body composition, both male and female subjects were classified in the "below average" category (43rd percentile). In a study[18] where a similar testing instrument was used (bioelectrical impedance device), body fat values for males were considerably lower (11.7%) compared with those for the present sample group (13.8%). Despite that, both male sample groups were characterised by similar BMI values (23.90 versus 23.96) and the subjects possessed less fat-free muscle mass than their British counterparts. In contrast, (the opposite picture emerged with Greek female Cadets whaving higher BMI values than British officer Cadets) the Greek female Cadets had higher BMI values than the British officer Cadets (24.3 versus 23.0) but lower body fat values (24.3% versus 25.2%). Generally, the female subjects in our study seem to be characterised by higher BMI values compared with those in other Military Academies,[17,18,19,20] indicating the possibility that an increase of body mass may be due to decreased physical activity and/or an increased food/fat consumption over the years in the U.S.[21] but also in Greece.[22,23] Furthermore, body fat values such as those found in males in this study can be associated with an increased risk of injury.[24] Although this component of fitness (body composition) does not directly affect military performance,[18] these low percentile scores indicate that the potential for performance impairment still exists, considering these body composition values represent a status below that of an average healthy person.

It should be noted that in the present study the subjects' fitness level was evaluated on separate gender scales. This may be subject to criticism, as all military personnel are obliged to perform the same occupational tasks irrespective of gender. However, the current procedure was followed for the following reasons: Firstly, this is a common tactic in most military Academies; Secondly, the HAA policy requires that all cadets should always improve their physical conditioning status as part of their military preparedness. Based on this line of thinking, a constant and gradual improvement in fitness scores will eventually eliminate gender differences in military tasks, which predominantly require endurance capabilities, so trained females should reach comparable levels to males.[26] In contrast, if common scales were used, females in a short time would have reached a plateau in their physical conditioning or possibly deteriorate due to psychological reasons. Another reason for using separate gender norms was the policy of the HAA for improving general fitness abilities and not those derived from the actual mission demands. These latter will be specified after graduation, when cadets by that time will be officers and will follow careers in a specific corps (Special Forces, Artillery, Infantry, Logistics, Supply & Transportation, Army Aviation, Armoured Vehicles etc,). In these corps the "passing" criteria for the physical conditioning tests will no longer be age and/or gender adjusted.

It is also noteworthy that performance in the "commonly used" physical conditioning tests (push ups, sit ups, pull ups and one mile run) placed subjects to the upper end (59th percentile) of the "average" category (50th -65th percentile);. When more standardised tests were used (CMJT, BPET, HDT, MSRT), the average score dropped to the lower end (50th percentile) of the "average" category. This is possibly due to a more stringent control placed on laboratory tests as compared to those of field tests. However, these field tests are widely considered to be measures of health-related fitness,[25] they are conducive to mass testing and require little to no equipment, a key feature for military testing that often involves evaluation of hundreds of participants.

Nonetheless, simplified versions of CMJT and SRT, such as vertical/broad jump and fingertips to floor/ sit-reach toe touching, could be supplementary to field tests in order to broaden the range of exercise abilities tested. Other researchers[26,27,28] (have utilized similar tests such as standing vertical and horizontal jump, in order to simulate occupational military tasks.) have utilised similar tests, including the standing vertical and horizontal jump, to simulate occupational military tasks. Additionally, since the scores in military field tests impose a systematic bias against larger cadets,[29,30,31] the use of more occupationally relevant and physically demanding tasks will eventually obliterate body mass bias. Through this procedure, subjects would not only be evaluated from a health-related and occupationally relevant fitness perspective, but also under fairer conditions.

It would be also interesting to compare the present fitness scores with those during physical fitness testing prior to Basic Combat Training and/ or correlating them with Basic Combat Training completion-discard rates. However, the outcome of physical fitness testing was "pass or fail", therefore, no pre-training status data was registered owing to the nature of this process. Consequently, there was no opportunity to correlate the status of pre-training fitness with the completion of the Basic Combat Training course in order to determine whether or not a relationship exists. This lack of information can be considered a limitation of the present study.

In summary, these data show that GAOCs were aerobically fit, they presented strength levels within the "average" category, whereas their muscular endurance and flexibility scores were"above the average" for healthy individuals of the respective age group. Although their body-fat percentages were greater than expected for military personnel, the benefit of enhanced muscular endurance ability may overcome this drawback. The categorisation of subjects in the present study will also provide a reference value for other Military Academies within NATO and for the HAA the obligation to design more efficient physical conditioning programmes in the future. These results also have important implications for developing nutrition education programmes in the Greek Armed Forces. With the increasing childhood obesity in Greece, greater resources are required in order to minimise the negative effects on adolescents and so, potentially, Army Officers. Finally, the difference (9%) in the percentiles scores obtained by field tests compared to laboratory tests, emphasize the need to utilize more standardised and/or occupationally relevant fitness tests in the HAA semestrial physical fitness examination, in order to ensure its validity and specificity.

Acknowledgements

Many thanks to the Greek Ministry of Defence for the financial support of this study. The views expressed in this manuscript are those of the authors and do not reflect the official policy or position of the Department of the Army, the Department of Defence, or the Greek Government.

Authors' affiliation: Hellenic Army Academy
Corresponding author: Assist.Prof Konstantinos Havenetidis, Faculty of Physical and Cultural Education, Hellenic Army Academy, Vari-Koropiou Avenue, 16673, Vari, Attiki, Greece
Email: havenetidisk@sse.gr

References

1. Havenetidis K, Kardaris D, Paxinos T. Profiles of musculoskeletal injuries among Greek Army officer cadets during Basic Combat Training. Mil Med 2011;176(3):297-303.
2. Hoyt RW, Opstad PK, Haugen AH, et al. Negative energy balance in male and female rangers: effects of 7 d of sustained exercise and food deprivation. Am J Clin Nutr 2006; 83:068–75.
3. Knapik JJ, Daniels W, Murphy M, et al. Physiological factors in infantry operations. Eur J Appl Physiol 1990; 60:233–8.
4. Sharp MA, Knapik JJ, Schoppe AW. Energy cost and efficiency of a demanding combined manual materials-handling task. Work 1994; 4:162–70.
5. Adams GM. Exercise Physiology Laboratory Manual. 3rd edition. California State University: WCB McGraw-Hill; 1998.
6. Pate RR. A new definition of youth fitness. Phys Sportsmed 1983; 11:77–83.
7. Corbin CB, Dowell LJ, Lindsey R, et al. Concepts in Physical Education. Dubuque, Iowa: W.C. Brown Company; 1983.
8. Hogan J. The structure of physical performance in occupational tasks. J Appl Psychol 1991; 76:495–507.
9. Vogel JA, Friedl KE. Army data: body composition and physical performance. In: Marriott BM, Grumstrup-Scott J, editors. Applications for Military Services. Washigton DC: National Academy Press; 1992.

10. Nieman DC. Exercise Testing and Prescription: A Health Related Approach, 4th edition. Mountain View, CA: Mayfield Publishing Company; 1999.
11. Hoffman J. Norms for fitness, performance and health: Leeds: Human Kinetics; 2006.
12. Ramsbottom R, Brewer J, Williams CA. Progressive shuttle run test to estimate maximal oxygen uptake. Br J Sports Med 1988; 22:141-4.
13. Heyward VH, Wagner DR. Applied Body Composition Assessment, Champaign, IL: Human Kinetics; 2004.
14. Blacker SD, Wilkinson DM, Bilzon, et al. Risk factors for training injuries among British Army recruits. Mil Med 2008; 173:278-86.
15. Santtila MH, Kyrolainen T, Vasankari S, et al. Physical Fitness Profiles in Young Finnish Men during the Years 1975–2004. Med Sci Sports Exerc 2006; 38(11):1990–4.
16. Knapik JJ, Jones BH, Reynolds KL, et al. Validity of self-assessed physical fitness. Am J. Prev Med 1992; 8:367–72.
17. Thomas DQ, Lumpp SA, Schreiber JA, et al. Physical fitness profile of Army ROTC cadets. J Strength Cond Res 2004; 18(4):904–7.
18. Harwood GE, Rayson MP, Nevill AM. Fitness, performance and risk of injury in British Army Officer Cadets. Mil Med 1999; 164(6):428-34.
19. Beutler AI, De La Motte SJ, Marshall SW, et al. Muscle strength and qualitative jump-landing differences in male and female military cadets: The jump-ACL study. J Sports Sci Med 2009; 8:663-71.
20. McMillian DJ, Moore H, Hatler BS, et al. Dynamic vs. static-stretching warm up: The effect on power and agility performance. J Strength Cond Res 2006; 20(3):492–9.
21. Knapik JJ, Sharp MA, Darakjy S, et al. Temporal changes in the physical fitness of US Army recruits. Sports Med 2006; 36(7):613-34.
22. Yannakoulia M, Panagiotakos D, Pitsavos C, et al. Five-year incidence of obesity and its determinants: the ATTICA study. Public Health Nutr 2009; 12(1):36-43.
23. Papandreou C, Mourad TA, Jildeh C, et al. Obesity in Mediterranean region (1997- 2007): a systematic review. Obes Rev 2008; 9(5):389-99.
24. Jones BH, Bovee MW, Harris JM, et al. Intrinsic risk factors for exercise related injuries among male and female Army trainees. Am J Sports Med 1993; 21:705-10.
25. Knapik JJ, Rieger W, Palkoska F, et al. United States Army physical readiness training: rationale and evaluation of the physical training doctrine. J Strength Cond Res 2009; 23(4):1353–62.
26. Leyk D. Effects of gender on operational physical performance. In: intrinsic and extrinsic factors affecting operational physical performance, RTO technical report TR-HFM-080 Optimizing operational physical fitness, NATO, 7.8-7.12, 2009.
27. Maud PJ. Fitness assessment defined. In: Foster C, Maud PJ, editors, 2nd edition. Champaign (IL): Human Kinetics; 2006.
28. Frykman P, Harman EA, Gutekunst DJ, et al. Effects of US Army standardized physical training and a weight lifting program on body composition. Med Sci Sports Exerc 2006; 38:S272.
29. Harman E, Frykman P, Gutekunst DJ et al. US Army standardized physical training vs. a weight-lifting based program: effects on soldier physical performance. Med Sci Sports Exerc 2006; 38:S272.
30. Kraemer WJ, Vescovi JD, Volek JS, et al. Effects of concurrent resistance and aerobic training on load-bearing performance and the Army Physical Fitness Test. Mil Med 2004; 169(12):994–9.
31. Vanderburgh PM. Correction factors for body mass bias in military physical fitness tests. Mil Med 2007; 172(7):738–42.

Definition of Terrorism Social and Political Effects

Gregor Bruce

Introduction

Social structure and order, governance of society and politics are dependent on good communication, and good communication requires agreement on definitions of terminology. Terrorism can dramatically influence the world, as shown by the far-reaching and prolonged effects of the attacks in New York on 11th September 2001. The definition of terrorism will affect communication and response to this issue and so have consequences for society and politics. However a suitable universal definition remains elusive because different bodies, organisations and government agencies have different definitions to suit their own particular role, purpose or bias.

Methodology

A broad internet literature search was performed by entering key words in widely used internet search engines such as Google and Yahoo. Key words used were "terrorism" (plus derivatives such as "terror", "terrorist, etc)" and "definition" (plus derivatives such as "define", "defining", etc). This revealed a large number of internet references but the vast majority were published for purposes other than improvement in knowledge or for scientific research. Most were the publications of political organisations across the full spectrum from left to right, pressure groups, lobby groups, "think tanks" with a biased point of view, commercial organisations and "journalists" or "researchers" expressing a pre-conceived point of view. Often the information presented was second or third hand and had been altered to suit the bias of the author. All but a few were rejected. Separating truth from disinformation is a hazard when researching terrorism.

Organisations, governments, national states and other bodies that have social and political influence were searched through the internet and classical texts on the topic of terrorism in order to examine their definitions of terrorism and how these definitions affect their social and political influence.

These searches were performed during January and February 2012.

A search of the printed literature was performed with the assistance of the Charles Sturt University Library using the key words of "terrorism" and "definition".

United Nations

Terrorism is international. The command and control of terrorist groups, the recruitment, training, active operations and the target audience can all be located in different countries and so counter-terrorist measures will not be effective unless all nations cooperate in agreeing to the characteristics of terrorist groups and their activities. Agreement on a common definition would be a step towards universal cooperation in the prevention of terrorism. The UN unsuccessfully attempted to get universal agreement after the 1972 Munich Olympic massacre. Some nations, particularly in Africa, Asia and the Middle East, were unwilling to label groups as terrorists if they sympathised with their aims, because of the perjorative aspects of the label. The West has also sympathised with groups which have committed terrorist activities. The Reagan administration supported the Nicaraguan Contras and there was Western support for the African National Congress in South Africa in the mid-1980s when their actions were terrorist. A universal definition will define terrorism irrespective of the aims of the group. As stated by Louise Richardson.[1] "The legitimacy or otherwise of the goals being sought (by a group) should be irrelevant to whether a group is (defined as) a terrorist group" and "so a terrorist is not a freedom fighter and a terrorist is not a guerrilla. A terrorist is a terrorist, no matter whether or not you like the goal s/he is trying to achieve, no matter whether or not you like the government s/he is trying to change".

Nonetheless, the UN has struggled to provide a definition that is accepted by all nations. In 2001 it adopted the International Convention for the Suppression of Terrorist Bombings even though they were unable to define the word "terrorist". The Convention only covered one very small aspect of terrorism. The UN[2] produced an interim draft definition in 2001. It down-plays political justification and lists acts of violence as terrorism if they are "resulting or likely to result in major economic loss, when the purpose of the conduct, by its nature or context, is to intimidate a population, or to compel a Government or an international organisation to

do or abstain from doing any act." In 2007 they were shifting to a consensus academic definition. On 1 December 2010 the Head of the UN Counter-Terrorism Committee Executive Directorate said that "the fact that there was not a universal definition of terrorism presented a challenge".[3]

Badey,as quoted by White[4] agreed that "nations are hampered by an inability to define and criminalise terrorism" and this remains a problem in achieving trans-national counter-terrorism.

Academic Research

Researchers and academic students of terrorism desire the intellectual discipline of a definition to enable focussing on a specified topic. This would facilitate communication between researchers, their organisations and their contribution to society's counter-terrorism measures if they are using common language and definitions. Most academic definitions emphasise the combination of violence, politics, sociology and psychology. The threat of violence is included as well as actual violence.

Walter Laqueur[5] uses the simple, broad definition "terrorism is the illegitimate use of force to achieve a political objective by targeting innocent people".

Tore Bjorgo[6] states "terrorism is a set of methods of combat rather than an identifiable ideology or movement, and involves premeditated use of violence against (primarily) non-combatants in order to achieve a psychological effect of fear on others than the immediate targets."

Fernando Reinares (cited on p.120 in reference 6) distinguishes three traits that define terrorism for the purpose of academic study. Firstly, it is an act of violence that produces widespread disproportionate emotional reactions such as fear and anxiety which are likely to influence attitudes and behaviour. Secondly, the violence is systemic and rather unpredictable and is usually directed against symbolic targets. Thirdly, the violence conveys messages and threats in order to communicate and gain social control.

A very useful guide to academic thought is the study by Schmid and Jongman[7] referred to by many authors (e.g. Hoffman, White, Richardson, Bjorgo) which examined 109 definitions and found 22 frequently used "definitional elements" (Table 1).

Agreement on definitions of terrorism will assist the research and study which may progress to counter measures for the benefit of democratic governments and society.

Legal Profession

The legal profession desires a definition that can be used for the successful prosecution and conviction of accused terrorists. Defence or an appeal by an accused terrorist is easier if the crimes are ambiguously defined.

Prosecutions in the US can be under the Homeland Security Act of 2002.[8] This Act emphasises the danger to human life, covers the critical infrastructure and key resources, but also includes the psychological and political aspects.

Terrorism is covered by the "Criminal Code Act 1995 Part 5.3 Divisions 100-106 pp 95-126" of the Australian Federal Government[9] and defines terrorism (groups and individuals) for the purpose of prevention, investigation and criminal prosecution. It is primarily legalistic but does acknowledge the psychological, social and political aims of such groups. Members of the group that planned a suicide attack on Holsworthy Army Barracks were prosecuted and convicted under anti-terrorism legislation.

Prosecutors in Australia and overseas can have more success using conventional charges under the criminal code because of the imprecision of the legal definition of terrorism, particularly if a violent terrorist act has taken place, whereas anti-terrorist legislation becomes more relevant if there is a threat of violence or if the terrorist act is still in the planning stage.

Accurate legal definition of terrorism is important for society and for governance to enable successful investigation and prosecution of terrorists within the established judicial system.

Law Enforcement and Counter-Terrorist Agencies

Law enforcement agencies involved in counter-terrorism and intelligence (e.g. FBI, Special Branch of Scotland Yard, Australian Federal Police Counter-Terrorism) need definitions of terrorism as guidelines for their task and legal endorsement for duties which are close to (and sometimes over) the boundaries of civil liberty. Consequently their definitions have more emphasis on actions and criminality than motivation and psychology so that the investigation of individuals and groups can be justified more on the basis of their activities rather than their presumed motives. Actual acts of terrorist violence are emphasised above the threats of the violence.

The US State Department[10] describes terrorism as "premeditated, politically motivated violence perpetrated against non-combatant targets by sub-national groups or clandestine agents, usually

intended to influence an audience". The FBI's definition is "the unlawful use of force or violence against persons or property to intimidate or coerce a Government, the civilian population, or any segment thereof, in furtherance of political or social objectives".[11] Similarly the Australian Federal Police are guided by the Criminal Code Act 1995.[9] These definitions legitimise the actions of these agencies in the counteraction and investigation of terrorism rather than increasing their understanding of it.

These definitions can have significant social and political implications. They can benefit society by empowering effective counter-terrorism measures. They can harm society if they allow measures that cross the boundaries of civil liberties.

Governments and Political Parties

There are two reasons why politicians or governments will place importance on the definition of terrorism.

Firstly it can be used for public relations or "spin" to persuade their electorate that they are taking appropriate steps to combat terrorism and gain acceptance of laws or measures that are more draconian than would be accepted for any other purpose. An example is President George W. Bush's use of the expression "War on Terrorism" which categorises terrorists as a conventional military enemy and legitimises conventional military action rather than counter-terrorist measures which can be interpreted by the US electorate as being "too soft." The Obama administration has shifted from military to counter-terrorism and has since actually been accused of being "too soft".

Australian Federal Governments have also reassured the general public regarding their efforts to counteract terrorism, particularly because they are aware that votes will be lost if there is a perception that governments are doing otherwise.

A side benefit for governments is the opportunity to introduce laws that are more repressive than is usually the case. The laws may be directed to terrorism but frequently are sufficiently extensive or intrusive to increase government power generally. Citizens are more accepting of the loss of individual civil rights in the name of counter-terrorism. Government abuse is an over-reaction to terrorism and can be followed by a backlash by citizens. One of the aims of a terrorist act is to precipitate an inappropriate reaction by governments.[12] Truthful definitions of terrorism by politicians can help reassure and educate the public and preserve their civil rights.

Secondly, governments and politicians can use definitions of terrorism to repress, victimise or demonise their opponents, civilians, political bodies and religions. This happens most frequently in authoritarian states but has occurred in democratic states, an example being the use of Guantanamo Bay to sequestrate individuals from the normal legal system in the USA by defining them as terrorists. There are many examples of perversion of definition by authoritarian states, such as the labelling of French and Greek Resistance fighters as "terrorists" by Nazi Germany and the March 2012 description of Syrian civilians as "terrorists" by Syrian President Bassar al Assad while they are being killed by Syrian Government agents.[13]

Misuse of the definition of terrorism can have far-reaching social and political consequences. Political parties and religions can be outlawed and persecuted. An individual who is convincingly defined as a terrorist loses many civil rights. If they happen to reside in certain areas of Afghanistan or Pakistan, they are at risk of being killed by a drone.

Terrorist Groups

Unsurprisingly, terrorists' definitions of terrorism are different from those of the remainder of society. They prefer terms such as freedom fighter, guerrilla, insurgent and revolutionary. Richardson's comment on these terms has been discussed earlier.[1] Hoffman[14] also describes the attempts of terrorist groups to evoke more acceptable images of themselves by the use of favourable descriptors or definitions, e.g. "freedom and liberation", "armies or other military organisational structure", "self-defence movements", "righteous vengeance".

Osama bin Laden described "good and bad terrorism".[1] "Terrorism can be commendable and it can be reprehensible. Terrifying an innocent person.... is objectionable and unjust, also unjustly terrorising people is not right. Whereas terrorising oppressors and criminals and thieves and robbers is necessary for the safety of people and protection of their property.....The terrorism we practise is of the commendable kind for it is directed at the tyrants and the aggressors and the enemies of Allah, the tyrants, the traitors who commit acts of treason against their own countries and their own faith and their own prophet and their own nation. Terrorising those and punishing them are necessary measures to straighten things and to make them right."

There will be continuing social and political consequences as long as terrorists continue to define themselves in these terms and act accordingly. Terrorism will exist indefinitely because there will always be individuals and groups that get reassurance and motivation from this type of self-

justification.

Australian Department Of Defence

The Australian Federal Government delineates the mission and provides the budget for the Department of Defence and the Australian Defence Force (ADF) for the defense of the Australian homeland and its international security interests, including defense against terrorism. Terrorism needs to be accurately defined by the ADF so that it can pursue this task appropriately.

Searching the ADF website does not reveal a precise definition but does reveal past discussion papers which have confirmed that a clear definition is needed to help future planning of counter-terrorist measures as distinct from planning for conventional military action. For example, Major Adam Boyd uses the FBI definition of terrorism and then states that "a pre-eminent strategic studies speaker" at the 2004 Australian Command and Staff Course was "adamant that Australia did not have a comprehensive strategy to combat macro-terrorism".[15]

The Department of Defence publishes an annual update which includes a section on terrorism but does not define the term.

The Australian Federal Government published a White Paper on Counter-Terrorism in 2010 but it does not include a definition of terrorism.[16]

The Medical Profession

There is interaction between terrorism and the medical profession, since victims of terrorism will require treatment for physical and/or psychological injury.

Doctors, particularly military doctors or doctors involved in humanitarian assistance, can be faced with ethical or judgmental decisions when treating suspected terrorists. Terrorism, particularly suicide terrorism, can produce mass casualties with a mix of terrorists and their victims, requiring ethical discipline from the medical team to allocate treatment to the casualties of greatest need. Definitions of terrorism are irrelevant in this situation.

Captured or deserting terrorists are assessed by forensic psychiatrists and psychologists.

Research into the medical aspects, physical and psychological, of terrorism requires a specific definition of terrorism relevant to medical research.

Arnold et al.,[17] point out at that terrorism "definitions have been crisis-centred, frequently reflecting the political perspectives of those who seek to define it" and that "a universal medical and public health definition of terrorism will facilitate clinical and scientific research, education, and communication about terrorism-related events or disasters". Their proposition is as follows:

"The intentional use of violence--real or threatened--against one or more non-combatants and/or those services essential for or protective of their health, resulting in adverse health effects in those immediately affected and their community, ranging from a loss of well-being or security to injury, illness, or death."

Definitions by the medical profession put more emphasis on the psychological effects on victims and regard threatened violence as significant as is actual violence. The definitions strive to be an accurate reflection of the reality of terrorism and avoid terms which may imply bias or an emotional response to terrorism.

Medical definitions of terrorism are for the purpose of medical research and must not influence treatment of casualties and participants of terrorism. This must be provided on conventional ethical indicators.

Media

The media use the word "terrorism" as a term that will persuade people to read newspapers and watch television news programmes. It does not use a precise definition but calls events "terrorism" to catch the attention of the public (eg "school bullying terrorism", "terrorism in the western suburbs of Sydney", "economic terrorism", "West Indies cricket pace attack terrorism"). The main education of the public on terrorism is via the media and frequent misuse of the word will result in it becoming a meaningless cliché.

Conclusion

There is an assortment of collectives and individuals with a vested interest in terrorism and they have defined the term in the form that suits their bias or perspective. They include organisations and alliances of nations, academics and researchers, the legal profession, the health profession, counter-terrorist and law enforcement agencies, governments that wish to protect their citizens, governments that wish to repress their citizens, terrorist groups and the media. They have different agenda, even within their groups, and so it is unlikely that there will be agreement on a common definition of terrorism. This will have social and political consequences.

Author's affiliation: GK Bruce Pty Ltd
Contact author: Greg Bruce, GK Bruce Pty Ltd, PO Box 1158, Hunters Hill, NSW 2110
Email: gkbruce@tpg.com.au

Table 1. Frequencies of definitional elements in 109 definitions of terrorism

	Element	Frequency %
1	Violence, force	83.5
2	Political	65
3	Fear, terror emphasised	51
4	Threat	47
5	(Psychological) effects and (anticipated) reactions	41.5
6	Victim - target differentiation	37.5
7	Purposive, planned, systematic, organised action	32
8	Method of combat, strategy, tactic	30.5
9	Extranormality, in breach of accepted rules, without humanitarian constraints	30
10	Coercion, extortion, induction of compliance	28
11	Publicity aspect	21.5
12	Arbitrariness; impersonal, random character; indiscrimination	21
13	Civilians, noncombatants, neutrals, outsiders as victims	17.5
14	Intimidation	17
15	Innocence of victims emphasised	15.5
16	Group, movement, organisation as perpetrator	14
17	Symbolic aspect, demonstration to others	13.5
18	Incalculability, unpredictability, unexpectedness of occurrence of violence	9
19	Clandestine, covert nature	9
20	Repetitiveness; serial or campaign character of violence	7
21	Criminal	6
22	Demands made on third parties	4

Source: Alex P. Schmidt, Albert J. Jongman et. al, Political Terrorism: A New Guide to Actors, Authors, Concepts, Data Bases, Theories, and Literature. New Brunswick, Transaction Books, 1988, pp. 5-6

References

1. Richardson, L. 2006 What terrorists want John Murray
2. UN Ad Hoc Committee on Terrorism 2001 Informal texts of article 2 of the draft comprehensive convention Document A/C.6/56/L.9. 2001 session of the Working Group of the Sixth Committee.
3. Head of UN Counter-Terrorism Committee Executive Directorate. 1 December 2010 Press Conference UN Website
4. White, J. R. 2012 Terrorism and homeland security (7th ed) Wadsworth
5. Laqueur, W., 1977 Terrorism London: Weidenfeld and Nicholson
6. Bjorgo, T. (Editor), Gupta, D. K., Maleckova, J., Horgan, J., Post, J., Merari, A., (. . .) Silke, A. 2005 Root causes of terrorism Routledge
7. Schmid, A. P., Albert J. Jongman, A. J. 1988 Political terrorism: A new guide to actors, authors, concepts, data bases, theories and literature New Brunswick, NJ: Transaction Books
8. US Department of Homeland Security Homeland Security Act of 2002 Congress of USA
9. Australian Federal Government (1995) Criminal Code Act 1995 Part 5.3 Divisions 100-106
10. US Department of Homeland Security Homeland Security Act of 2002 Congress of USA Title 22 of the United States Code, Section 2656f [d]
11. Federal Bureau of Investigation Reports and Publications 2005
12. Griset, P. L., Mahan, S. 2003 Terrorism in perspective Sage Publications http://www.fbi.gov/stats-services/publications/terrorism-2002-2005
13. SBS Television 18 March 2012 World News
14. Hoffman, B. 2006 Inside terrorism (Revised edition) Columbia University Press
15. The relevance of current defence strategic policy in light of an altered international security environment Major Adam Boyd, Australian Army 2004 Australian Department of Defence Publication http://www.defence.gov.au/adc/docs/publications2010/PublcnsGeddes2004_300310_RelevanceofCurrent.pdf
16. Counter-Terrorism White Paper Securing Australia | Protecting Our Community 2010 Published by the Department of the Prime Minister and Cabinet http://www.dsto.defence.gov.au/attachments/counter-terrorism-white-paper.pdf
17. Arnold JL, Ortenwall P, Birnbaum ML, et al..... A proposed universal medical and public health definition of terrorism Prehospital Disaster Medicine 2003 Apr-Jun;18(2):47-52.

Post-Exposure Prophylaxis for Hepatitis B Virus after Exposure to a Person-Borne Improvised Explosive Device

Dr Csongor G. Oltvolgyi (FRACGP) and Dr Duncan B. Wallace (FRANZCP)

Abstract

The case of a member of the Australian Defence Force exposed to a person-borne improvised explosive device is presented. Indications for post-exposure prophylaxis against blood-borne viruses in this context are discussed, with specific focus on the 'non-response' to prior hepatitis B vaccination of this individual. This case received post-exposure prophylaxis against hepatitis B only, consisting of hepatitis B immunoglobulin and a course of intradermal vaccine. At six months this member remained free of serological evidence of infection. It is recommended that in the Australian Defence Force hepatitis B non-responder status be formally recorded by way of medical employment classification review, and that consideration be given to occupation groups other than health care workers where there might be benefit in routine testing for seroconversion after a primary course of hepatitis B vaccination.

Conflicts of interest: none declared.

The opinions expressed in this paper are those of the authors and do not necessarily reflect Australian Defence Force policy.

Background

The Australian Defence Force (ADF) has been on operations in Afghanistan since late 2001, and in significantly increased numbers since 2006. An ever-present risk on this operation continues to be that posed by improvised explosive devices (IEDs). In a decade of coalition operations in Afghanistan over 40% of fatalities have been as a result of IEDs[1]. This figure is reflected in Australia's experience in this war.

There are several ways in which an IED attack can be carried out. One of these is the person-borne IED (PBIED), colloquially known as a suicide bomber. Physical consequences of exposure to such blasts can include significant exposure to human tissue, from droplets through to penetrating bone fragments (Figure 1). If not fatal, such exposure can represent a risk of transmission of blood-borne viruses (BBV). Braverman et al. described a case report of bone fragments removed from a hepatitis B-negative PBIED victim which tested positive for hepatitis B surface antigen[2].

The ADF has a program of routine vaccinations provided to all members, and additional vaccinations are administered in anticipation of specific

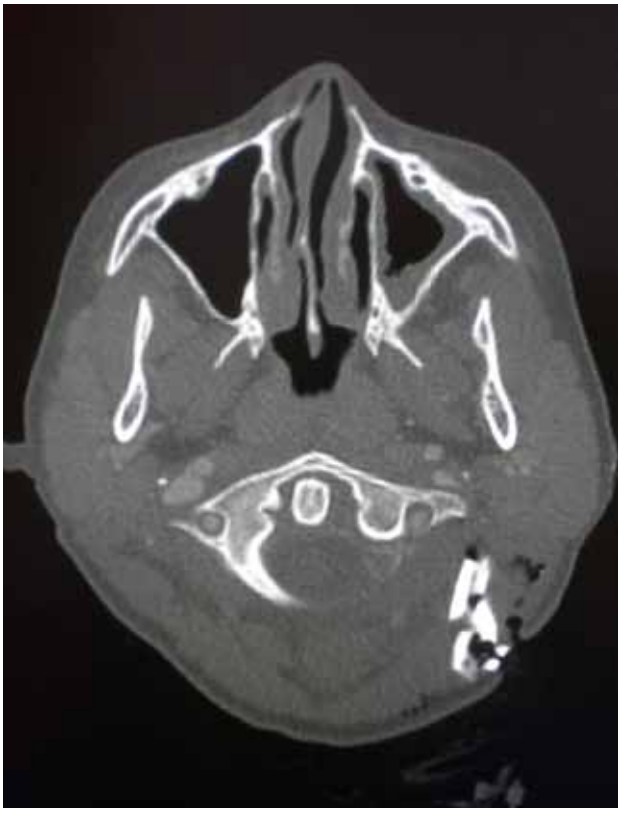


Figure 1. PEP for HBV after exposure to PBIED

exposures[3]. Hepatitis B vaccine is included in the routine vaccinations, and as there are no vaccines available to protect against hepatitis C and HIV, it is thus the only clinically important BBV against which there is presumed protection.

We describe the use of post-exposure prophylaxis (PEP) against hepatitis B virus (HBV) following exposure to a PBIED.

History

A 40 year-old male ADF member was exposed to a PBIED in Southern Afghanistan in early 2012. It was calculated that the suicide bomber was carrying approximately five pounds (~2.3 kg) of home-made explosives (HME), without significant fragmentary additives. The device was detonated without warning inside a building in a civilian area. The ADF member, who was wearing full disruptive pattern camouflage uniform with combat body armour but no helmet or eye protection, was located approximately three metres from the explosion, on the other side of a makeshift plywood wall. The force of the blast blew out the flimsy wall, exposing the member to the source of the blast. The only fatality was the suicide bomber himself.

The member was evacuated to a nearby coalition facility where he had a preliminary medical assessment. He did not require admission and was instructed to follow up in 24 hrs. The member returned to his primary duties in the interim.

Clinical findings

The first presentation by the member to an ADF role 1 medical facility was 27 hrs after the blast.

The member was able to recall the entire event, save for a few moments prior to and immediately after the blast. He was initially dazed and confused, and reported nausea and headache for approximately 24 hrs after the incident. After the blast his uniform, armour, and skin were covered in debris, including small dark flecks of firmly adherent material. He also recalled coughing, spitting, and clearing his eyes of debris for a time after the blast. Given the proximity of the bomber it is conceivable that some of this material contained human remains (HR).

In additional to mucosal exposure, the member also had a 5 cm graze on the neck (containing dried blood), and a 5mm full-thickness cut to the tip of the right middle finger.

The member had evidence of a full primary course of ADT vaccination in 1991, a booster in 2001, and an adult dTp_a booster (Boostrix®, GSK) administered in 2011. There was also evidence of a full primary course of hepatitis B vaccine (Engerix-B Adult®, GSK) in 1991. It was noted that there were subsequent doses administered in 1997 and 2003, but there were no anti-HBs titres in the deployed volume of the unit medical record (UMR), the medical record available to the treating medical officer in Afghanistan. Through contact with the member's ADF health centre in Australia, we were able to determine that an anti-HBs titre of 0.1 mIU/mL had been recorded in late 2002. Thus it is possible that the 4th and 5th doses of HBV vaccine were administered because of an absence of evidence for prior seroconversion. As a result, it was deemed that the member may still not have protective antibodies at this presentation.

Management

Post-exposure prophylaxis advice was sought via telephone from an infectious diseases physician in Australia.

The risk of contracting HIV in such an exposure incident from a source of unknown status was deemed to be very low. Post-exposure prophylaxis with zidovudine and lamivudine (Combivir®, GSK) was offered, but declined by the member after a discussion about the risk of contracting HIV, and the risks and benefits of treatment.

The risk of contracting HBV was considered to be significantly higher. Whilst prevalence data for blood-borne viruses in Afghanistan are scarce, it is apparent that the prevalence of HBV is significantly higher than HIV, in the range of 1.5 – 12.3 %, depending on the population studied[4-9]. Hepatitis B is also far more infectious than HIV10,11. For these reasons the member consented to HBV post-exposure prophylaxis. Hepatitis B immunoglobulin (HyperHEP B®, Talecris), at a dose of 1100 IU, was administered intramuscularly 33 hours after the blast. It was not possible to wait for a baseline anti-HBs titre, as this had to be performed in Germany, with a one-to-two week turn-around. In addition to the HBIG, an accelerated course of intradermal HBV vaccine (Engerix-B Adult®, GSK) was commenced because of the presumption that the member was a HBV vaccine 'non-responder'.

Outcome

The member had serology performed for HIV, HBV, and HCV at baseline, one, three, and six months.

Because of overnight laboratory closure, baseline bloods were not drawn until the day after administration of HBIG. This is likely to have contributed to the baseline anti-HBs titre of 72.6

mIU/mL. Negative baseline serology was returned for HIV, HCV, and HBsAg.

Anti-HBs was 244.2 mIU/mL, 156.8 mIU/mL, and 40 mIU/mL at one, three, and six months respectively (while the six-month test was performed at a different laboratory to the first three, the trend supports the suspicion of persisting non-responder status). Tests for HBsAg, and HIV and HCV antibodies were all negative at three and six months.

Discussion

Management of BBV post-exposure prophylaxis in any setting can be very complex. The comprehension of risk can be a challenge at the best of times, yet at a time of significant anxiety the patient, with the doctor's assistance, is required to assimilate complex epidemiology, modes of transmission, and efficacy of treatment all in a short period of time. Ultimately the decision to receive prophylaxis is a personal one, but guidelines can be helpful. The Centers for Disease Control and Prevention (CDC) in the United States have produced a guideline on BBV post-exposure prophylaxis in blast incidents by adapting existing recommendations for occupational and non-occupational exposures to blast mechanisms[12]. These guidelines were referred to in the management of this case (Table 1). The member was considered to have at least a category 2 exposure on the CDC scale.

It is widely reported that approximately 5-15 % of healthy immunocompetent individuals do not raise an adequate antibody response,[14,15] and that this is positively correlated with age.[16] The product information for the ADF recombinant vaccines, Engerix®-B (GSK) and Twinrix® (GSK), state that non-response is as low as 4 % and 0.7 %, respectively, after a correctly administered primary course.[17,18] Such non-response can persist despite multiple additional doses of vaccine, so vaccination in high-risk groups may need to be tailored to the individual.[19] The National Health and Medical Research Council (NHMRC) recommends testing for anti-HBs titres only in people with impaired immunity or with increased occupational risk, for example healthcare workers[15] , so it is unusual to have a titre performed on a military member who is not a healthcare worker. Such post-vaccination testing should be performed one to two months after the final dose of vaccine and if the anti-HBs titre is found to be > 10 mIU/mL in an immunocompetent person they can be assumed to have long-term protection and therefore not require any further testing[20] .

The management of people who are identified as not having seroconverted after a primary course of HBV vaccine is varied. The NHMRC recommends a fourth double dose or a further three doses at monthly intervals.[15] Another option, as in this case, is a course of intradermal vaccination using one quarter of the usual adult dose (5 mcg in 0.25 mL). This has been shown to seroconvert approximately 90% of healthcare workers who were non-responders to a primary intramuscular course of vaccine,[21,22] though long-term data are lacking for both this and the additional intramuscular dose approaches.[20]

Table 1. Post-exposure recommendations in the setting of blast, and with unknown vaccination status. Adapted from CDC12 with permission.

Risk category	HIV	HBV	HCV	Tetanus
Cat. 1 Penetrating injuries or non-intact skin exposures	Generally no action	Intervene	Consider testing	Intervene
Cat. 2 Mucous membrane exposures	Generally no action	Intervene	Generally no action	No action
Cat. 3 Superficial exposure of intact skin	No action	No action	No action	No action

In this case an assumption was made that the member was not immune to HBV. This was because of a negative anti-HBs titre before his fifth and final HBV vaccination in 2003 (though it is worth noting that some people who mount an effective humoral response early after vaccination no longer have measurable antibodies when retested at least one year later[13], causing some to question the use of antibody levels as a measure of protection). People who fail to raise an effective early humoral response to HBV vaccination, defined as an anti-HBs titre < 10 mIU/mL, are often termed 'non-responders'.

This case raises a few important questions regarding HBV vaccination in military members. Whilst the vaccine is routinely administered in the military, and is a requirement for deployment on operations, only healthcare workers have their anti-HBs levels checked. That said, it is hard to make a case for

population serological testing on the basis of this one exposure. Is it possible to define a sub-group of the military who should have serological testing, in addition to healthcare workers? Are all combat corps of the Army an appropriate group? What about those who are most likely to be exposed to injured enemy and human remains, for example special forces? What about those who are likely to conduct cache searches (unpublished data reveal a number of needle-stick injuries necessitating PEP in this context)? Can certain operations be anticipated to harbour greater risk, necessitating pre-deployment serological testing of an appropriate sub-group? In the context of current operations the authors suggest that, as a minimum, consideration should be given to pre-deployment anti-HBs testing of special forces elements, including their engineering support. Operational health advisors may identify other at-risk groups that should also be tested.

It is not known why this member had HBV serology performed after four doses of vaccine. However, once a person is identified as a non-responder, it is recommended by the authors that an attempt is made to induce seroconversion with further vaccine doses, using one of the techniques described above. If the person is a persistent non-responder, they should be carefully counselled about the implications of this, including education about high risk exposures, and the requirement for HBIG in the case of exposure.[23] For military members, non-responder status should be clearly recorded, ideally by Medical Employment Classification review, so that this is immediately apparent when managing a potential post-exposure prophylaxis case. Access in the deployed environment to serological testing, HBIG, and vaccine, may need to be considered prior to deployment. Appropriate first aid measures after contact with potentially contaminated materials should also be included as part of pre-deployment medical training, if not ADF-wide annual first aid training.

Hepatitis B is a serious but preventable illness. As long as military operations are conducted in regions where its occurrence is relatively high , and where access to a laboratory for performance of early anti-HBs titres is not available, consideration needs to be given as to how to manage the risk in the small but significant number of vaccine non-responders.

Acknowledgements:

We thank Associate Professor David Looke, Staff Specialist Infectious Diseases and Clinical Microbiology, for clinical advice and review of the manuscript, and Dr Brad McCall, Public Health Physician, for review of the manuscript.

Authors' affiliation: Department of Defence
Corresponding author: Csongor Oltvolgyi, B1-1-M054, Air and Space Operations Centre, Headquarters Joint Operations Command, Kings Hwy, Bungendore NSW 2621
Email: csongor_oltvolgyi@health.qld.gov.au

References

1. iCasualties.org [internet]. iCasualties.org; c2009 [updated 2012 Feb 8; cited 2012 Feb 8]. Available from: http://icasualties.org/OEF/index.aspx.
2. Braverman I, Wexler D, Oren M. A novel mode of infection with hepatitis B: penetrating bone fragments due to explosion of a suicide bomber. IMAJ 2002;4:528-529.
3. Australian Defence Force Publication 1.2.2.1. Immunisation procedures. [cited 2012 Feb 2]. Available from: http://intranet.defence.gov.au/home/documents/adfdocs/ADFP/adfp1_2_2_1.htm.
4. Centers for Disease Control and Prevention. Hepatitis B. In: CDC health information for international travel. Atlanta: CDC; 2012.
5. Todd CS, Abed AMS, Strathdee SA, et al. HIV, hepatitis C, and hepatitis B infections and associated risk behaviour in injection drug users, Kabul, Afghanistan. Emerg Infect Dis 2007;13:1327-1331.
6. Todd CS, Nasir A, Stanekzai MR, et al. HIV, hepatitis B, and hepatitis C prevalence and associated risk behaviors among female sex workers in three Afghan cities. AIDS 2010;24 (Suppl 2):S69-75.
7. Khan S, Attaullah S. Share of Afghanistan populace in hepatitis B and hepatitis C infection's pool: is it worthwhile? Virol J 2011;8:216. EPub 2011 May 11.
8. Todd CS, Ahmadzai M, Atiqzai F, et al. Seroprevalence and correlates of HIV, syphilis, and hepatitis B and C virus among intrapartum patients in Kabul, Afghanistan. BMC Infect Dis 2008;8:119. Epub 2008 Sep 17.
9. Quddus A, Luby SP, Jamal Z, et al. Prevalence of hepatitis B among Afghan refugees living in Balochistan, Pakistan. Int J Infect Dis 2006;10:242-247.
10. Hilleman MR. Comparative biology and pathogenesis of AIDS and hepatitis B viruses: related but different. AIDS Res Hum Retroviruses 1994;10:1409-1419.

11. World Health Organisation. Fact sheet No 204: Hepatitis B. [updated 2008 Aug; cited 2008 Feb 2]. Available from: http://www.who.int/mediacentre/factsheets/fs204/en/.
12. Centers for Disease Control and Prevention. Recommendations for postexposure interventions to prevent infection with hepatitis B virus, hepatitis C virus, or human immunodeficiency virus, and tetanus in persons wounded during bombings and similar mass-casualty events – United States, 2008. In: Morbidity and mortality weekly report 2008;57. Atlanta:CDC.
13. McMahon BJ, Dentinger CM, Bruden D, et al. Antibody level and protection after hepatitis B vaccine: results of a 22-year follow-up study and response to a booster dose. J Infect Dis 2009;200:1390-1396.
14. Siebert D, Locarnini S. Hepatitis B: issues in laboratory diagnosis and vaccination. Aust Prescr 1998;21:72-75.
15. National Health and Medical Research Council. Hepatitis B. In: The Australian immunisation handbook. 9th ed. Canberra: Department of Health and Ageing; 2008.
16. Fisman DN, Agrawal D, Leder K. The effect of age on immunologic response to recombinant hepatitis B vaccine: a meta-analysis. CID 2002;35:1368-1375.
17. GlaxoSmithKline. Engerix®-B product information. [updated 2011 Jun 21; cited 2012 Feb 1]. Available from: http://www.gsk.com.au/resources.ashx/vaccineproductschilddataproinfo/306/FileName/9C9D07C8FC17ED0CDDFF8CC691138AD5/Engerix_B_(Preservative_Containing)_Pi_v3.pdf.
18. GlaxoSmithKline. Twinrix® (720/20) and Twinrix® junior (360/10) product information. [updated 2011 Dec 23; cited 2012 Feb 1]. Available from: http://www.gsk.com.au/resources.ashx/vaccineproductschilddataproinfo/348/FileName/572E9F5DB1D66F8A22F1B7FA49A66382/Twinrix_(Preservative_Containing)_v4.0_clean_pdf_format.pdf.
19. Heininger U, Gambon M, Gruber V, et al. Successful hepatitis B immunization in non- and low responding health care workers. Hum Vaccin 2010;6:725-728.
20. Centers for Disease Control and Prevention. A comprehensive immunization strategy to eliminate transmission of hepatitis B virus infection in the United States: recommendations of the Advisory Committee on Immunization Practices (ACIP); Part 2: immunization of adults. In: Morbidity and mortality weekly report 2006;55. Atlanta:CDC.
21. Playford EG, Hogan PG, Bansal AS, et al. Intradermal recombinant hepatitis B vaccine for healthcare workers who fail to respond to intramuscular vaccine. Infect Control Hosp Epidemiol 2002;23:87-90.
22. Levitz RE, Cooper BW, Regan HC. Immunization with high-dose intradermal recombinant hepatitis B vaccine in healthcare workers who failed to respond to intramuscular vaccination. Infect Control Hosp Epidemiol 1995;16:88-91.
23. Hoy J, Richmond J. Standard precautions and infection control. In: Bradford D, Hoy J, Matthews G, editors. HIV, viral hepatitis and STIs: a guide for primary care. Darlinghurst: ASHM; 2008. p. 146-151.

Army Malaria Institute – its Evolution and Achievements Third Decade (2nd Half) : 1990-1995

Karl H. Rieckmann, Stephen P. Frances, Barbara M. Kotecka, Robert D. Cooper, G. Dennis Shanks, Anthony W. Sweeney, Michael D. Edstein

Abstract

The second half of the third decade (1990-1995) after the establishment of the Army Malaria Research Unit was characterised by substantial progress in meeting the challenges posed by drug-resistant malaria. In view of the rapid emergence of drug resistance, laboratory/field studies were carried out to develop novel and improved methods to assess and monitor changes in patterns of parasite susceptibility to antimalarial drugs. The high malaria attack rates observed during training exercises in Papua New Guinea (PNG) in 1988/1989 underscored the urgent need to provide better protection for soldiers deployed to malarious areas. This led to doxycycline substituting pyrimethamine/dapsone (Maloprim®) for prevention of falciparum malaria, but relapsing vivax malaria remained a problem. Laboratory investigations with tafenoquine indicated that this long-acting drug could eventually replace the cumbersome primaquine eradication course for prevention of vivax malaria. Furthermore, alternative antibiotics and halofantrine might be able to be used for prevention and treatment of falciparum malaria. Other studies showed that, apart from acting synergistically with dapsone, proguanil and PS-15 (experimental antifolate) potentiated the activity of atovaquone (new antimalarial) against drug-resistant parasites. Further investigations with Mannich base compounds revealed some of them to be more potent than amodiaquine or pyronaridine. Personal protection measures against insect bites were expanded by testing the effectiveness of various formulations of insect repellents when applied to skin, uniforms and bednets. After completing the survey of *Anopheles farauti* in northern Australia during the previous quinquennium, its spatial distribution was analysed in relation to detailed topography and historical climate records. Further anopheline surveys were performed in the Torres Strait and four PNG provinces to enhance future malaria control activities in these areas.

Background

The malaria situation continued to deteriorate in many tropical areas of the world during the mid- to late- 1980s.[1] In fact, most countries had made little progress in controlling malaria during the two decades following the establishment of the small Army malaria research laboratory in 1965.[2,3] The ability of developing countries to control malaria was hampered by the slow pace of development, rudimentary health infrastructures and various administrative, social and technical problems. Parasites were also becoming increasingly resistant to antimalarial drugs. This was facilitated by the widespread movement of people looking for work, food or a peaceful environment, and the proliferation of mining, agricultural or deforestation projects. Although chloroquine resistance was considered to affect only *Plasmodium falciparum*, the Army Malaria Research Unit (AMRU) documented in 1989 that *P. vivax* could also develop resistance to this drug.[4]

Significant progress was made at AMRU during the first half of the third decade (1985-1990) in using *in vitro* cultivation of *P. falciparum* for identifying the presence and degree of parasite drug resistance and for screening the activity of potentially useful antimalarial drugs.[5] In addition, the introduction of high performance liquid chromatography (HPLC) technology for measuring drug concentrations in body fluids and of the bioassay for estimating serum/plasma drug activity greatly increased the ability of AMRU to undertake investigations concerning the *in vivo* effectiveness of various drugs and drug combinations.[5] In fact, the use of these newly-established procedures suggested that daily proguanil (Paludrine®, 200 mg), in combination with a low dose of dapsone (diamino-diphenyl sulfone, 10 mg), might be more effective against drug-resistant malaria than a standard weekly regimen of pyrimethamine (12.5 mg)/dapsone (100 mg) (Maloprim®) prophylaxis. This was subsequently

confirmed in a field trial conducted in a malarious area of Papua New Guinea (PNG) during 1987/1988.[5]

During and after a 3-4 week field deployment of 163 Australian Defence Force (ADF) personnel to PNG in 1988/1989, about 6% of them developed falciparum malaria and about 20% developed vivax malaria.[6] Not only was weekly pyrimethamine/ dapsone/ chloroquine no longer providing adequate protection against falciparum malaria, but the 14-day (42-dose) primaquine eradication course was unable to prevent acute attacks of vivax malaria in an unacceptably high number of soldiers following their return to Australia. Alternative prophylactic regimens were obviously needed. Although favourable results had been obtained with proguanil plus low-dose dapsone, this drug combination was not registered for clinical use. Two other drugs – doxycycline and mefloquine – were registered, the latter only in 1988. Consequently, 224 Australian soldiers volunteered to take one of these two drugs while in PNG. Preliminary results showed that both drugs suppressed the blood stages of *P. falciparum* and *P. vivax*, and that both were potential candidates to replace pyrimethamine/dapsone/chloroquine for malaria prophylaxis.[6]

Studies were also carried out with anopheline mosquitoes and non-human primates to determine their susceptibility to different strains of *P. falciparum* and *P. vivax*.[5] Routine malaria transmission would provide suitable models for assessing the activity of experimental antimalarial agents against different stages of the human malaria life cycle.

The survey of anopheline mosquitoes in northern Australia, spanning a period of 6 years, was completed in 1990.[5] This was the most extensive characterisation of malaria vectors in a region that remains receptive to the re-introduction of malaria. During earlier field investigations with *Culicinomyces*, another mosquito pathogen – *Amblyospora* – had been discovered parasitising *Culex annulirostris*.[5] The complete life cycle of this microsporidian parasite in mosquitoes and its copepod intermediate host was able to be established and the feasibility of using it as a biological control agent was explored. However, the complexity of the parasite-host system, coupled with technical problems relating to larger-scale production, precluded the use of *Amblyospora* as a biological control agent for mosquitoes, at least at that time. Studies were also initiated to evaluate alternatives to the standard ADF mosquito repellent because it was not particularly well accepted by soldiers in the field and, besides, did the repellent require such a high deet concentration (95%)? The investigations showed the potential value of alternative repellent approaches to reduce contact with mosquitoes and chigger mites, including the impregnation of uniforms and bednets with permethrin, a synthetic pyrethroid compound.[5]

Staff and facilities

Professor Karl Rieckmann continued to direct activities at AMRU and was ably supported by Deputy Director and Commanding Officer, Lieutenant Colonel Anthony Sweeney. During this time, the Unit was privileged to receive the advice and support of the Army Malaria Research Advisory Board (AMRAB), chaired by the Director General of Army Health Services (DGAHS).

Figure 1. Army Malaria Research Advisory Board (1995)

Standing (L to R): LTCOL A. Gill, LTCOL A.W. Sweeney, COL M. Heugh, X, Prof J. Egerton.

Sitting (L to R): Prof W.J. O' Sullivan, BRIG B.G. Stevens, Prof K.H. Rieckmann, BRIG P.T.R. Buckley (DGAHS), Prof T.C. Sorrell.

Pharmacological investigations were somewhat curtailed because of the unexpected re-assignment of Major Robert Veenendaal to Canberra in 1990. Although Major Michael Edstein started a 3-year exchange posting at the Armed Forces Research Institute of Medical Services (AFRIMS), Bangkok in the same year, he was able to make important pharmacological contributions to drug studies of mutual interest to AMRU and AFRIMS. In 1991, Captain Anthony Yeo (Medical Officer) started a 3-year assignment during which he examined the activity of various drugs and drug combinations on *P. falciparum*. With the further development of close ties with the US Army, Lieutenant Colonel G. Dennis Shanks was posted by the Walter Reed Army Institute of Research (WRAIR) to AMRU in 1992 for 3 years. During that time he was involved with malaria field studies and provided clinical support and advice to the ADF regarding malaria. Also in 1992, Major Steve Frances replaced Major Edstein at AFRIMS with a 3-year exchange posting to identify insect repellents providing improved protection against mosquitoes

and chigger mites. Ms Barbara Kotecka and Major Robert Cooper continued their studies investigating the *in vitro* activity of antimalarial drugs and the distribution/speciation of anopheline mosquitoes, respectively.

During this quinquennium, various overseas investigators spent several weeks to years at AMRU and contributed significantly to malaria research activities. Sponsored by the World Health Organization, Australian International Development Assistance Bureau, or Rotary Against Malaria, they included Mr George Taleo (Vanuatu), Dr Li Xiuhong and Dr Tian Liping (China), Prof Bui Dai, and Dr Nhuyen Thi Nhu Mai (Vietnam). With the wholehearted support of AMRAB, the scope and impact of research activities were advanced substantially through collaboration and interaction with other national and international institutions (Table 1).

Table 1. List of institutions and individuals collaborating with the Army Malaria Research Unit.

Australian National University, John Curtin School of Medicine, Department of Medicinal Chemistry, Canberra, Australia. (Dr. G. Barlin)

Armed Forces Research Institute of Medical Sciences (AFRIMS), Bangkok, Thailand. (Dr T. Brewer, Dr. G. D. Shanks)

Australian International Development Assistance Bureau, Canberra, Australia. (Vietnam-Australia Malaria Control Project)

Center for Disease Control, Atlanta, USA. (Dr. W. Collins)

Gorgas Memorial Laboratory, Panama City, Panama. (Dr. O. Nicanor)

Jacobus Pharmaceutical Company, USA (Dr. D. Jacobus)

ICT Diagnostics, Sydney, Australia. (Dr. M. Garcia)

Ministry of Health and Medical Services, Honiara, Solomon Islands. (Dr. N. Kere)

National Health and Medical Research Council, Canberra, Australia. (Dr. G. Davis)

Ok Tedi Mining Limited, Tabubil, PNG. (Dr. P. Spicer, Dr. J. Schuurkamp)

Papua New Guinea Institute of Medical Research, Maprik, PNG. (Dr. B. Genton, Mr. K. Lorry)

Rotary Against Malaria, Sydney, Australia. (Dr. B. Hanley)

Solomon Islands Malaria Training and Research Institute, Honiara, Solomon Islands. (Dr. B. Bakote'e)

University of Sydney, Department of Biochemistry, Sydney, Australia. (Dr. R. Christopherson)

US Army Malaria Research Institute, Nairobi, Kenya. (Dr. G.D. Shanks)

US Naval Medical Research Unit No. 2, Jakarta, Indonesia. (Dr. K. Baird)

Walter and Eliza Hall Institute, Melbourne, Australia. (Dr. J. Reeder)

Walter Reed Army Institute of Medical Research, Experimental Therapeutics Division, Washington DC, USA. (Dr. W.K Milhous)

Wellcome Research Laboratories, Bangkok, Thailand. (Dr. N. White)

World Health Organization, Geneva, Switzerland

World Health Organization, Western Pacific Region, Manila, Philippines. (Dr. Matsushima, Dr. D. Parkinson, Dr. K. Palmer)

Malaria situation

Malaria continued to be a problem for Australians travelling overseas because of parasite resistance to an increasing number of antimalarial drugs.[7] The high incidence of malaria in Australian soldiers during 1988 and 1989 declined dramatically following the replacement of weekly pyrimethamine/dapsone by daily doxycycline (100 mg) at the start of the 1990-1995 quinquennium. During the deployment of almost 2,000 Australian soldiers to Cambodia, Somalia and Rwanda, only 8 soldiers developed malaria while on daily doxycycline prophylaxis.[8] This was probably related to inadequate compliance rather than parasite resistance to doxycycline. The antibiotic was usually well tolerated by soldiers taking it with food and protecting themselves from sunlight. Between 1-2% of soldiers were placed on weekly mefloquine (250 mg) prophylaxis because of gastrointestinal intolerance, sun-sensitisation, or contraindications precluding the use of doxycycline.

Although mefloquine was recommended widely as first-line malaria prophylaxis in drug-resistant areas, AMRU preferred the use of doxycycline. In addition to killing asexual blood stages, the tetracyclines acted against the pre-erythrocytic liver stages of *P. falciparum*,[9] a feature not shared by mefloquine. A further reason to use doxycycline was mefloquine's exceedingly long pharmacological half-life which would favour the development of parasite resistance to the drug if it were used extensively for malaria prophylaxis. In fact, it was already no longer able to suppress or cure falciparum malaria in some parts of Cambodia and Thailand. For example, during the deployment of various military contingents to the United Nations Transitional Authority in Cambodia (UNTAC), mefloquine was ineffective in protecting soldiers from countries other than Australia against falciparum malaria. On the other hand, only 2 of 500 Australians on doxycycline prophylaxis developed malaria during their deployment to Cambodia during 1992. Because far more malaria was observed in other contingents, prophylaxis with doxycycline, introduced in Cambodia by the Australian contingent, was later adopted as the standard malaria prophylaxis by UNTAC. The value of doxycycline was further proven by the low malaria attack rates observed by ADF personnel during deployments to Rwanda and Somalia.[8]

Activities

The emergence of chloroquine resistance in *P. falciparum* along the Thai-Cambodian border in the late 1950s led to world-wide efforts to develop alternative drugs for the prophylaxis and treatment of falciparum malaria.[10] In the meantime *P. vivax*, the other main species infecting humans, remained susceptible to chloroquine everywhere. However, the landmark discovery by AMRU in 1989 that *P. vivax* had developed resistance to chloroquine in PNG[4] was a reminder that parasites and vectors in Australia's neighbourhood could be appreciably different from those encountered elsewhere in the world. Furthermore, the large number of ADF soldiers who developed falciparum and vivax malaria in 1988/1989[6] indicated the need for more effective alternative drug regimens and improved measures to control and avoid contact with anopheline mosquitoes.

With this in mind, more and more of AMRU's activities during this quinquennium were focussed on addressing practical problems confronting the ADF. Research activities were usually performed under one of the following broad categories:

1. Drug resistance and diagnosis;
2. Drug development and evaluation; and
3. Vector control, biology and geographical distribution.

Drug resistance and diagnosis

Field assessment of drug resistance

Assessment of parasite resistance to drugs in Australian soldiers while deployed overseas or after return to Australia was proving difficult to carry out for a variety of operational reasons. However, in view of the rapid changes in patterns of parasite susceptibility to drugs, it was important to obtain information regarding the degree and extent of drug resistance from malarious areas of possible future deployment.[11,12] Not only could this be helpful in the formulation of more appropriate prophylactic regimens, but medical personnel in the field would be alerted to the possibility of treatment failures, thereby guiding them in the selection of alternative drug regimens. To meet these objectives, studies were initiated in collaboration with other institutions to determine the parasite susceptibility of parasites from different geographical areas using both established and novel procedures. The findings would of course also assist local health authorities in controlling malaria more effectively.

- Assessment of drug resistance needed for optimum prophylaxis and treatment

Drug resistance in PNG

Assessment of the drug resistance pattern in PNG was commenced in collaboration with the PNG Institute of Medical Research (PNGIMR) facility at Maprik, Sepik Province. Using the WHO *in vitro*

microtest,[13] about three-fourths of the isolates were found to be resistant to chloroquine, with one-fourth of them also being resistant to amodiaquine. Another significant finding was the greatly reduced susceptibility of about one-third of the isolates to pyrimethamine, indicating that a substantial number of non-immune patients were no longer being cured by the current standard treatment used in PNG, viz. 3 days of quinine and a single dose of sulfadoxine/pyrimethamine. On the other hand, the 10-fold greater parasite susceptibility to cycloguanil, the active metabolite of proguanil, supported earlier findings[5] that proguanil, used in combination with dapsone, should be evaluated for the treatment and prophylaxis of malaria. Subsequent examination of these specimens at the Walter and Eliza Hall Institute (WEHI) found no point mutations in dihydrofolate reductase (DHFR) and dihydropteroate synthetase (DHPS) genes in any of the 13 patients who were found to be sensitive to pyrimethamine and proguanil by the *in vitro* test.[14] The findings confirmed the value of the *in vitro* field test whenever local circumstances render it difficult or impossible to use highly sophisticated (and expensive) genotyping technology to determine the parasite susceptibility to antifolate drugs.

Parasite susceptibility to various drugs using the *in vitro* test[13] was also determined at Ok Tedi Mining Limited (OTML) located in a highland area of the Western Province of PNG. There was no malaria transmission in the township, but many employees, recruited from different areas of PNG, became ill with malaria after their 3-week annual vacation to their home provinces. In collaboration with the Medical Department of the company, AMRU was able to document the widespread prevalence throughout PNG of *P. falciparum* resistance to chloroquine, amodiaquine and pyrimethamine. Furthermore, *P. vivax* resistance to chloroquine was common, especially so in the island provinces. However, this species remained sensitive to amodiaquine, pyrimethamine and cycloguanil.[15]

- Widespread parasite resistance to standard antimalarial drugs in PNG demonstrated by the *in vitro* test

Field *in vitro* test using disposable environmental bag

In many malarious countries, specially-prepared gas mixtures needed to support the growth of parasites *in vitro* are not readily available, or are unaffordable, or the field incubators are too small to accommodate standard environmental chambers. These problems were partly circumvented in the WHO microtest kit by the use of candle jars in which pure paraffin candles are able to generate a suitably O_2-depleted and CO_2-enriched environment.[13,16] As the "candle-jar" was more bulky and expensive than other items in the kit, investigations were carried out to determine whether disposable plastic environmental bags, used by many hospital and pathology laboratories to culture other micro-organisms, might facilitate assessment of the drug susceptibility of malaria parasites. After examining various bags, the Becton Dickinson Bio Bag type C was found to be very satisfactory, with parasite maturation being comparable to that obtained in the specially-prepared gas mixture (5% O_2, 5%CO_2 and 90% N_2) and superior to that obtained in the candle-jar.[17]

- Disposable environmental bag used successfully for the *in vitro* test

Bioassay for field use

As was pointed out previously,[5] the bioassay developed at AMRU could be employed in various ways to complement drug analysis of serum/plasma specimens by HPLC, and it was also useful for assessing drug resistance under field conditions.

After earlier development of a bioassay for cycloguanil,[5] further investigations at AMRU produced bioassays for mefloquine,[18] artemisinin,[19] and chloroquine.[20] During more detailed studies with chloroquine, it became clear that the bioassay measured the total antimalarial activity of the parent drug and its active metabolites, desethylchloroquine and bis-desethylchloroquine, whereas HPLC analysis measures parent drug and metabolite concentrations.[20]

In collaboration with the Department of Health, Vanuatu and the Oxford Group, the bioassay was used in 1993 to determine whether observed treatment failures were due to true drug resistance or whether they were related to difficulties in administering chloroquine to children under field conditions[21] (young children dislike the bitter taste of chloroquine and, in understaffed clinics or hospitals located in endemic areas, it is often difficult to supervise adequate ingestion of the entire treatment course). The findings showed that most children who had a recurrence of parasitaemia after treatment also had much lower-than-average chloroquine plasma concentrations. This demonstrated the value of the bioassay, used in conjunction with the *in vitro* microtest, to exclude inadequate ingestion/absorption before ascribing treatment failures to drug resistance. A distinct advantage of the bioassay was that only small amounts of blood (usually finger-tip specimens) were required for assessment. Furthermore, this assay could be employed in many malarious areas where HPLC equipment, expensive reagents and specially-trained staff are often not available.

As part of the medical support to the Australian contingent of UNTAC, the bioassay was also used to determine chloroquine concentrations in plasma specimens obtained from 26 Australian soldiers on chloroquine plus doxycycline for malaria prophylaxis.[22] In addition, plasma chloroquine and doxycycline concentrations were monitored by HPLC analysis. Although the bioassay relies on the measurement of total antimalarial plasma activity, chloroquine equivalent concentrations could still be measured by the bioassay (with similar results to HPLC analysis) despite the presence of doxycycline in the plasma samples. The reason for this was that chloroquine is a rapidly-acting drug which inhibits schizont maturation during the first 30 hours of incubation (endpoint of the *in vitro* test), whereas therapeutic concentrations of doxycycline have a delayed effect upon parasite growth and maturation which only becomes apparent after 3 to 4 days of incubation.[23] This is in accord with doxycycline's mode of action against the *P. falciparum* apicoplast, with a loss of apicoplast function occurring late in the life-cycle with a delayed inhibition of protein synthesis. These results suggested that the bioassay could be used successfully in the field to estimate the concentration of rapidly-acting drugs when administered in combination with slower-acting ones.

- Bioassay used to determine drug resistance in Vanuatu and Cambodia

Non-microscopic malaria diagnosis

Definitive diagnosis of malaria can only be established by microscopic examination of blood films. However, in highly synchronous infections of *P. falciparum*, parasites are sometimes missed if blood is not examined during the first half of the 48-hour asexual blood cycle before they disappear from the peripheral blood circulation into the brain and other organs.[24] But more commonly, diagnostic problems are due to technical difficulties experienced in identifying parasites during routine examination of blood films, particularly at low parasite densities. Although this can be partly remedied by special training in malaria microscopy, the availability of a non-microscopic test would be a distinct advantage. In 1994, ICT Diagnostics developed the ICT Malaria Pf test card which used an immuno-chromatographic approach to capture Pf Histidine Rich Protein-2 antigen by antibodies specific for this antigen.

The first field evaluation of this test card was carried out by AMRU in collaboration with ICT Diagnostics, Sydney, and the Solomon Islands Malaria Training and Research Institute (SIMTRI), Honiara. After testing finger-tip specimens of blood from 251 symptomatic patients reporting to the outpatients clinic of the Honiara Central Hospital, the accuracy of this test proved to be good, with a sensitivity of 100% and a specificity of 96.2% for parasite densities exceeding 80/μL blood.[25] These findings suggested that this 6-minute card test might prove to be a simple and practical means of making a rapid diagnosis of falciparum malaria at all levels of health care. As a result, the card test was introduced for use in the ADF under circumstances in which a microscopic diagnosis would be difficult or unreliable, such as under field conditions where pathology support is limited.

- Diagnosis of falciparum malaria achieved without a microscope

Drug development and evaluation

The proven effectiveness of doxycycline in preventing malaria in Australian soldiers in PNG led to further field and laboratory investigations with this and other antibiotics. As doxycycline does not affect the tissue stages of *P. vivax*, further studies were carried out with primaquine and a potentially new alternative to primaquine – tafenoquine, in efforts to decrease the inordinately high number of soldiers returning to Australia with relapsing vivax malaria. Studies were also carried out with halofantrine, a new drug which had been developed primarily as a possible replacement for mefloquine in drug-resistant areas.

Meanwhile, the long-standing interest of AMRU in proguanil, used in combination with other drugs, continued unabated. Does proguanil metabolism affect its effectiveness in combination with dapsone? What about its combination with the new antimalarial - atovaquone? Might another antifolate – PS-15 (phenoxypropoxybiguanide) - be more potent than proguanil?

Investigations were also initiated with the artemisinins which appeared ready to replace quinine as the main drug for treating chloroquine- or amodiaquine-resistant falciparum malaria. In addition, further *in vitro* and *ex vivo* studies were carried out with Mannich base compounds to identify candidates with a greater antimalarial activity than amodiaquine or pyronaridine.

In carrying out these investigations, new *in vitro*, *ex vivo* and analytical methods and procedures were developed, and used alongside older ones, to assess drug activity against parasites with varying degrees of drug susceptibility. They were also used during pharmacokinetic studies to optimise drug regimens for malaria prophylaxis and treatment.

Investigations with antibiotics other than doxycycline

As already pointed out, doxycycline became the cornerstone for malaria prophylaxis during this period. Nevertheless, studies with other antibiotics were initiated because doxycyline cannot always be used, e.g. during pregnancy or early childhood, and because parasites may eventually become resistant to it. When multidrug-resistant parasites were incubated *in vitro* with various antibiotics for 96 hours (rather than 30 or 48 hours), inhibitory concentrations were similar to plasma drug concentrations observed after *in vivo* administration of chloramphenicol,[26] ciprofloxacin,[27] or azithromycin.[23] For example, the mean minimum inhibitory concentration (MIC) of azithromycin was reduced from 6.2 μg at 48 hours to 0.08 μg at 96 hours after the start of incubation. Since peak and trough serum concentrations after 500 mg azithromycin were about 0.4 and 0.04 μg, respectively, and serum protein binding was low, courses of azithromycin treatment lasting 3-4 days might be sufficient to cure malaria infections. However, because azithromycin (and other antibiotics) clear parasites slowly, it would probably have to be taken in combination with a rapidly-acting, non-curative blood schizontocide, such as artesunate, to quickly abort an acute attack of malaria. In addition to its potential value for treating malaria infections, this drug might also prove to be useful for preventing drug-resistant malaria.

- Short azithromycin course might cure falciparum malaria

Doxycycline and primaquine combination for causal prophylaxis

Doxycycline acts against blood stages of both *P. falciparum* and *P. vivax* but primaquine is needed to eliminate residual hypnozoite liver stages of *P. vivax*. Co-administration of daily doxycycline (100 mg) and primaquine (22.5 mg) for 14 days was required to prevent both falciparum and vivax malaria after leaving malarious areas. For various reasons, compliance with this lengthy "eradication course" was not optimal. But even with good drug compliance, an increasing number of individuals were developing vivax malaria after returning to Australia, presumably because the hypnozoite liver stages were becoming increasingly tolerant to primaquine.

Earlier observations in ADF personnel deployed to PNG in 1988/1989 had indicated that doxycycline, in combination with a low dose of primaquine, might be effective in killing the pre-erythrocytic liver stages of both *P. falciparum* and *P. vivax*.[5] If such causal prophylactic activity could be confirmed, this would be operationally very significant because doxycycline and primaquine would have to be taken for only a few days, instead of 14 days, after leaving a malarious area. Although the tetracyclines, such as doxycycline, are able to kill pre-erythrocytic liver stages of *P. falciparum*, they have little or no activity against the hypnozoite liver stages of *P vivax*.[9] Based on earlier observations with rhesus monkeys infected with *P. cynomolgi*,[28] it was hoped that a low dose of 7.5 mg primaquine, administered together with 100 mg doxycycline, might be able to exert sufficient synergistic drug activity against the pre-erythrocytic liver stages of *P. vivax* to enable prophylaxis to be discontinued within a few days after return to Australia.

The deployment of a 53-man detachment of 3rd Combat Engineer Regiment to PNG in 1993 for a 42-day road and bridge building exercise provided a good opportunity to verify the effectiveness of such a drug combination in a heavily malarious area of the country.[29] Malaria was not observed when men took a daily dose of 100 mg doxycycline and 7.5 mg primaquine while in PNG and for 3 days after return to Australia, despite intense exposure to malaria. However, 2 of the soldiers taking this drug combination developed falciparum malaria within 3 weeks after return and 7 of them developed vivax malaria between 3 and 16 weeks after leaving PNG. This occurred despite adequate doxycycline plasma concentrations. Clearly, discontinuation of this prophylactic regimen 3 days after return to Australia could not be relied upon to suppress all pre-erythrocytic parasites in some individuals who had been exposed to heavily-infected mosquitoes.[29] Consequently, 14-day courses of doxycycline and primaquine would still need to be taken after leaving an endemic area until less cumbersome post-exposure courses became available.

- Short courses of doxycyline and primaquine are unable to replace cumbersome 14-day courses for malaria eradication

Tafenoquine as a possible replacement for primaquine

Tafenoquine (also known as WR238605 and Etaquine) was a new 8-aminoquinoline drug being developed as an alternative to primaquine by the Walter Reed Army Institute of Research (WRAIR), Washington, DC, USA. Preliminary results at AMRU showed that tafenoquine had a relatively low level of intrinsic blood stage activity and acted slowly *in vitro* against *P. falciparum* and *P. vivax*. Slow clearance of parasitaemia was also observed after administration of a 3-day course of tafenoquine to six *Aotus* monkeys infected with the chloroquine- and pyrimethamine-resistant AMRU 1 strain of *P. vivax*.[30] Because of the paucity of *Aotus* monkeys at

AMRU, collaborative studies were undertaken with Gorgas Memorial Laboratory in Panama where a larger group of *Aotus* monkeys could be inoculated with the AMRU 1 isolate of *P. vivax*.[31] The studies confirmed that tafenoquine was a slow-acting blood schizontocide which persisted in the body for much longer than primaquine. Although 3mg/kg was not curative, the addition of 30 mg/kg chloroquine (well-below curative dosage) usually cured monkeys of their infections. However, identical dosages of primaquine combined with chloroquine failed to cure any monkeys. The findings indicated that these drug combinations were more effective than either drug used alone and that tafenoquine was far more effective than primaquine in achieving clearance of parasitaemia and curing blood-induced vivax infections.

Since the main value of tafenoquine would be to replace primaquine as a tissue schizontocide (active against liver stages of *P. vivax*), studies were initiated to determine whether the AMRU 1 strain could be transmitted via mosquitoes (*Anopheles farauti*) to either *Aotus* or *Saimiri* monkeys. Although early results were encouraging,[5] too few monkeys were available at AMRU to carry out definitive tissue schizontocidal studies. After consultation with Dr William Collins at the Center for Disease Control in Atlanta, the parasite isolate was forwarded to him, resulting in the routine transmission of this first chloroquine-resistant isolate of *P. vivax* between mosquitoes and monkeys in his large simian colony.

- Tafenoquine has a much longer half-life and greater blood schizontocidal activity against *P. vivax* than primaquine

Halofantrine for prevention of falciparum malaria

Halofantrine, a phenanethrenemethanol drug initially developed by WRAIR, was used successfully for treating drug-resistant falciparum malaria starting in the late 1980s. As the pharmacokinetics of halofantrine had mostly been studied in healthy volunteers, a collaborative study was carried out, in collaboration with SIMTRI, to determine the disposition of halofantrine and its principal metabolite, N-desbutyl-halofantrine after treatment of 6 adult Melanesian patients infected with falciparum malaria. All patients responded well to treatment with 500 mg halofantrine given 3 times at 6-hourly intervals, indicating that the observed pharmacokinetic parameters, such as maximum plasma concentrations and elimination half-lives of halofantrine and N-desbutylhalofantrine, were adequate to achieve good therapeutic outcomes.[32]

By the early 1990s, however, halofantrine was failing to cure falciparum malaria in Thailand.[33] Although taking it with food increased the absorption of the drug, about half of the patients were not cured after treatment.[34] But as with many other drugs, halofantrine was still effective in areas outside of southeast Asia. In French troops deployed to Central Africa for 4 months, a short course of halofantrine at the end of deployment prevented soldiers from developing malaria after return to France.[35]

The role that post-exposure treatment with halofantrine might play in preventing malaria was further investigated in PNG. In collaboration with Ok Tedi Mining Ltd located in a non-malarious highland area of the Western Province, 345 copper miners received a 3- or 6- day course of halofantrine after returning to the mine from 2-4 weeks home leave in various rural areas of the country. None of the men receiving either regimen of halofantrine developed falciparum malaria, whereas 36% of accompanying family members developed malaria within 1 month after return.[36] Although halofantrine cannot be used for long-term prophylaxis and has been largely superseded due to cardiovascular adverse events, the PNG findings suggested that individuals exposed to malaria for a short period (preferably shorter than the malaria incubation period) might not develop symptomatic malaria if an effective treatment course of halofantrine is administered immediately after leaving an endemic area.

- Falciparum malaria is suppressed by halofantrine administered after short visits to malarious areas

Proguanil conversion to cycloguanil affects synergy with dapsone

The marked synergism between proguanil and dapsone, observed during earlier studies,[5] led to further *in vitro*, *ex vivo* and *in vivo* investigations using both drugs. Proguanil, a biguanide prodrug, is metabolised by hepatic enzymes to the active triazine metabolite, cycloguanil. Synergism occurs between cycloguanil and dapsone with the triazine inhibiting the malarial parasite enzymes DHFR and the sulfone inhibiting DHPS, resulting in inhibition of folate metabolism. In addition to confirming that cycloguanil, not proguanil, was the main determinant of antimalarial activity,[37] studies at AMRU showed that cycloguanil's activity could not be potentiated by combining it with another DHFR inhibitor – pyrimethamine.[38] Furthermore, the pharmacokinetics of dapsone were not altered when administered in combination with proguanil in healthy volunteers.[39]

Additional investigations were also carried out to determine whether inter-individual variability in the oxidative activation of proguanil to cycloguanil could influence the effectiveness of malaria prophylaxis

or treatment. Was the poor protection to proguanil/dapsone prophylaxis in Thai soldiers solely due to a high level of parasite resistance to antifolate drugs or was poor metabolic conversion from proguanil to cycloguanil a contributing factor? The latter could have been partly responsible because 23% of 207 Thai soldiers were classified as poor metabolisers (PMs) of proguanil, resulting in low plasma concentrations of cycloguanil.[40] By contrast, all 25 Australian soldiers receiving daily 200 mg proguanil and 8 mg dapsone during a short-term deployment to Thailand were extensive metabolisers (EMs) of proguanil, and this may have contributed to their protection against malaria.

A further collaborative study with AFRIMS showed that Thai patients had a limited ability to convert proguanil to cycloguanil, even at a very high dose of proguanil.[41] Additional *in vitro* and *ex vivo* studies with cycloguanil-resistant isolates of *P. falciparum* suggested that low plasma cycloguanil concentrations, coupled with high levels of drug resistance, limited the value of proguanil plus dapsone in Thailand.[42] However, this drug combination might still be useful in other ethnic groups who were better able to biotransform proguanil to cycloguanil, especially in areas with a greater parasite susceptibility to cycloguanil.

This was demonstrated by collecting blood samples from Australian soldiers on proguanil/dapsone prophylaxis and incubating their sera with parasites previously obtained from Thai patients who had not been cured by proguanil/atovaquone or proguanil/dapsone. Parasite growth was inhibited at plasma dilutions of 4, 8 and 16, indicating that Australian soldiers on proguanil/dapsone prophylaxis, most of whom are EMs of proguanil, would probably be protected if exposed to parasites with a drug susceptibility similar to these isolates from Thailand.[42]

- Proguanil/dapsone synergistic activity is affected by the ability to biotransform proguanil to cycloguanil

Proguanil plus Atovaquone – a new antimalarial drug combination

Atovaquone, a novel hydroxynaphthoquinone which inhibits pyrimidine biosynthesis in parasite mitochondria, was developed primarily for treating *Pneumocystis carinii* and *Toxoplasma gondii* in HIV-infected patients. It was also shown to abort clinical attacks of malaria, but about 30% of patients had a recrudescence of parasitaemia.[43] When proguanil was combined with atovaquone, cure rates above 90% were obtained.

Unlike dapsone, atovaquone was potentiated *in vitro* by proguanil to a much greater degree than by its cycloguanil metabolite.[44,45] Studies at AMRU also showed that the MIC of atovaquone was considerably higher in cultures containing 50% serum than in those containing 10% serum. Presumably this was due to the substantial binding of atovaquone to plasma proteins, with less "free" drug being available to inhibit parasite growth (similar findings were observed with pyrimethamine, well-known for its high protein binding to plasma of about 94%). By contrast, proguanil and cycloguanil had similar MIC values at high and low serum concentrations. These findings were a reminder that chemical analysis of "total" drug concentrations does not necessarily equate to the "free" drug available for killing parasites.

As part of a collaborative effort with AFRIMS and Wellcome Research Laboratories in the United Kingdom, AMRU determined the pharmacokinetics of proguanil in Thai patients receiving either proguanil alone or in combination with atovaquone. Proguanil and cycloguanil plasma concentrations were similar in the two groups, with the group that received the drug combination showing slightly higher values for proguanil and its metabolite.[46] In a further study involving Thai malaria patients treated with proguanil plus atovaquone, *ex vivo* antimalarial plasma activity (measured by bioassay) was similar in PM and EM patients, indicating that an individual's ability to metabolise proguanil did not appear to affect the *in vitro* antimalarial activity of this drug combination. This suggested that the phenotypic status of individuals was of little importance in determining the outcome of treatment with proguanil plus atovaquone and that, unlike proguanil plus dapsone, the synergistic activity of this combination was determined primarily by proguanil rather than by that of its metabolite.[46]

In collaboration with the Department of Biochemistry at the University of Sydney, studies were also conducted to investigate the effects of pyrimidine antagonists, such as atovaquone, on the third and fourth steps of the pathway for the de-novo biosynthesis of pyrimidine nucleotides.[47,48] The results indicated the need for further studies to understand the regulation of nucleotide metabolism and the mechanism of action of pyrimidine antagonists in preventing DNA synthesis in the parasite.

- Proguanil/atovaquone synergistic activity is not affected by the ability to biotransform proguanil to cycloguanil

Other potential atovaquone drug combinations

In view of the above findings, a further study was started to determine whether the addition of

dapsone might enhance the synergistic activity between proguanil and atovaquone by potentiating the activity of proguanil's metabolite – cycloguanil. Preliminary results seemed to indicate that this might be the case, raising the possibility that two synergistic actions – that of proguanil/atovaquone and cycloguanil/dapsone - in a triple combination of proguanil, atovaquone and dapsone might enable the dosage of the component drugs to be reduced, thereby possibly reducing potential adverse drug reactions.[49] Additional *in vitro* and *in vivo* studies involving *Saimiri* monkeys showed that folinic acid did not reverse antifolate activity against *P. falciparum*, suggesting that it could be used, if necessary, to prevent bone marrow depression.[50,51]

So far, these various studies with atovaquone had demonstrated synergism between this drug and inhibitors of the folate pathway. How might doxycycline, already the main drug used for malaria prophylaxis, interact with atovaquone? As both drugs depress the activity of dihydroorotate dehydrogenase - suppressing *de novo* pyrimidine synthesis in the parasite - they might potentiate each other's antimalarial activity. In fact, when serum samples from 52 Australian soldiers on doxycycline prophylaxis were incubated *in vitro* with atovaquone, this proved to be the case, indicating that these drugs were rational partners for malaria prophylaxis or treatment.[52,53]

- Atovaquone activity is increased by combining this new drug with proguanil/dapsone or doxycycline

A new generation antifolate: PS-15 and its metabolite (WR99210)

WR99210, a potent diamino triazine developed by WRAIR during the early 1970s, was shown to be remarkably effective against multidrug-resistant strains of *P. falciparum*.[54] Unfortunately, severe gastrointestinal reactions and poor bioavailability during subsequent clinical trials precluded the further development of this antifolate drug. Nevertheless, interest in WR99210 was maintained, particularly because of its lack of cross resistance with other antifolates, such as pyrimethamine and cycloguanil.

Recently, PS-15 (also known as WR250417) had been developed as a prodrug for WR99210.[55] The concept for its design was based on the conversion of proguanil to its active metabolite, cycloguanil. It was hoped that PS-15 would circumvent the intolerance and poor bioavailability seen with WR99210, but still maintain potent antimalarial activity.

In collaboration with the Jacobus Pharmaceutical Company, Princeton, USA, AMRU administered PS-15 and WR99210 to non-infected *Saimiri sciureus* monkeys and subsequently used HPLC analysis to determine serum drug concentrations[56] and the bioassay to determine the serum *ex vivo* activity against *P. falciparum*. Previous studies had shown that these monkeys provided excellent preliminary information about the degree and duration of serum antimalarial activity of promising experimental drugs because of the ability of their serum to support the growth of *P. falciparum in vitro*.[57] After drug administration, substantial and sustained serum antimalarial activity was observed in monkeys receiving PS-15, but not in those receiving WR99210 (probably due to poor bioavailability).[58] Although PS-15 had some intrinsic antimalarial activity, the results indicated that WR99210 was primarily responsible for the antimalarial activity observed after PS-15 administration.

As a result of these observations, and the lack of any observable gastrointestinal toxicity with PS-15, *in vitro* studies were carried out which compared the activity of various antifolate compounds against six multidrug-resistant isolates or clones of *P. falciparum*.[59] They showed that WR99210 had a complete lack of cross-resistance to other antifolates and that, if humans were able to tolerate, absorb and metabolise PS-15 equally as well as *Saimiri* monkeys, it should be a prime candidate for further development as an antimalarial drug. Furthermore, marked synergistic activity was observed *in vitro* between the metabolite WR99210 and dapsone or sulfamethoxazole, but not atovaquone.[45] When PS-15 was co- administered with sulfamethoxazole to *Saimiri* monkeys, serum antimalarial activity (after biotransformation from PS-15 to WR99210) was more pronounced than in monkeys receiving PS-15 alone. But at 24 and 48 hours there was little difference in activity, probably because most of the sulfonamide had been eliminated from the blood circulation.[60] Unlike the *in vitro* findings with WR99210, the activity of PS-15 was not potentiated by dapsone, but it was potentiated by atovaquone.[45] This suggested that the synergistic interactions of proguanil and PS-15 may be similar: namely antimalarial activity of parent drugs potentiated by atovaquone, and that of their triazine metabolites potentiated by dapsone or sulfamethoxazole.

Ex vivo studies were also performed to compare the *in vitro* antimalarial activity of serum samples obtained from *Saimiri* monkeys treated with PS-15 and proguanil triple drug combinations. The results showed that the synergistic activity of PS-15/atovaquone/dapsone was greater than for proguanil/atovaquone/dapsone.[49] Significantly, folinic acid did

not reverse the antimalarial activity of these drug combinations.[48,49] In view of these findings, it was felt that further studies were needed to assess the value of this antifolate class of biguanide precursors for the prevention and treatment of multidrug-resistant falciparum malaria.

- PS-15 (and metabolite) is far more potent than proguanil (and metabolite), either alone or in combination with atovaquone and/or dapsone

Artemisinin compounds – pharmacokinetic information needed

Artemisinin (qinhaosu) is derived from *Artemisia annua*, (qinghao), a medicinal plant used extensively in China as a febrifuge for hundreds of years. As artemisinin and its derivatives, such as artesunate and artemether, act more rapidly than quinine (and other drugs) in aborting acute attacks of malaria, increasing consideration was being given to using them to treat acute infections of drug-resistant malaria. However, recrudescence rates of 20-50% were observed when mainly empirical regimens of these drugs were given for less than 5 days. Clearly, more information was needed on the pharmacokinetic properties of the artemisinins to support clinical studies aimed at improving treatment regimens.

Chemical assays were proving to be rather insensitive and unreliable because these drugs were thermally labile and lacked an ultra-violet or fluorescent chromophore. In a collaborative study with WRAIR, plasma samples were analysed at AMRU using an HPLC method incorporating reductive electrochemical detection. Although artemisinin, artesunate and dihydroartemisinin (the principal metabolite of artesunate) could be detected at concentrations as low as 10 ng/mL, this method was too cumbersome for routine drug analysis, with the need for rigorous deoxygenation of samples and the mobile phase.

The bioassay appeared to offer a more accurate (detection limit down to about 1 ng/mL) and a considerably cheaper alternative for use in pharmacokinetic studies, although it obviously could not differentiate between serum concentrations of parent compounds and their metabolites.[19] By measuring total antimalarial serum activity, preliminary pharmacokinetic information was obtained about the degree and duration of artemisinin and its derivatives after their administration to *Saimiri* monkeys. The results indicated that the artemisinins were more potent but were eliminated more rapidly than standard antimalarial drugs. Significantly, antimalarial activity was reduced when serum samples were incubated at higher erythrocyte concentrations, suggesting that the artemisinins, similar to chloroquine, were selectively concentrated by erythrocytes. In view of these findings, the bioassay might play a significant role during clinical evaluation of alternative artemisinin therapeutic regimens.

- Bioassay provides a sensitive method for use during pharmacokinetic evaluation of various artemisinin compounds

Figure 2. Dr Barbara Kotecka preparing cultures to assess the in vitro activity of drugs against Plasmodium falciparum

Mannich bases with greater activity than amodiaquine or pyronaridine

Early studies with amodiaquine[61] and pyronaridine[62] had shown these two Mannich base drugs to have greater antimalarial activity than chloroquine or pyrimethamine.

Since parasites were becoming increasingly tolerant to both Mannich bases, the search for more potent compounds in this class[5] was continued as part of an ongoing collaborative effort with Dr Gordon Barlin at the Australian National University, Canberra.[63-70] Apart from *in vitro* screening by standard morphological and radioisotopic methods, a new visual technique was developed and used successfully to screen 44 compounds.[71]

Ten of the most active compounds were administered to *Saimiri* monkeys as a single dose and their antimalarial serum activity over the next 7 days was compared with that of amodiaquine and pyronaridine.[72] Four of the compounds had a greater and more prolonged *ex vivo* activity than pyronaridine against the standard K1 isolate. They were also similarly active against four other drug-resistant strains of *P. falciparum*, one of which was resistant to mefloquine and halofantrine, and the other resistant to atovaquone. The marked degree and duration of activity after a single dose of these drugs indicated the need for further studies in

infected *Aotus* monkeys to determine the potential value of this class of compounds for the prophylaxis and treatment of multidrug-resistant malaria.

These findings also provided further evidence of the value of the *ex vivo* monkey model for the development of antimalarial drugs. Furthermore, by using non-infected monkeys for obtaining preliminary information about potential antimalarial drugs, *in vivo* evaluation could be restricted to those drugs showing the greatest promise for further development. Since evaluation of drugs in infected *Aotus* monkeys has significant ethical and economic implications, any experimental approaches minimising the use of such monkeys deserved strong support.

- Visual test developed for the *in vitro* screening of antimalarial drugs
- Some Mannich bases are substantially more active *ex vivo* than amodiaquine and pyronaridine

Vector control, biology and distribution

Insect repellents

Deet – applied to skin. The topical application of mosquito repellents was (and continues to be) an important means of protecting oneself against malaria. As the current ADF repellent (95% deet) was not popular with soldiers in the field,[5] laboratory and field tests were conducted in Thailand during 1992-1993 to evaluate protection provided by novel repellents and other formulations of deet.

Early on, it was established that deet (diethylmethylbenzamide) and dimethylphthalate provided shorter protection against *Anopheles dirus*, an important malaria vector in Thailand, than against *Aedes albopictus*.[73] With this background information, the efficacy of deet was compared with two experimental chemicals developed by the US Army - AI3-37220 and CIC-4. Using 25% ethanol solutions of each repellent, *An. dirus* was more susceptible to CIC-4 than either deet or AI3-37220 in the laboratory. However, a field study in Chantaburi province, southeastern Thailand, showed that AI3-37220 provided significantly better protection (>95% for 4 hours) than the other two repellents (<95% after 2 hours).[74]

In a further study, different deet formulations were evaluated in Sisaket province, northeastern Thailand. An applicator stick containing 33% deet provided >87.1% protection against *Culex* sp. for 5 hours, and 50% deet provided >93.3% protection against *Anopheles* spp. for 8 hours. By comparison, the US Extended Duration Repellent Formulation containing 33% deet provided complete protection for 6 hours against *An. dirus* and *Cx. vishnui*. Although similar levels of protection were observed with 50 and 75% deet in ethanol, 25% solutions were less effective.[75]

The repellent trials in Chantaburi and Sisaket provinces also presented the opportunity to examine *Anopheles* mosquitoes from untreated participants for the presence of *Plasmodium* circumsporozoite (CS) proteins using an ELISA technique. *P. vivax* CS protein was detected in 3.4% (2/54) of *An. karwari* and 4.8% (2/42) of *An. barbirostris*, but *P. falciparum* CS protein was detected in only 0.3% (1/276) of *An. dirus*. This suggested that *An. barbirostris* may play a role in transmission of *P. vivax* in Chantaburi Province, Thailand.[76]

- Deet and other repellents provide good protection against malaria vectors in Thailand

Permethrin – impregnated in uniforms and bednets.

Following earlier work with permethrin-impregnated uniforms and bednets,[5] further studies revealed that the knockdown and mortality of *An. farauti* and *Ae. aegypti* were similar following 15-180 seconds exposure to permethrin treated bednets and polyester/cotton fabrics.[77] Despite the presence of permethrin (0.007-0.068 mg/cm^2) in bednet material, 18-50% of *An. farauti* s.s. adults successfully blood fed through the nets and, although all were knocked down after 60 minutes, up to 78% recovered within 24 hours. The results indicated that this important vector of malaria could obtain a bloodmeal through treated bednet material and they emphasised the importance of avoiding contact with bednets while sleeping.

Collaborative studies with the US Department of Agriculture, Agricultural Research Service, Gainesville, Florida, showed that permethrin persisted in uniforms and provided good protection after 5 gentle cold water washes. These findings corroborated recommended guidelines of re-treating after 3-4 washes. As a result, AMRU recommended the use of permethrin treated uniforms, and this protective measure was instituted by the ADF in the early 1990s.[78]

In 1992 a joint trial was conducted with AFRIMS in northeastern Thailand to compare the incidence of malaria in Thai soldiers whose uniforms, covering both arms and legs, had received a high-pressure spray of permethrin with those whose uniforms had not been sprayed.[79] Bioassays of treated clothing, using laboratory reared *An. dirus*, showed permethrin remained in treated clothing for up to 90 days. Both permethrin-treated and untreated fabric provided >84% protection from biting *An. dirus* for the duration of the study. However, the impregnated

uniforms did not provide protection against malaria in this highly malarious area of Thailand. This was probably related to soldiers wearing casual attire (shorts and T-shirts) after work hours, exposing them to vector mosquitoes when they were at their most active.

During this trial, the prevalence and incidence of antibodies to *Orientia tsutsugamushi*, the aetiologic agent for scrub typhus, was also tested in these soldiers.[80] The point prevalence of antibodies varied from 0 to 4.1%, which was low compared to other regions in Thailand. An increased incidence of antibodies was observed during the wetter months of the year, indicating an increased risk for local and foreign military personnel.

- Permethrin treatment of uniforms and bednets is effective against mosquitoes. Limitations in the use of impregnated uniforms against malaria in Thailand

"Chiggers"

Previous work at AMRU with chiggers, insect vectors of scrub typhus (*Orientia tsutsugamushi*),[5] was continued during Major Frances' assignment at AFRIMS in Thailand from 1992 onwards. Following the establishment of a laboratory colony of *Leptotrombidium deliense* naturally infected with *O. tsutsugamushi* at AFRIMS,[81] the development and persistence of antibodies to *O. tsutsugamushi* in laboratory rats and mice could be observed.[82] In addition, it allowed investigation of the vertical[83] and horizontal[84] transmission of this pathogen. The observed novel transmission of *O. tsutsugamushi* to co-feeding mites could possibly explain the presence of rickettsiae in trombiculid genera not considered to be the main vectors of this organism.[85] Subsequent field studies in Nonthaburi Province (near Bangkok) showed that, contrary to findings with laboratory mice, rats were good sentinels, with chiggers attached to 44% of 202 rats and 2 rodents developing infections with *O. tsutsugamushi*. Out of a total of 314 engorged *L. deliense* obtained from sentinel rats, 2 (0.6%) were naturally infected with *O. tsutsugamushi*. This study also showed that the risk of exposure to *O. tsutsugamushi* is greater during the wet season, and that only a relatively small number of chigger attachments are needed to infect potential hosts.[86] A further field study in Phitsanulok Province, central Thailand, in 1993, showed that, contrary to expectations, *Blankaartia acuscutellaris* was not a vector of scrub typhus.[87]

Insect repellent studies against two vectors of scrub typhus – *L. deliense* and *L. fletcheri* - showed that low concentrations of permethrin, dimethylphthalate, deet, benzyl benzoate, di-n-propyl 2-5 pyridine-dicarboxylate, AI3-37220, 2-hydroxymethyl-cyclohexyl acetic acid lactone and a high concentration of dibutylphthalate (DBP) were toxic for uninfected larvae of both species. The median knockdown times for all chemicals were longer for *L. deliense* infected with *O. tsutsugamushi* than uninfected larvae. The study indicated that low concentrations of test chemicals, except DBP, should be effective against two important vectors of scrub typhus in southeast Asia.[88]

- Ecology of scrub typhus vectors and evaluation of repellents in Thailand

Distribution of Anopheline mosquitoes in northern Australia

The work on the anophelines in northern Australia was completed in 1991 with a survey of the Torres Strait, an area of scattered islands lying between Cape York and Western Province of PNG. While the mainland of Australia is malaria free, outbreaks of the disease regularly occur throughout these islands because people bring malaria parasites with them when moving between the coastal villages of Western Province and the Australian islands (Boigu and Saibai are only 7 km off the coast of Western Province). During the survey of the Torres Strait conducted in April-May 1991, *Anopheles farauti* 1 and *Anopheles hilli* were found on the larger islands of Boigu, Saibai, Badu, Moa, Horn, and Prince of Wales. Both species have been known to transmit malaria in Australia. Their larvae tolerate brackish water, an adaptation which would support their dispersal through these island groups.

Retrospective spatial analysis of potential malaria vectors, surveyed in northern Australia between 1985-1990,[5] showed that the three sibling species of *Anopheles farauti* were distributed around the coast and 50-100 km inland north of 20° S latitude and east of 129° E longitude. In some areas the three species were found together but the overall patterns of occurrences for each species were different.[89-91] These apparent dissimilarities in realised distribution suggested that there might be differences in ecological factors influencing the range of the individual species. This was investigated with the use of computer based Geographical Information Systems (GIS) data sets and the ecological niche modelling (ENM) software GARP (Genetic Algorithm for Rule-set Prediction). The prediction of species range by GARP uses point localities where species are known to occur together with environmental data for the geographic region of interest. Inputs for model construction included species occurrence data from field surveys together with high resolution environmental information for northern Australia

based on historical climate records. Two data mining software packages, CART (classification and regression tree analysis) and KnowledgeSeeker, were selected to search for significant environmental factors associated with species presence. The overall objective was to identify the key environmental factors responsible for defining the geographical ranges of the different vector species, as such factors are of epidemiological significance for malaria control.

The results revealed consistent agreement in the variables ranked by both data mining methods. This permitted the selection of reduced sets of environmental data to develop GARP models for the three target species with equivalent predictive accuracy to those which used all of the environmental information. Atmospheric moisture was shown to be a key predictor for all three species of *Anopheles farauti* in Australia.[91]

However, the GARP results were less satisfactory for describing the realised distribution of *Anopheles farauti* s.s. The distribution of this important vector of malaria in the Southwest Pacific Region includes New Guinea, the Solomon Islands and Vanuatu, as well as northern Australia. Systematic surveys in Australia's Northern Territory and Queensland have indicated that its distribution is predominantly within 5 km from the coast.[92] This species utilises brackish water sites for larval development, but larvae have also been found in freshwater environments,[93] suggesting that its coastal distribution is determined by environmental factors other than simple proximity to the sea. But the GARP models were unable to identify combinations of environmental parameters that successfully delimited the coastal distribution of this species.

Further analyses were undertaken with a novel exploratory method which permits visualisation and analysis of environmental gradients across distribution boundaries. The use of this range boundary tool to investigate environmental variable changes across the known range boundary limits of *An. farauti* s.s. in northern Australia identified the importance of elevation. The inclusion of this topographical variable resulted in models which included all of the record sites of this species in northern Australia and successfully reconstructed its narrow coastal distribution.[94]

- Environmental factors are shown to influence the distribution of *Anopheles farauti*.

Distribution and Speciation of Anopheles mosquitoes in Papua New Guinea.

Following on from Operation Anopheles in northern Australia, similar mosquito surveys were conducted in PNG. This highly malarious country has close ties with Australia and, historically, was administered by it until gaining independence in 1975. During training exercises in PNG, Australian soldiers are at a high risk of acquiring malaria when exposed to malaria vectors.[6] Although the primary vectors were considered to be members of the *An. punctulatus* group - *An. farauti* 1, *An. punctulatus* and *An. koliensis*, very little was known about their distribution or what other species might be present throughout PNG.

Faunal surveys were conducted in Western Province (1992), the Sepik region (1993), Gulf Province (1994), and Madang Province (1995). As this vast region is sparsely populated and has a limited road network, the work could only be accomplished with the support provided by crew and helicopters from Army Aviation's 162 Reconnaissance Squadron.[5] Excellent support was also provided by Preventive Medicine personnel from the PNG Defence Force.

These surveys relied heavily on the use of dry ice (CO_2) baited light traps to attract and collect adult mosquitoes. Up to 10 traps were set up each evening and retrieved the next morning, thus enabling a wide area to be surveyed with much less effort than the laborious human landing catches. As dry ice was unavailable, it had to be made from CO_2 gas cylinders. Because of the inaccessibility of field sites, this survey would not have been possible without Caribou aircraft from 25 Squadron pre-positioning these cylinders at various sites. Towards the end of the survey, another potential mosquito attractant – octenol - was being trialled for collecting culicine mosquitoes for arbovirus surveillance. As it appeared to have distinct advantages over CO_2 for use in remote locations, a trial was conducted during 1995 in the Madang area comparing different trapping methods for various species of anopheline mosquitoes. While octenol alone attracted more mosquitoes than light alone, it lured fewer mosquitoes into its traps than those with CO_2 alone or CO_2 plus octenol.[95] Continued reliance on dry ice for attracting anopheline mosquitoes appeared to be necessary.

Collections of larvae and adults were processed in the same way as described previously.[5] Initially, DNA probes were only available to identify the sibling species of *An. farauti*: *An. farauti* 1, *An. farauti* 2, and *An. farauti* 3.[96] However, by 1994, DNA probes had been developed for all the known members of the *An. punctulatus* group and were made available to support this work.[97]

Anopheles farauti 1 was found to be a dominant coastal species throughout the survey area. Preferred larval habitats were coastal swamps and

lagoons where the flow of streams is blocked by sand bars. Such breeding sites are ubiquitous along the coastline of PNG, their large size allowing huge population densities which were reflected in adult trap collections exceeding 7000 mosquitoes per night. *An. farauti* 1 is usually the primary vector of malaria in PNG, but it will also feed on domestic animals (pigs and dogs). In these surveys it was often found well outside its flight range of humans and domestic animals, indicating that this species also feeds upon native birds and animals.

Anopheles farauti 2 occurred throughout the surveyed region and it was the dominant species in Western Province. Significantly, this species had previously not been recorded to be present in PNG.[98] Although it was most abundant in inland lowland river valleys and flood plains, it was also found breeding along the coast and up to 1500 m in highland areas. This species was also found in human-made sites, such as wheel ruts and earthen drains. Although it was observed biting humans, nothing is known of its possible role in malaria transmission.

Anopheles farauti 3, *An. meraukensis* and *An. novaguinensis* were recorded from only a few localities in Western Province. In this region the climate tends to be different from the rest of PNG - it is monsoonal with distinct wet and dry seasons similar to northern Australia where these species are common. Larval habitats included swamps and the edges of rivers and streams, though it was also found in smaller ground pools. This is the first record of these species in PNG and nothing is known about their behaviour. However, due to their limited distribution and low numbers, they are unlikely to play a role in malaria transmission.

Anopheles farauti 4 larvae were commonly found in small ground pools to large swamps of inland river valleys and flood plains north of the central highlands. Nothing is known about the malaria transmission potential of this species.

An. punctulatus and *An. koliensis* are considered to be primary vectors of malaria in PNG. Both species were abundant throughout the survey region in inland, lowland river valleys and flood plains. *An. punctulatus* was also commonly collected in the highlands >1000 m. Larvae of both species were found breeding in natural pools of water, but the highly invasive *An. punctulatus* was also breeding in small pools created by human or animal activity, such as foot or hoof prints, pig wallows, shallow drains around village dwellings, and wheel ruts on roads.

Anopheles karwari, an Asian immigrant first discovered in the Sepik region in 1957,[99] was found in several localities in the Sepik region. It is a dangerous vector of malaria in Asia but nothing is known of its role in malaria transmission in PNG.

Anopheles longirostris and An. bancrofti were also found throughout the survey area. The former species was identified from adult mosquitoes collected mainly in the inland regions of the Sepik and Madang provinces. Larvae of the latter species were most frequently collected from swamps and lagoons of inland flood plains of the Sepik and Fly rivers. Their role in malaria transmission has not been determined.

- New information has been obtained about Anopheles vectors of malaria in four provinces of Papua New Guinea

Conclusions

The second half of the third decade (1990-1995) experienced a marked expansion of activities. Novel and well-established procedures were used to assess drug resistance of malaria parasites and to develop and evaluate antimalarial drugs in laboratory and field settings. The value of doxycycline for malaria prophylaxis was confirmed and, in addition, other antibiotics were investigated as possible alternative drugs. Despite efforts to curtail the cumbersome and lengthy primaquine eradication course, the prevention of vivax malaria remained difficult. Results of initial studies with a long-acting analogue of primaquine – tafenoquine – appeared to indicate that this new drug might be more effective than primaquine. Further studies with proguanil combinations indicated that another new drug – atovaquone – might be a suitable partner for prophylaxis of drug-resistant falciparum malaria. Investigations with PS-15 showed this experimental antifolate compound to be considerably more potent than proguanil. In the search for better tools to combat drug-resistant malaria, progress was also made during laboratory-based studies with other drugs, including artemisinin and Mannich base compounds. Personal protection measures against mosquitoes and other disease vectors were broadened to delineate the efficacy and persistence of different formulations of standard and novel repellents applied to the skin and impregnated into uniforms and bednets. Retrospective spatial analysis of potential malaria vectors, surveyed in northern Australia between 1985-1990, showed atmospheric humidity to be a key predictor of the widespread distribution of the three sibling species of *An. farauti*. After completing the survey of the Torres Strait islands, from which malaria is intermittently imported into Australia, surveys of four provinces in PNG revealed the presence of numerous species

of anopheline mosquitoes, some of which had not been identified previously in PNG. New knowledge gained about larval breeding sites and geographic distribution of these potential malaria vectors should not only be of benefit for future field deployments, but also assist local health authorities in their malaria control activities.

Acknowledgement

The opinions expressed are those of the authors and do not necessarily reflect those of the Joint Health Command or any extant Australian Defence Force Policy.

Authors' affiliation: Australian Army Malaria Institute
Corresponding author: Emeritus Professor Karl H. Rieckmann
Email: krieckmann@iprimus.com.au

Highlights

1990 Major M.D. Edstein commences 3-year exchange posting to Armed Forces Research Institute of Medical Sciences, Thailand.

Doxycycline replaces pyrimethamine/dapsone as standard malaria prophylaxis.

1991 Tafenoquine (new primaquine analogue) acts slowly against blood stages of *P. falciparum* and *P. vivax*.

Proguanil biotransformation in Australian and Thai soldiers.

Mosquito survey in Torres Strait.

1992 Major S. P. Frances commences 3- year posting to Armed Forces Research Institute of Medical Sciences, Thailand.

Lieutenant Colonel G. D. Shanks (US Army) commences 3-year posting to AMRU.

Evaluation of halofantrine at OK Tedi Mining Ltd.

Tafenoquine active against liver stages of chloroquine-resistant *P. vivax* (AMRU1) strain in *Aotus* monkeys.

In vitro assessment of non-tetracycline antibiotics, e.g. azithromycin, and artemisinin derivatives

Mannich base compounds screened by *in vitro* visual test.

Absorption of PS-15 (new antifolate prodrug) by *Saimiri* monkeys much better than its active metabolite WR 99210.

Persistence of permethrin in uniforms during field trial in Thailand.

Mosquito survey in Western Province, Papua New Guinea (PNG)

1993 Doxycycline prophylaxis proven effective and well-tolerated in 2000 Australian soldiers, with only 1-2% switching over to mefloquine prophylaxis.

Shorter courses of doxycycline plus primaquine unable to replace 14-day post-exposure course.

In vitro results suggest that 3-4 day azithromycin course probably sufficient to cure falciparum infections, but must be taken with rapidly acting drug.

Atovaquone (new drug) potentiated by proguanil, but not its metabolite cycloguanil (reverse of proguanil/dapsone synergy)

Marked degree and duration of activity of some Mannich compounds against highly resistant isolates of *P. falciparum*.

Permethrin-impregnated uniforms provide good protection after 5 cold water washes

Mosquito survey in Sepik region, PNG

1994 Tafenoquine more effective than primaquine against *P. vivax* (AMRU 1 isolate).

Proguanil/atovaquone synergistic activity not affected by patients' ability to transform proguanil to cycloguanil.

PS-15 combined with atovaquone and dapsone far more effective than proguanil triple drug combination.

Studies with Mannich bases confirm the value of using uninfected monkeys to obtain preliminary information about the degree and duration of activity of antimalarial drugs.

Evaluation of different deet formulations and new insect repellents.

Investigations with insect vectors of scrub typhus completed.

Retrospective spatial analysis of potential malaria vectors in Australia.

Mosquito survey in Gulf region, PNG.

1995 First field evaluation of non-microscopic test card for falciparum malaria.

Mannich bases more active than chemically-related drugs against drug-resistant malaria.

Pharmacokinetic information on artemisinin derivatives obtained more reliably by bioassay than chemical analysis.

Proguanil biotransformation not increased by administering high proguanil dose to poor metabolisers of the drug.

Mosquito survey in Madang Province, PNG

References

1. Rieckmann KH. The chequered history of malaria control: are new and better tools the ultimate answer? Ann Trop Med Parasitol 2006; 100(8):647-662.
2. Rieckmann KH, Sweeney AW. Army Malaria Institute: its evolution and achievements. First decade: 1965-1975. JMVH 2012; 20 (2):17-24.
3. Rieckmann KH, Edstein MD, Cooper RD, Sweeney AW. Army Malaria Institute: its evolution and achievements. Second decade: 1975-1985. JMVH 2012; 20 (3): 9-20.
4. Rieckmann KH, Davis DR, Hutton DC. *Plasmodium* vivax resistance to chloroquine. Lancet 1989; 2:1183-1184.
5. Rieckmann KH, Sweeney AW, Edstein MD, Cooper RD, Frances SP. Army Malaria Institute: its evolution and achievements. Third decade (first half): 1985-1990. JMVH 2012; 20 (4): 59-70.
6. Rieckmann KH, Yeo AET, Davis DR, Hutton DC, Wheatley PF, Simpson R. Recent military experience with malaria prophylaxis. Med J Aust 1993; 158: 446-449.
7. Chen SCA, Dwyer DE, Merrell H, Holland DJ, Robinson T, Walker J, Rieckmann KH. Multidrug-resistant *Plasmodium falciparum* malaria. Comm Dis Intell 1993: 17: 332-334.
8. Shanks GD, Roessler P, Edstein MD, Rieckmann KH. Doxycycline for malaria prophylaxis in Australian soldiers deployed to United Nations missions in Somalia and Cambodia. Milit Med 1995; 160: 443-445.
9. Rieckmann KH. Antibiotics. In: Handbook of Experimental Pharmacology: Antimalarial Drugs, 1984;Volume 68/II, Chapter 14, pp. 443-470. Springer-Verlag. Berlin, Heidelberg, New York.
10. Rieckmann KH. Falciparum malaria: The urgent need for safe and effective drugs. Ann Rev Med 1983; 34:321-335.
11. Rieckmann KH. Monitoring the response of malaria infections to treatment. Bull Wld Hlth Org 1990; 68: 759-760.
12. Hess FI, Iannuzzi A, Leafasia J, Cowdrey D, Nothdurft, HD, Von Sonnenburg F, Loscher T, Rieckmann KH. Risk factors of chloroquine resistance in *Plasmodium falciparum* malaria. Acta Tropica 1996; 61: 293-306.
13. Rieckmann KH, Sax LJ, Campbell GH, Mreme JE. Drug sensitivity of *Plasmodium falciparum*. An in-vitro microtechnique. Lancet 1978; 1:22-23
14. Reeder JC, Rieckmann KH, Genton B, Lorry K, Wines B. Cowman AF. Point mutations in the dihydrofolate reductase and dihydropteroate synthetase genes and *in vitro* susceptibility to pyrimethamine and cycloguanil of *Plasmodium falciparum* isolates from Papua New Guinea. Am J Trop Med Hyg 1996; 55: 209-213.
15. Schuurkamp GJ, Spicer PE, Kereu RK, Bulungol PK, Rieckmann KH. Chloroquine-resistant *Plasmodium* vivax in Papua New Guinea. Trans R Soc Trop Med Hyg 1992; 86: 121-122.

16. WHO: *In vitro* micro-test (Mark III) for the assessment of the response of *Plasmodium falciparum* to chloroquine, mefloquine, quinine, amodiaquine sulfadoxine/pyrimethamine and artemisinin. Instruction for use of the *in vitro* microtest kit (Mark III). Geneva: World Health Organization; 2001.
17. Van Huyssen W, Rieckmann KH. Disposable environmental chamber for assessing the drug susceptibility of malaria parasites. Trop Med Parasitol 1993; 44: 329-330.
18. Yeo AET, Rieckmann KH. A bioassay for mefloquine. J Parasitol 1992; 78: 1096-1097.
19. Xiuhong Li, Rieckmann KH. A bioassay for derivatives of qinghaosu (artemisinin). Trop Med Parasitol 1992; 43: 195-196.
20. Kotecka BM, Rieckmann KH. Chloroquine bioassay using malaria microcultures. Am J Trop Med Hyg 1993; 49: 460-464.
21. Maitland K, Williams TN, Kotecka BM, Edstein MD, Rieckmann KH. Plasma chloroquine concentrations in young and older malaria patients treated with chloroquine. Acta Tropica 1997; 66: 155-161.
22. Kotecka BM, Edstein MD, Rieckmann KH. Chloroquine bioassay of plasma specimens obtained from soldiers on chloroquine plus doxycycline for malaria prophylaxis. Int J Parasitol 1996; 26: 1325-1329.
23. Yeo AET, Rieckmann KH. Increased antimalarial activity of azithromycin during prolonged exposure of *Plasmodium falciparum in vitro*. Int J Parasitol 1995; 25: 531-532.
24. Rieckmann KH. Poor correlation between density of malaria parasitaemia and clinical symptoms in the Solomon Islands. Comment. Med J Aust 1995; 162: 389.
25. Garcia M, Kirimoama S, Marlborough D, Leafasia J, Rieckmann KH. Immunochromatographic test for malaria diagnosis. Lancet 1996; 347: 1549.
26. Yeo AET, Rieckmann KH. The *in vitro* antimalarial activity of chloramphenicol against *Plasmodium falciparum*. Acta Tropica 1994; 56: 51-54.
27. Yeo AET, Rieckmann KH. Prolonged exposure of *Plasmodium falciparum* to ciprofloxacin increases antimalarial activity. J Parasitol 1994; 80: 158-160
28. Schmidt LH. Enhancement of the curative activity of primaquine by concomitant administration of mirincamycin. Antimicrob Agents Chemother 1985; 27: 151-157.
29. Shanks GD, Barnett A, Edstein MD, Rieckmann KH. Effectiveness of doxycycline combined with primaquine for malaria prophylaxis. Med J Aust 1995; 162: 306-310.
30. Cooper RD, Milhous WK, Rieckmann KH. The efficacy of WR238605 against the blood stages of a chloroquine resistant strain of *Plasmodium* vivax. Trans. R. Soc. Trop. Med. Hyg 1994; 88: 691-692.
31. Obaldia N, Rossan RN, Cooper RD, Kyle DE, Nuzum EO, Rieckmann KH, Shanks GD. WR238605, chloroquine and their combinations as blood schizontocides against a chloroquine-resistant strain of *Plasmodium* vivax in *Aotus* monkeys. Am J Trop Med Hyg 1997; 56: 508-510.
32. Veenendaal JR, Parkinson AD, Kere N, Rieckmann KH, Edstein MD. Pharmacokinetics of halofantrine and n-desbutylhalofantrine in patients with falciparum malaria following a multiple dose regimen of halofantrine. Eur J Clin Pharmacol 1991; 41: 161-164.
33. Shanks GD, Watt G, Edstein MD, Webster HK, Suriyamongkol V, Watanasook C, Panpunnung S, Kowinwiphat W. Halofantrine for the treatment of mefloquine chemoprophylaxis failures in *Plasmodium falciparum* infections. Am J Trop Med Hyg 1991; 45:488–49.
34. Shanks GD, Watt G, Edstein MD, Webster HK, Loesuttiviboon L, Wechgritaya S. Halofantrine given with food for falciparum malaria. Trans R Soc Trop Med Hyg 1992; 86:233-234.
35. Baudon D, Bernard J, Mouliat-Pelat JP, Martet G, Sarrouy J, Touzé JE, Spiegel A, Lantrade P, Picq JJ. Efficacy of radical treatment with halofantrine on the prevention of imported *Plasmodium falciparum* malaria. Ann Soc Belg Med Trop 1992; 72:263-270.
36. Shanks GD, Edstein MD, Kereu RK, Spicer PE, Rieckmann KH. Postexposure administration of halofantrine for the prevention of malaria. Clin Infect Dis 1993; 17: 628-631.
37. Yeo AET, Edstein MD, Shanks GD, Rieckmann KH. A statistical analysis of the antimalarial activity of proguanil and cycloguanil in human volunteers. Ann Trop Med Parasitol 1994; 88: 587-594.
38. Yeo AET, Rieckmann KH. The activity *in vitro* of cycloguanil and pyrimethamine in combination against *Plasmodium falciparum*. Trans R Soc Trop Med Hyg 1992; 86: 234.
39. Edstein MD, Rieckmann KH. Lack of effect of proguanil on the pharmacokinetics of dapsone in healthy

volunteers. Chemotherapy 1993; 39: 235-241.

40. Edstein MD, Shanks GD, Teja-isavadharm P, Rieckmann KH, Webster HK. Oxidative activation of proguanil and dapsone acetylation in Thai soldiers. Br J Clin Pharmacol 1994; 37: 67-70.
41. Edstein MD, Looareesuwan S, Wilairatana P, Vanijanonta S, Kyle DE, Rieckmann KH. Disposition of proguanil in Thai patients with uncomplicated falciparum malaria. Am J Trop Med Hyg 1997; 56: 498-502.
42. Edstein MD, Yeo AET, Shanks GD, Rieckmann KH. *Ex vivo* antimalarial activity of proguanil combined with dapsone against cycloguanil-resistant *Plasmodium falciparum*. Acta Tropica 1997; 66: 127-135.
43. Looareesuwan S, Viravan C, Webster HK, Kyle DE, Hutchinson DB, Canfield CJ. Clinical studies of atovaquone alone or in combination with other antimalarial drugs, for treatment of acute uncomplicated malaria in Thailand. Am J Trop Med Hyg 1996; 54: 62-66.
44. Canfield CJ, Pudney M, Gutteridge WE. Interactions of atovaquone with other antimalarial drugs against *Plasmodium falciparum in vitro*. Exp Parasitol 1995; 80: 373-381.
45. Yeo AET, Seymour KK, Rieckmann KH, Christopherson RI. Effects of dual combinations of antifolates with atovaquone or dapsone on nucleotide levels in *Plasmodium falciparum*. Biochem Pharmacol 1997; 53: 943-950.
46. Edstein MD, Yeo AET, Kyle DE, Looareesuwan S, Wilairatana P, Rieckmann KH. Proguanil polymorphism does not affect the antimalarial activity of proguanil combined with atovaquone *in vitro*. Trans R Soc Trop Med Hyg 1996; 90: 418-421.
47. Seymour K, Lyons SD, P*hilli*ps L, Rieckmann KH, Christopherson RI. Cytotoxic effects of inhibitors of *de novo* pyrimidine biosynthesis upon *Plasmodium falciparum*. Biochemistry 1994; 33: 5268-5274.
48. Seymour KK, Yeo AET, Rieckmann KH, Christopherson RI. dCTP levels are maintained in *Plasmodium falciparum* subjected to pyrimidine deficiency or excess. Ann Trop Med Parasitol 1997; 91: 603-609.
49. Yeo AET, Edstein MD, Rieckmann KH. Antimalarial activity of the triple combination of proguanil, atovaquone and dapsone. Acta Tropica 1997; 67: 207-214.
50. Yeo AET, Seymour KK, Rieckmann KH, Christopherson RI. Effects of folic and folinic acids on the activities of cycloguanil and WR99210 against *Plasmodium falciparum* in erythrocytic culture. Ann Trop Med Parasitol 1997a; 91: 17-23.
51. Yeo AET, Rieckmann KH. The activity of triple combinations of antifolate biguanides, with and without folinic acid, against *Plasmodium falciparum in vitro*. Ann Trop Med Parasitol 1997b; 91: 247-251.
52. Yeo AET, Edstein MD, Shanks GD, Rieckmann KH. Potentiation of the antimalarial activity of atovaquone by doxycycline against *Plasmodium falciparum in vitro*. Parasitol Res 1997; 83: 489-491.
53. Yeo AET, Rieckmann KH, Christopherson RI. Indirect inhibition by antibiotics of nucleotide and deoxynucleotide biosynthesis in *Plasmodium falciparum*. Southeast Asian J Trop Med Public Health 1998; 29: 24-26.
54. Rieckmann KH. The *in vitro* activity of experimental antimalarial compounds against strains of *Plasmodium falciparum* with varying degrees of sensitivity to pyrimethamine and chloroquine. In: Chemotherapy of Malaria and Resistance to Antimalarials. Geneva: World Health Organization. Techn Rep Ser 1973; No. 529, p. 58.
55. Canfield CJ, Milhous WK, Ager AL, Rossan RN, Sweeney TR, Lewis NJ, Jacobus DP. PS-15: a potent, orally active antimalarial from a new class of folic acid antagonists. Am J Trop Med Hyg 1993; 49: 121-126.
56. Edstein MD, Corcoran KD, Shanks GD, Ngampo chjana, M, Hansukjariya P, Sattabongkot, Webster HK, Rieckmann KH. Evaluation of WR250417 (a proguanil analog) for causal prophylactic activity in the *Plasmodium* cynomolgi - macaca mulatta model. Am J Trop Med Hyg 1994; 50: 181-186.
57. Kotecka BM, Rieckmann KH. *In vivo-in vitro* model for assessing chloroquine activity in monkeys. Chemotherapy 1995; 41: 134-140.
58. Rieckmann KH, Yeo AET, Edstein MD. Activity of PS-15 and its metabolite, WR99210, against *Plasmodium falciparum* in an *in vivo-in vitro* model. Trans R Soc Trop Med Hyg 1996; 90: 568-571.
59. Edstein MD, Bahr S, Kotecka B, Shanks GD, Rieckmann KH. *In vitro* activities of the biguanide PS-15 and its metabolite, WR99210, against cycloguanil-resistant *Plasmodium falciparum* isolates from Thailand. Antimicrob Agents Chemother 1997; 41: 2300-2301.
60. Yeo AET, Rieckmann KH. The activity of PS-15 in combination with sulfamethoxazole. Trop Med Parasitol

1994; 45: 136-137.

61. Rieckmann KH. Determination of the drug sensitivity of *Plasmodium falciparum*. J Am Med Assoc 1971; 217: 573-578.
62. Fu S, Xiao SH. Pyronaridine: a new antimalarial drug. Parasitology Today 1991; 7: 310-313.
63. Barlin GB, Jiravinyu C, Butcher GA, Kotecka B, Rieckmann KH. The *in vitro* and *in vivo* antimalarial activity of some Mannich bases derived from 4-(7'-trifluoromethyl-1', 5'-naphthyridin-4'-ylamino) phenol, 2-(7'trifluoromethyl-quinolin-4'-ylamino) phenol, and 4'-chloro-5-(7"-trifluoromethylquinolin-4"-ylamino) biphenyl-2-ols. Ann Trop Med Parasitol 1992a; 86: 323-331.
64. Barlin GB, Nguyen TMT, Kotecka B, Rieckmann KH. Potential Antimalarials. XV. Di-Mannich bases of 2-(7'Chloroquinolin-4'-yl-amino) phenol and 2-(7'-bromo (and trifluoromethyl)- 1', 5'-naphthyridin-4'-ylamino]phenol. Aust J Chem 1992b; 45: 1651-1662.
65. Barlin GB, Tian FL, Kotecka B, Rieckmann KH. Potential Antimalarials. XVI. 4'-Chloro-3'[7"-chloro (and trifluoromethyl) quinolin-4"-yl]amino-5-(substituted amino) methylbiphenyl-4-ols and 4'-bromo (and 3'-trifluoromethyl)-3-(subsituted amino) methyl-5-7"-trifluoromethylquinolin-4"-yl) aminobiphenyl-2-ols. Aust J Chem 1992c; 45: 1845-1955.
66. Barlin GB, Nguyen TMT, Kotecka B. Rieckmann KH (1993). Potential Antimalarials. XVII. Di-and mono-Mannich bases of 2(and 4)-[2(and 8)-trifluoromethylquinolin-4-ylamino]phenol. Aust J Chem 1993a; 46: 21-29.
67. Barlin GB, Ireland SJ, Nguyen TMT, Kotecka B, Rieckmann KH. Potential Antimalarials. XVIII. Some mono- and di Mannich bases of 3-[7-chloro(and trifluoromethyl) quinolin-4-ylamino]phenol. Aust J Chem 1993b; 46: 1685-1693.
68. Barlin GB, Ireland SJ, Jiravinuy C, Nguyen TMT, Kotecka B, Rieckmann KH. Potential Antimalarials. XIX. Syntheses and testing of a-(piperidin-2-yl)-a-(7'-trifluoromethylquinolin-4-yl) methanol and a -(7-bromo-1,5-naphthyridin-4-yl)-a-(piperidin-2'-yl) methanol. Aust J Chem 1993c; 46: 1695-1703.
69. Barlin GB, Ireland SJ, Trang MTN, Kotecka B, Rieckmann KH. Potential antimalarials. XX. Mannich base derivatives of 2-[7-chloroquine-4-ylamino and 7-bromo (and 7-trifluoromethyl)-1,4-naphthyridin-4-ylamino]-4-chloro-(or 4-or 6-5-butyl or 4-or-5-floro)phenols and 4 (or 6)-t-butyl-2-(7 trifluoromethylquinolin-4-ylamino) phenol. Aust J Chem 1994a; 47: 1143-1154.
70. Barlin GB, Ireland SJ, Trang MTN, Kotecka B, Rieckmann KH. Potential antimalarials. XXI. Mannich base derivatives of 4-[7-chloro(and 7-trifluromethyl)quinolin-4-ylamino]phenols. Aust J Chem 1994b; 47: 1553-1560.
71. Kotecka B, Rieckmann KH. An inexpensive and simple method for screening potential antimalarial drugs. Trop Med Parasitol 1992; 43: 9-12.
72. Kotecka BM, Barlin GB, Edstein MD, Rieckmann KH. New quinoline di-Mannich bases with greater antimalarial activity than chloroquine, amodiaquine or pyronaridine. Antimicrob Agents Chemother 1997; 41: 1369-1374.
73. Frances SP, Eikarat N, Sripongsai B, Eamsila C. Response of *Anopheles dirus* and *Aedes albopictus* to repellents in the laboratory. J Am Mosq Control Assoc 1993; 9:474-476.
74. Frances SP, Klein TA, Hildebrandt DW, Burge R, Noigamol C, Eikarat N, Sripongsai B, Wirtz RA. Laboratory and field evaluation of the repellents, deet, CIC-4 and AI3-37220, against *Anopheles dirus* (Diptera: Culicidae) in Thailand. J Med Entomol 1996a; 33:511-515.
75. Frances SP, Eamsila C, Pilakasiri C, Linthicum KJ. Effectiveness of repellent formulations containing deet against mosquitoes in northeastern Thailand. J Am Mosq Control Assoc 1996b 12: 331-333.
76. Frances SP, Klein TA, Wirtz RA, Eamsila C, Pilakasiri C, Linthicum KJ. *Plasmodium falciparum* and *Plasmodium* vivax circumsoprozoite proteins in anophelines collected in eastern Thailand. J Med Entomol 1996c; 33:990-991.
77. Frances SP, Sweeney AW. Response of *Anopheles farauti* to permethrin-impregnated net and cloth fabrics in the laboratory. J Am Mosq Control Assoc 1996; 12:321-324.
78. Health Manual 21, Australian Defence Force.
79. Eamsila C, Frances SP, Strickman D. Evaluation of permethrin-treated military uniforms for personal protection against malaria in northeastern Thailand. J. Am. Mosq. Control Assoc 1994; 10:515-521.

80. Frances SP, Eamsila C, Strickman D. Antibodies to *Orientia tsutsugamush*i in soldiers in northeastern Thailand. Southeast Asian J Trop Med Publ Hlth 1997; 28:666-668.

81. Frances SP, Watcharapichat P, Phulsuksombati D, Tanskul P. Occurrence of *Orientia tsutsugamush*i in rodents and chiggers (Acari: Trombiculidae) in an orchard near Bangkok, Thailand. J Med Entomol 1999a; 36:449-453.

82. Frances SP, Watcharapichat P, Phulsuksombati D. Development and persistence of antibodies to *Orientia tsutsugamush*i in the roof rat, Rattus rattus and laboratory mice following attachment of naturally infected *Leptotrombidium deliense*. Acta Tropica 2000b; 77: 279-285.

83. Frances SP, Watcharapichat P, Phulsuksombati D. Vertical transmission of *Orientia tsutsugamush*i in two lines of naturally infected *Leptotrombidium deliense* (Acari: Trombiculidae). J Med Entomol 2001a; 38:17-21.

84. Frances SP. Potential for horizontal transmission of *Orientia tsutsugamush*i by chigger mites (Acari: Trombiculidae). Int J Acarol 2005; 31: 75-82.

85. Frances SP, Watcharapichat P, Phulsuksombati D, Tanskul P. Transmission of *Orientia tsutsugamush*i, the aetiologic agent for scrub typhus, to co-feeding mites. Parasitol 2000a; 120:601-607.

86. Frances SP, Watcharapichat P, Phulsuksombati D, Tanskul P, Linthicum KJ. Seasonal occurrence of *Leptotrombidium deliense* (Acari: Trombiculidae) attached to sentinel rodents in an orchard near Bangkok, Thailand. J Med Entomol 1999b; 36:869-874.

87. Frances SP, Watcharapichat P, Phulsuksombati D, Tanskul P. Investigation of the role of *Blankaartia acuscutellaris* (Acari: Trombiculidae) as a vector of scrub typhus in central Thailand. Southeast Asian J Trop Med Publ Hlth 2001b; 32:863-866.

88. Frances SP, Khlaimanee N. Laboratory tests of arthropod repellents against *Leptotrombidium deliense*-noninfected and infected with Rickettia tsutsugamushi- and noninfected *L. fletcheri*. J Med Entomol 1996; 33:232-235.

89. Sweeney AW, Cooper RD, Frances SP. Distribution of the sibling species of *Anopheles farauti* in the Cape York peninsula, northern Queensland, Australia. J Am Mosq Control Assoc 1990; 6: 425-429.

90. Cooper RD, Frances SP, Sweeney AW. Distribution of members of the *Anopheles farauti* complex in the Northern Territory of Australia. J Am Mosq Control Assoc 1995; 11:66-71.

91. Cooper RD, Frances SP, Waterson DGE, Piper RG, Sweeney AW. The distribution of anopheline mosquitoes in northern Australia. J Am Mosq Control Assoc 1996; 12: 656-663.

92. Sweeney AW, Beebe NW, Cooper RD. Analysis of environmental factors influencing the range of anopheline mosquitoes in northern Australia using a genetic algorithm and data mining methods. Ecological Modelling 2007; 203: 375-386.

93. Sweeney AW. Larval salinity tolerances of the sibling species of *Anopheles farauti*. J Am Mosq Control Assoc 1987; 3: 589-592.

94. Sweeney AW, Beebe NW, Cooper RD, Bauer J T, Peterson AT. Environmental Factors Associated with Distribution and Range Limits of Malaria Vector *Anopheles farauti* in Australia. J Med Entomol 2006; 43:1068-1075.

95. Cooper RD, Frances SP, Popat S, Waterson DGE. The effectiveness of light, 1-octen-3-ol, and carbon dioxide as attractants for anopheline mosquitoes in Madang Province, Papua New Guinea. J Am Mosq Control Assoc 2004; 20: 239-242.

96. Cooper L, Cooper RD, Burkot TR. The *Anopheles* punctulatus complex: DNA probes for identifying the Australian species using isotopic, chromogenic, and chemiluminescence detection systems. Exp Parasitol 1991; 73: 27-35.

97. Beebe NW, Foley DH, Saul A, Cooper L, Bryan JH, Burkot TR. DNA probes for identifying members of the *Anopheles* punctulatus complex in Papua New Guinea. Am J Trop Med Hyg 1994; 50: 229-234.

98. Cooper RD, Waterson DGE, Kupo M, Foley DH, Beebe NW, Sweeney AW. Anopheline mosquitoes of the Western Province of Papua New Guinea. J Am Mosq Control Assoc 1997; 13:5-12.

99. Peters W, Standfast HA. Report on a malaria survey in the Sepik District. Med J Aust 1957; 1: 861-868.

JOURNAL OF MILITARY AND VETERANS' HEALTH

CALL TO AUTHORS

The Journal of Military and Veterans' Health is a peer reviewed quarterly publication published by the Australasian Military Medicine Association. Its Editorial Board has identified the following themes for future editions:

Edition	Theme	Publication Date	Closure of article submission date
October 2013	Initial Trauma Management	21 October 2013	30 August 2013
January 2014	General Papers	20 January 2014	29 November 2013
April 2014	Medical Aspects of World War I	21 April 2014	28 February 2014

Categories for the above include:

Original Research/Original Articles, Short Communication, Review Articles, Reprinted Articles, Case Studies, Abstracts from the Literature, Biographies, History, Boiok Reviews, Commentary and View from the Front.

The Editor would be delighted to receive articles for consideration on these themes. However, please note that although these are identified themes for 2013 and 2014, we encourage authors to continue to submit articles on a range of topics on military medicine and veterans' health.

Submission of articles should be done online via JMVH website www.jmvh.org where the Instructions to Authors can also be found.

Should you have any queries, in relation to JMVH, please do not hesitate to contact the Editor via the AMMA Secretariat on 61 3 6234 7844 or editorial@jmvh.org or secretariat@amma.asn.au.

Do you have an article of interest to our readers?

Authors are invited to submit articles of relevance to the Editor for consideration for publication in the Journal of Military and Veterans' Health (JMVH)

Categories include:

Informative and commentary
Original Research/Original Articles
Short Communication
Review Articles
Reprinted Articles
Case Studies
Abstracts from the Literature
Letters to the editor
Biographies
History
Obituaries
Book Reviews
Commentary
View from the Front

See the JMVH website for authors' instructions and
Submit your article online at www.jmvh.org

JMVH

www.jmvh.org