

AD \_\_\_\_\_

Award Number: W81XWH-05-2-0081

TITLE: Military Nutrition Research: Four Tasks to Address Personnel Readiness and Warfighter Performance

PRINCIPAL INVESTIGATOR: Donna Ryan, M.D.

CONTRACTING ORGANIZATION: Pennington Biomedical Research Center  
Baton Rouge, LA 70808

REPORT DATE: March 2007

TYPE OF REPORT: Final Addendum

PREPARED FOR: U.S. Army Medical Research and Materiel Command  
Fort Detrick, Maryland 21702-5012

DISTRIBUTION STATEMENT: Approved for Public Release;  
Distribution Unlimited

The views, opinions and/or findings contained in this report are those of the author(s) and should not be construed as an official Department of the Army position, policy or decision unless so designated by other documentation.

| REPORT DOCUMENTATION PAGE                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                  |             |                                  |                            | Form Approved<br>OMB No. 0704-0188           |                                           |
|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-------------|----------------------------------|----------------------------|----------------------------------------------|-------------------------------------------|
| Public reporting burden for this collection of information is estimated to average 1 hour per response, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing and reviewing this collection of information. Send comments regarding this burden estimate or any other aspect of this collection of information, including suggestions for reducing this burden to Department of Defense, Washington Headquarters Services, Directorate for Information Operations and Reports (0704-0188), 1215 Jefferson Davis Highway, Suite 1204, Arlington, VA 22202-4302. Respondents should be aware that notwithstanding any other provision of law, no person shall be subject to any penalty for failing to comply with a collection of information if it does not display a currently valid OMB control number. PLEASE DO NOT RETURN YOUR FORM TO THE ABOVE ADDRESS.                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                         |             |                                  |                            |                                              |                                           |
| 1. REPORT DATE<br>01-03-2007                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                               |             | 2. REPORT TYPE<br>Final Addendum |                            | 3. DATES COVERED<br>1 Mar 2006 – 28 Feb 2007 |                                           |
| 4. TITLE AND SUBTITLE<br><br>Military Nutrition Research: Four Tasks to Address Personnel Readiness and Warfighter Performance                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                             |             |                                  |                            | 5a. CONTRACT NUMBER                          |                                           |
|                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                            |             |                                  |                            | 5b. GRANT NUMBER<br>W81XWH-05-2-0081         |                                           |
|                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                            |             |                                  |                            | 5c. PROGRAM ELEMENT NUMBER                   |                                           |
| 6. AUTHOR(S)<br><br>Donna Ryan, M.D.<br><br>Email: <a href="mailto:ryandh@pbrc.edu">ryandh@pbrc.edu</a>                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                    |             |                                  |                            | 5d. PROJECT NUMBER                           |                                           |
|                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                            |             |                                  |                            | 5e. TASK NUMBER                              |                                           |
|                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                            |             |                                  |                            | 5f. WORK UNIT NUMBER                         |                                           |
| 7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES)<br><br>Pennington Biomedical Research Center<br>Baton Rouge, LA 70808                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                   |             |                                  |                            | 8. PERFORMING ORGANIZATION REPORT NUMBER     |                                           |
| 9. SPONSORING / MONITORING AGENCY NAME(S) AND ADDRESS(ES)<br>U.S. Army Medical Research and Materiel Command<br>Fort Detrick, Maryland 21702-5012                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                          |             |                                  |                            | 10. SPONSOR/MONITOR'S ACRONYM(S)             |                                           |
|                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                            |             |                                  |                            | 11. SPONSOR/MONITOR'S REPORT NUMBER(S)       |                                           |
| 12. DISTRIBUTION / AVAILABILITY STATEMENT<br>Approved for Public Release; Distribution Unlimited                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                           |             |                                  |                            |                                              |                                           |
| 13. SUPPLEMENTARY NOTES                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                    |             |                                  |                            |                                              |                                           |
| 14. ABSTRACT<br>Background: Pennington Biomedical Research Center (PBRC) continues a 19 year collaborative effort with the Department of Defense (DoD) in this research effort. Objectives: To assess and evaluate novel ways to sustain warfighter performance during high intensity missions at home and abroad, under specially funded cooperative agreements between the US Army Medical Research and Materiel Command (USAMRMC) and PBRC, PBRC provides high quality analytical laboratory, nutrition database and metabolic unit support for military nutrition clinical research protocols. Specific Aims: PBRC performed four research tasks as follows: Task 1: Clinical Laboratory for Human Samples; Task 2: Stable Isotope Laboratory; Task 3: Nutrient Database Laboratory; Task 4: Metabolic Unit Project. During the year of this report, the four tasks supported eight projects directed by USARIEM investigators. Study Design: In consultation with the Project Officer at US Army Research Institute of Environmental Medicine (USARIEM), PBRC Task Leaders for Tasks 1 and 2 determine the number, timing, type of sample and type of analysis. Analysis of relevant endpoints in the PBRC Clinical Laboratory and Stable Isotope Laboratory provides information useful for determination of energy expenditure, water turnover, body composition, clinical biochemistry and metabolism. In consultation with the Project Officer, the Task Leader for Task 3 provides assistance with food intake analysis and nutrient composition. In consultation with the Project Officer, the Task Leader for Task 4 provides access to the PBRC Metabolic Unit, where capabilities exist for clinical studies of relevance to energy metabolism using state of the art facilities for metabolic chambers and magnetic resonance spectroscopy. |             |                                  |                            |                                              |                                           |
| 15. SUBJECT TERMS<br>metabolism, nutrition, energy expenditure, readiness, performance                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                     |             |                                  |                            |                                              |                                           |
| 16. SECURITY CLASSIFICATION OF:                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                            |             |                                  | 17. LIMITATION OF ABSTRACT | 18. NUMBER OF PAGES                          | 19a. NAME OF RESPONSIBLE PERSON           |
| a. REPORT                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                  | b. ABSTRACT | c. THIS PAGE                     |                            |                                              | USAMRMC                                   |
| U                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                          | U           | U                                | UU                         | 53                                           | 19b. TELEPHONE NUMBER (include area code) |

## Table of Contents

|                                        |    |
|----------------------------------------|----|
| Cover . . . . .                        | 1  |
| SF 298. . . . .                        | 2  |
| Table of Contents . . . . .            | 4  |
| Introduction . . . . .                 | 5  |
| Body . . . . .                         | 6  |
| Key Research Accomplishments . . . . . | 26 |
| Reportable Outcomes . . . . .          | 28 |
| Conclusion . . . . .                   | 29 |
| References . . . . .                   | 30 |
| Appendices . . . . .                   | 39 |

## INTRODUCTION

The Pennington Biomedical Research Center (PBRC) has a 19-year history of collaborative research with the Department of Defense (DOD). A series of specially funded cooperative agreements between the PBRC and the U.S. Army Medical Research and Materiel Command (USAMRMC) has provided high quality analytical laboratory, nutrition database, and metabolic unit support for DOD nutrition related research programs. The program currently supports the RDT&E funded Military Nutrition Research Programs at the U.S. Army Soldier Systems Center (Natick, Massachusetts) and the U.S. Army Research Institute of Environmental Medicine (USARIEM) laboratories, as well as the Ration Sustainment Testing program. PBRC personnel have traveled to DOD field studies to collect samples, which are returned to the PBRC for laboratory analyses. Additionally, the PBRC has conducted research that complements and extends USARIEM's intramural program in areas of nutritional neuroscience, stress, physical, and mental performance, and garrison feeding. Though funded through earmarks, the PBRC program has been periodically successfully peer reviewed by an external panel from the Committee on Military Nutrition Research (CMNR), Institute of Medicine (1988, 1990, 1996, and 2002). This joint effort of PBRC and military researchers has led to significant improvements of operational rations, better understanding of warfighter energy and nutritional requirements, and modifications in garrison feeding.

This project continues a research program that has been in place since 1988. The project is for the fourth specially funded cooperative agreement series that began in 1988. The previous cooperative agreements are listed below.

| <b>Dates of Award</b> | <b>Title</b>                                                                                                                          | <b>Funding</b> |
|-----------------------|---------------------------------------------------------------------------------------------------------------------------------------|----------------|
| 7/1/97-12/31/04       | Military Nutrition Research: Eight Tasks to Address Medical Factors Limiting Soldier Effectiveness                                    | \$16,710,748   |
| 4/1/92-3/30/98        | Military Nutrition Research: Six Tasks to Address Medical Factors Limiting Soldier Effectiveness                                      | \$11,340,567   |
| 8/1/88-7/31/92        | Effect of Food, Diet and Nutrition on Military Readiness and Preparedness of Army Personnel and Dependents in a Peacetime Environment | \$3,845,000    |

In each of the three previous cooperative agreements, the CMNR of the National Academy of Sciences provided peer review prior to and during program implementation. Additionally, USARIEM approved all projects and provided consultation on research design for all projects that emanated from PBRC. Any modifications to the original research plan were approved by USARIEM prior to implementation.

In November 2006, a review was conducted of the research at USARIEM by the American Institute of Biological Sciences and the projects and tasks outlined in this document were approved.

## BODY

PBRC provides high quality support of military nutrition clinical research protocols through the execution of the four tasks outlined below. PBRC and the US Army Medical Research and Materiel Command (USAMRMC) cooperate in this specially funded agreement to assess and evaluate novel ways to sustain warfighter performance during high intensity missions at home and abroad. In November 2006 the Military Nutrition Division's research program was reviewed by the American Institute of Biological Sciences. Dr. Jennifer Rood presented a description of the tasks outlined below as part of the review. Members of the review panel were Drs. Mark Davis (chair), Susan Racette, Kathryn Schmidt, and Robert Hickner. In all cases, both the Military Nutrition Division and the corresponding PBRC tasks received a positive review and the continuing cooperative relationship was fully supported.

### Task 1: Clinical Laboratory for Human Samples (Jennifer Rood, Ph.D., Task Leader)

- After consultation with U. S. Army Research Institute of Environmental Medicine (USARIEM), samples from USARIEM research projects were received, processed, and analyzed by the PBRC laboratory.
- The number, timing, type of sample and analyses performed were determined in consultation with USARIEM investigators.
- Results were forwarded to USARIEM and publications generated.

### Task 2: Stable Isotope Laboratory (Jennifer Rood, Ph.D., Task Leader)

- After consultation with USARIEM, samples for analysis of energy expenditure, body composition and protein/metabolite turnover studies were analyzed by the PBRC laboratory.
- The number, timing, type of sample and analyses performed were determined in consultation with USARIEM investigators.
- Results were forwarded to USARIEM and scientific publications will be generated.

### Task 3: Nutrient Database Laboratory (Catherine Champagne, Ph.D., Task Leader)

- After consultation with USARIEM, food intake analysis services can be performed and nutrient database services provided.
- The number and timing of field data collection services provided by PBRC personnel can be determined in collaboration with USARIEM.
- Results can be forwarded to USARIEM and scientific publications will be generated.

### Task 4: Metabolic Unit Services (Steven Smith, MD, Task Leader)

- Protocols can be developed in consultation with USARIEM and implemented using the PBRC metabolic unit clinical facility.
- PBRC can provide for metabolic kitchen services, inpatient unit nursing, clinical laboratory assessment, energy expenditure assessment, body composition assessment, food intake assessment and in-vivo bio imaging using multinuclear magnetic resonance spectroscopy.
- In consultation with USARIEM, scientific publications can be generated.

During the year of this report, the four tasks were used to support the following eight projects directed by USARIEM investigators:

1. Project 1 - The effect of fitness level, caloric intake, and protein intake on short-term nitrogen balance during a 1000-calorie increase in daily energy expenditure (PI: CPT Matthew Pikosky, Ph.D.) - supported by Tasks 1 and 2.
2. Project 2 - Comparison of a PDA-based Method vs. Written Records for Assessing Energy Intake and Expenditure (PI: Gaston Bathalon, Ph.D.) - supported by Task 2.
3. Project 3 - Response of Biomarkers of Bone Remodeling to Military Recruit Training (PI: Rachel Evans, Ph.D.) - supported by Task 1.
4. Blood biochemistries following repeated days of First Strike Ration or Meal, Ready to Eat consumption (PI: Scott Montain, Ph.D.) – supported by Task 1 and 2.
5. Impact of Basic Combat Training on iron Status and Cognitive Function Among Female Military Personnel (PI: Harris Lieberman, Ph.D.) – supported by Task 1.
6. The Effect of a Prolonged Period Aboard a Submarine on Bone Strength and Metabolism. (PI: Rachel Evans, Ph.D.) – supported by Task 1.
7. Effects of Physical Training and Iron Status on Body Composition, Muscle Function, and Bone Health in Rats (PI: James McClung, Ph.D.) – supported by Task 1.
8. Efficiency of casein-whey drink for optimizing plasma amino acid availability (PI: Scott Montain, Ph.D.) – supported by Task 1.

Detailed descriptions of each of the eight projects are described below:

**Project 1** - The effect of fitness level, caloric intake, and protein intake on short-term nitrogen balance during a 1000-calorie increase in daily energy expenditure .

## **Background**

During heavy sustained military operations, nutrition is a major factor that impacts the health and performance of warfighters. However, for this population, the importance of dietary protein for “optimal” nutrition is not resolved. The extent to which total daily energy expenditure, caloric intake, and fitness level affect the balance between whole-body protein synthesis and whole-body protein breakdown and, thus, dietary protein requirements, remain debated. Sedentary individuals who increase activity exhibit a short-term increase in nitrogen loss from the body, assumed to reflect increased dietary protein requirement, but whether physically fit individuals, such as warfighters, respond similarly is unknown. Additionally, while whole-body protein catabolism increases when energy intake does not meet energy expenditure, the degree to which an increase in dietary protein may preserve whole-body protein synthesis and reduce catabolism, during inadequate intake and increased energy expenditure is unknown.

The purpose of this study is two fold: 1) to determine if, during increased energy expenditure, physically fit individuals exhibit a smaller, larger, or equivalent increase in protein requirement as sedentary individuals and 2) to determine if, during periods of heavy physical activity and inadequate intake, an increase in protein intake will enhance conservation of whole-body protein content.

## Hypotheses

1. In fit and sedentary individuals, daily nitrogen balance becomes negative when TDEE increases significantly, even when energy intake matches expenditure.
2. Due to metabolic adaptations enhancing whole body protein synthetic capacity and/or blunting proteolysis, fit individuals will preserve nitrogen balance better than sedentary individuals when TDEE increases significantly.
3. For fit individuals who significantly increase TDEE, but not energy intake, consuming a greater fraction of energy as dietary protein will improve nitrogen balance by enhancing whole-body protein synthetic rate, blunting proteolysis, or both.
4. For fit individuals who significantly increase TDEE, but not energy intake, consuming a greater fraction of energy as dietary protein will sustain or enhance gluconeogenesis while blunting proteolysis, and this effect will diminish with time as negative energy balance continues.

## Technical Objectives

1. Over 7 days, determine nitrogen balance and measure changes in rates of whole body protein turnover and gluconeogenesis, in sedentary and fit individuals who increase TDEE by 1000 calories while consuming an energy sufficient diet that provides 55% of total calories from carbohydrate and 0.9 g protein/kg body weight.
2. Over 7 days, determine nitrogen balance and measure changes in rates of whole body protein turnover and gluconeogenesis, in fit individuals expending an additional 1000 calories while consuming a hypocaloric diet (deficit of 1000 calories) that provides 55% of total calories from carbohydrate and 0.9 g protein/kg body weight.
3. Over 7 days, determine nitrogen balance and measure changes in rates of whole body protein turnover and gluconeogenesis, in fit individuals expending an additional 1000 calories while consuming a hypocaloric diet (deficit of 1000 calories) that provides 55% of total calories from carbohydrate and 1.8 g protein/kg body weight.
4. Monitor plasma concentrations of the following:
  - a. Hormones related to energy metabolism (i.e. insulin, glucagon, catecholamines).
  - b. Metabolically active substances that affect protein metabolism (i.e. growth hormone, cortisol, cytokines).
  - c. Products of energy metabolism (i.e. urea nitrogen, free fatty acids, glycerol).
5. Use the respirometry chamber to confirm that TDEE is both predictable and reproducible when a detailed activity schedule is followed.

## Military Relevance

This research addresses USARIEM STP III.B. Task 1 (Fueling optimal performance) and Task 2 (Enhancing/defending the body's biological matrix). Protein synthesis maintains physiological function, including immune response, neurotransmission, muscle mass and, thereby, performance.

Controlled studies investigating whole-body protein turnover and dietary protein requirement of physically fit individual's abruptly increasing total daily energy expenditure (TDEE) have not been conducted. At high levels of energy expenditure it is unclear how much dietary protein is required to meet the body's demand for energy and protein synthesis, or how a caloric deficit may impact this requirement. In fact, the 1999 Institute of Medicine report recommends research to quantitate the effect of energy deficit on protein requirements (1). The results of this experiment will be used to evaluate the adequacy of current Military Dietary

Reference Intake (MDRI) and Nutritional Standards for Operational Rations (NSOR) for protein and to make recommendations regarding the optimal protein content of operational rations and/or nutritional supplements. In addition, data from this study will be used to further develop, refine, and validate the Dynamic Nutrition Model being developed by The U.S. Army Soldier and Biological Chemical Command.

## **Project 2 – Comparison of a PDA-based Method vs. Written Records for Assessing Energy Intake and Expenditure**

### **Background**

Dietary intake studies in military dining facilities have generally relied upon a visual estimation method to collect dietary data to assess energy intake. However, this method is only suitable for group feeding and, thus, is not feasible for studies involving free-living individuals (1). Reports show that energy intakes calculated from self-recorded food records are, on average, underestimated 10 – 30% when compared with energy expenditure determined by DLW or indirect calorimetry (2-4). The discrepancies are due mostly to underreporting of food intake which can be the failure to record all foods consumed or the underestimation of portion sizes. Likewise, research comparing written, self-reported physical activity against DLW demonstrates that self reports overestimate energy expenditure by approximately 10% (5-7). Clearly, there is a need to improve the self-reporting of food intake and physical activity in order to determine the energy balance of individuals with greater accuracy and precision (8).

Successful weight management requires a careful balance between energy intake (food and beverage consumed) and energy expenditure (activities of daily living). Self-monitoring of food intake and physical activity has been identified as one of the most effective strategies for weight loss and maintenance (9-10). Self-monitoring at the beginning of a weight loss program assesses eating and exercise behavior and continues throughout the program to evaluate behavioral changes and compliance with program goals (11). Evidence indicates that individuals who self-monitor their food intake and physical activities are more successful at weight loss than those inconsistent with self-monitoring (12). Current findings support the IOM recommendations of keeping a 3-day food intake record and a physical activity diary (or using an activity monitor such as a pedometer) every 3 months to help maintain weight loss (13). Therefore, valid and accurate self-monitoring techniques are necessary to assess deviations in energy balance to assist overweight and overfat Soldiers in successful weight loss and weight management (14-15).

Objective assessment of energy expenditure can be measured by a variety of methods, with varying degrees of accuracy. Components of TEE include resting energy expenditure (REE), which typically encompasses 50% - 70% of TEE; the thermic effect of food (TEF), which accounts for another 10%; and the remainder is physical activity. Physical activity is the most variable component (16) and is comprised of activities of daily living (bathing, feeding, and grooming, for example), sports and leisure, and occupational activities. The amount of TEE accounted for by physical activity and thus, the error of misreporting energy expenditure is greater for active individuals than sedentary.

Doubly labeled water, a reference method for validation of energy intake and expenditure, is a widely used criterion for estimating TEE in a free-living population (17). In DLW, TEE is calculated from carbon dioxide production and an estimate of the respiratory ratio and has been extensively validated against gas exchange in small animals and humans (18-19). When free living individuals are weight-stable, energy intake will be equal to TEE. Therefore, DLW allows investigators to compare the accuracy of self-reported energy intake.



Objective assessment of energy intake can also be measured by a variety of methods with varying degrees of accuracy. Seven-day weighted food intake records are often used when evaluating other dietary assessment methods such as the 24-hour recall, food frequency record, or other novel methods (20). Written self-reported food intake records are a common method for collecting energy intake. The volunteer writes descriptive information of all foods and beverages consumed over a period of time, typically 3 – 7 consecutive days (recording food intake for more than 7 days has shown greater error in reporting bias) (21). While keeping a food record, individuals are instructed to accurately describe all foods and beverages consumed including the name of the food/beverage (brand name, if applicable), preparation method(s), recipes for mixed foods and beverages, and accurate portion sizes. Portion sizes are measured (with a scale or other household measures such as cups or tablespoons), estimated against food models or pictures, or estimated without the use of an aid. Ideally, recording is done at the time the food(s) and/or beverage(s) is/are consumed, thereby, increasing recording accuracy of the foods and beverages consumed. Individuals often view the written food record as time-consuming. To reduce the omission of forgotten foods and clarify entries, the food intake records are reviewed with respondents by trained study personnel.

Studies indicate that underreporters of food intake are likely to be female (22), older (23), or overweight (24-25). Additionally, income, education (26), social desirability (27), body image, and history of dieting or restrained eating (28) are likely to influence accuracy of reporting. The Three-Factor Eating Questionnaire (TFEQ) is internally consistent and valid and is suggested as the best tool for psychometrically measuring the degree of dietary restraint (29). Therefore, we will control for dietary restraint by using results from the TFEQ such as disinhibition, hunger, and dietary restraint. Additionally, we will match groups for gender, age, and body mass index.

Because of the quantity and diversity of information collected in food intake records, coding (data entry) and nutrient analysis can be time consuming. A hybrid method of the “checklist” form in which respondents check off consumed food items at each eating occasion may be easier than recording a complete description of the food, and the costs of data processing can be minimized (30). The checklist method may be most effective when assessing a limited set of nutrients, e.g., macronutrient intake (carbohydrates, fats, proteins, and alcohol). Another type of checklist is the close-ended method consisting of foods in different food groups in which the respondent indicates whether that food group has been consumed concurrent with actual intake (31). Portion size is recorded in an open-ended manner or in specific categories. The BalanceLog® program utilizes a checklist-type database and therefore can potentially improve recording accuracy while simultaneously reducing the recording burden on volunteers and investigators.

BalanceLog® is a computer software program that helps individuals create a personalized program to reach and maintain their weight goals. It provides a database of more than 4,000 foods and 300 exercises in which an individual can accurately monitor energy intake and expenditure. The sources of nutritional analysis for foods in the database include the U.S. Department of Agriculture (USDA) National Nutrient Databases, brand name label data, restaurant data and data obtained from manufacturers. The caloric expenditure for each exercise in the database are based on metabolic equivalents from the Compendium of Physical Activities (32), the American College of Sports Medicine guidelines (33) and manufacturer supplied data for exercise equipment.

Although BalanceLog® is used as a self-monitoring tool for individuals to monitor energy intake and expenditure, no validation studies assessing the accuracy of the PDA-based BalanceLog® program compared to energy balance calculated from TEE as measured by DLW have been performed (personal communication, Haugen, January 2005). Likewise, utilization of BalanceLog® for recording energy intake and expenditure has not been compared with data obtained from written food intake and physical activity records. Therefore, the PDA-based

BalanceLog® program is a novel, cost-effective method to estimate energy intake and expenditure in a free-living environment. The BalanceLog® tool may provide researchers a less resource-intensive approach to estimate energy intake/expenditure as well as providing Soldiers with a self-monitoring tool for weight management that reduces recording burden.

## **Hypotheses**

BalanceLog® software used in conjunction with a PDA will be as accurate as written records for assessing energy intake and energy expenditure in weight stable volunteers when compared to TEE as measured by DLW.

## **Technical Objectives**

1. Compare the accuracy of reported energy intake of written 3-day and 7-day food records and PDA-based software (BalanceLog®) against energy intake calculated from TEE as measured by DLW in weight stable volunteers.
2. Compare the accuracy of reported energy expenditure between written 3-day and 7-day activity records and PDA-based software (BalanceLog®) against TEE as measured by DLW in weight stable volunteers.

## **Military Relevance**

The 2002 Survey of Health Related Behaviors Among Military Personnel suggests that the military Services need to address rising rates of overweight among those on active duty, defined as a body mass index (BMI)  $> 25.0 \text{ kg/m}^2$  (34). The epidemic of overweight and obesity in the U.S. affects the military in that it reduces the pool of recruits that are of normal weight and decreases the retention of new recruits due to failure to adequately manage their body weights (11). As a result, Dr. William Winkenwerder, Jr., Assistant Secretary of Defense for Health Affairs, has identified overweight as one of five key challenges facing military medicine (35). With guidance from Dr. Winkenwerder and the Surgeons General, the DoD Prevention, Safety, and Health Promotion Council targeted overweight as a key health promotion issue in 2004. Clearly, weight management is a DoD targeted health concern and as a result, U. S. Army Medical Research and Materiel Command (USAMRMC) established Task Area 3.X, Weight Management Strategies, to emphasize the importance of research designed to aid overweight Soldiers.

Military personnel must successfully manage their body weight and body fat over the course of their careers or face the possibility of early discharge for noncompliance with AR 600-9, The Army Weight Control Program (36). In fact, in fiscal year 2003, 3017 Service personnel across the DoD were discharged for failure to meet weight/body fat standards; of these, 2705 were soldiers (personal communication, Grissom, September 2004). Recent reports indicate that the BMI of Army recruits is increasing as is the incidence of overweight in active duty Soldiers (37). Indeed, 62.3% of male and 32.4% of female Service personnel across the Department of Defense (DoD) are overweight, as defined by BMI  $> 25.0 \text{ kg/m}^2$  (38). Similar overweight prevalence rates of 60.1% for male and 40.6% of female Soldiers assigned to Forts Leonard Wood, Jackson, and Bragg were observed (39). We reported that 11% of male and 22% of female active duty Soldiers exceeded their screening weight-for-height and body fat standard and, therefore, meeting criteria to be placed on the Army Weight Control Program (unpublished observations, Bathalon, November 2004).

This project focuses on methods that can be considered for use in future studies of overweight and overfat Soldiers. The Army Surgeon General has authorized the use of 'Weigh-

to-Stay' as the baseline standard of care intervention for Soldiers placed on the Army Weight Control Program. This education and intervention-based program provides overweight Soldiers with various techniques to help with weight loss. One strategy for successful weight loss and maintenance as discussed in 'Weigh-to-Stay' is self-monitoring of food intake and physical activities to monitor energy balance. Therefore, the PDA-based BalanceLog® program is a potential self-monitoring tool that Soldiers might use to manage their weight. In fact, 27% of male Soldiers and 32% of female Soldiers on the Army Weight Control Program at Fort Bragg reported that use of a PDA would be of interest to them for weight management (unpublished observations, Bathalon, January 2005). The use of PDA technology could advance self-monitoring for military (as well as civilian) volunteers participating in weight loss/maintenance programs. Thus, the protocol supports efforts to assist military personnel with weight management, a key health promotion and military retention issue (40).

### **Project 3 - Response of Biomarkers of Bone Remodeling to Military Recruit Training**

#### **Background**

Stress fracture, an overuse injury to bone, is one of the most common and potentially debilitating overuse injuries seen in military recruits. Stress fractures develop when unaccustomed mechanical loading results in fatigue microdamage at sites of accelerated bone remodeling. There are reports of clinical and experimental observations of abundant remodeling and associated porosity in the vicinity of stress fractures. High rates of bone remodeling can be identified by measuring biochemical byproducts of bone resorption and formation, however the utility of using bone turnover and other biomarkers of bone remodeling to assess the bone remodeling response during short-term training regimens is not well studied.

#### **Hypotheses**

Bone turnover markers and other serum markers may be useful for monitoring the course of bone adaptation during a 16 week period of recruit training.

#### **Technical Objectives**

To determine the clinical utility of using bone turnover markers and other serum markers associated with bone remodeling as noninvasive, sensitive tools for monitoring the course of bone adaptation during a 16-week period of recruit training.

#### **Military Relevance**

Relatively little information is available regarding the effects of either acute or chronic exercise on biomarkers of bone turnover during military training regimens. The utility of using markers of bone turnover to monitor the bone remodeling response in military recruits is greatly understudied. Additionally, it is unclear whether women, who sustain stress fracture at much higher rates than men, have a greater bone turnover response to exercise. The results from this study will allow us to identify serum markers of bone formation and resorption that are sensitive to short-term physical training regimens, and that have the potential to be used in future studies that will attempt to relate observed changes in bone quality with changes in the formation:resorption ratio, particularly with respect to onset of stress fracture. Additional analyses of the relationship between bone turnover and the hormonal, dietary, and inflammatory

response may lead to dietary or other medical interventions that optimize bone health of military service members during strenuous physical training regimens.

#### **Project 4 - Blood biochemistries following repeated days of First Strike Ration or Meal, Ready to Eat consumption**

##### **Background**

The First Strike Ration (FSR) is a compact, eat-on-the-move ration designed to be consumed during short-term high-intensity missions of approximately 3 days. It substantially reduces the size and weight burden relative to current field ration (MRE), and its eat-on-move capabilities are expected to enhance consumption, nutritional intake, and mobility. Prototype versions of the FSR have received high acceptability and desirability scores (41), and the ration was recently accepted for accelerated procurement.

USARIEM actively participated on two field studies that compared the FSR prototype to MRE. Initially, the acceptability of the FSR prototype was tested with 124 Warfighters performing patrol missions in Eastern Afghanistan (41). 68% of those who ate the FSR over 3 day mission reported that they liked the FSR “moderately” or “extremely” (8 & 9 on 9 point scale), and 63% reported that they would prefer the FSR over the MRE for their next mission. Only 20% preferred the MRE over the FSR. More recently, USARIEM collaborated with the USDA Forest Service and the University of Montana (42) to perform the only performance type test of the FSR to date. In that study, the efficacy of FSR for improving voluntary work productivity and cognitive performance capability of wildland firefighters was assessed while they performed 2 days of actual fire suppression. We found that volunteers consuming the FSR self-selected to perform 30 min more moderate intensity work per day ( $p < 0.05$ ) and rest 30 min/day less ( $p < 0.05$ ) than when they were provided 2 MRE per day (41). Associated with this behavior, total energy intake ( $5250 \pm 580$  vs.  $4396 \pm 599$  kcal), carbohydrate intake ( $698 \pm 76$  vs.  $545 \pm 82$  g) and caffeine intake ( $347 \pm 262$  vs.  $55 \pm 65$  mg) were greater ( $p < 0.05$ ) when eating FSR vs. MRE, respectively, over 2 days of arduous labor. Data analysis on cognitive data is still underway. While saliva samples taken after 2 days of arduous work revealed no differences between FSR or MRE for salivary cortisol ( $0.09$  vs.  $0.11$   $\mu\text{g/dl}$ ) or testosterone levels ( $94$  vs.  $92$   $\text{pg/ml}$ ), no blood samples were taken to compare nutritional or metabolic status consequent to eating FSR vs. MRE. Equally important, the FSR consumed by the firefighters was partially manufactured by contractor and partially made by Combat Feeding Directorate. No acceptability or performance data are available on FSR made solely by contractor.

To accomplish the objectives of this study, we plan to again team up with the USDA Forest Service and the University of Montana. In preliminary discussions, both parties have provided verbal endorsement. Equally important, Hotshot Crew Chiefs have already provided verbal commitment to enable their crews to participate. Wildland firefighters offer several advantages over military units for this type of investigation. First, each team member ( $n=20$  per fire team) performs roughly the same task as his/her other team members, and all members of a fire team are exposed to same environmental situation. These two factors should minimize within group variability. As wildland firefighters have energy expenditures that are similar to those of soldiers performing patrol-type missions (43-44), the magnitude of under-eating experienced by firefighters participating in this study are expected to be representative of what can be expected by military personnel.

##### **Hypotheses**

1. Blood biochemical profiles will be similar between volunteers consuming the 1 FSR or 2 MRE during repeated days of arduous work.

2. Consumption of the FSR will be greater than consumption of the MRE.
3. Ration acceptability will be rated higher by volunteers consuming the FSR versus the MRE.

### **Technical Objectives**

1. Evaluate volunteers' biochemical blood profile after consuming either 1 FSR/day or 2 MRE/day over 3-5 days of arduous work.
2. Assess each volunteer's daily consumption of the FSR and the MRE and activity profile.
3. Assess the acceptability of the FSR versus the MRE contingent upon whether this information is desired (at time of implementation) by Combat Feeding Directorate, NSC.

### **Military Relevance**

Before the FSR can be fielded, the manufacturer produced item must meet predefined acceptability criteria. One criteria is that sufficient food will be eaten that nutrient intake is equally as good if not better than existing field ration (Meal, Ready to Eat; MRE). The purpose of this experiment is to determine if FSR sustains nutritional and metabolic status as effectively as the MRE. This project will test if FSR meets this requirement by comparing blood chemistries between volunteers who consume 1 FSR/day to volunteers who consume 2 MRE/day while performing at least 3 days of arduous work. Two MRE/day were selected as this is the amount of food typically consumed by Infantry-type Soldiers during missions where FSR would be provided. The research supports USARIEM ATO IV.MD.2005.02 (Nutritionally Optimized First Strike Ration), as well as the Ration Sustainment Testing (RST) program.

### **Project 5. Impact of Basic Combat Training on Iron Status and Cognitive Function Among Female Military Personnel**

#### **Background**

Iron is a nutritionally essential trace element known to impart a series of critical biological functions through incorporation into proteins and enzymes. Perhaps the best known function of iron is through its incorporation into heme-containing proteins, including hemoglobin for oxygen transport, myoglobin for muscle storage of oxygen, and cytochromes for the production of ATP. Iron is also known to incorporate into a series of nonheme enzymes, including NADH dehydrogenase and succinate dehydrogenase, which are required for the electron transport chain and function in energy metabolism (45). Other enzymes that require iron for biochemical function include catalase, aconitase, phosphoenolpyruvate carboxykinase, and ribonucleotide reductase.

Despite the fact that iron metabolism has been well studied and that many of the factors leading to iron deficiency have been identified, iron deficiency continues to be the most common nutritional deficiency disorder in the world (46-47). Iron deficiency occurs in stages, beginning with the depletion of body iron stores, indicated by reduced serum ferritin levels. Once iron stores have been depleted, the iron supply to tissue is reduced, as indicated by reduced serum iron concentration, increased total iron binding capacity, and diminished transferrin saturation. The final stage of iron deficiency occurs as limited serum and tissue iron supply cause depressed production of hemoglobin, resulting in anemia.

The consequences of iron deficiency range from subtle to severe, and could limit the health and performance of the modern warfighter. Perhaps the best described consequence of iron deficiency is reduced work performance. Classical human studies have demonstrated reduced performance and work productivity with varying degrees of anemia (48-50). Animal studies have demonstrated that reduced iron status results in impaired oxidative energy production in skeletal muscle, resulting in diminished capacity for prolonged exercise due to less efficient glucose oxidation and increased use of the gluconeogenic pathway (51). Other well described negative consequences of iron deficiency include diminished intellectual performance, altered body temperature regulation, reduced immunity and resistance to infections, and adverse pregnancy outcomes (52).

## **Hypotheses**

The purpose of this study is to assess the impact of BCT on the prevalence of iron deficiency and iron deficiency anemia among enlisted female military personnel. A second purpose is to determine whether changes in iron status associated with BCT have a detrimental effect on cognitive function.

As cross-sectional studies indicate an increase in the prevalence of iron deficiency and iron deficiency anemia in female military personnel immediately following BCT (53), we hypothesize that this longitudinal study will identify a similar decrement in iron status. Based upon the available literature in the field, we believe that decrements in iron status will be associated with changes in cognitive function.

## **Technical Objectives**

1. Determine the prevalence of iron deficiency and iron deficiency anemia among females upon initial entry to BCT?
2. Determine the impact of BCT on the prevalence of iron deficiency and iron deficiency anemia?
3. Determine if decrements in iron status observed during BCT associated with changes in cognitive function?

## **Military Relevance**

Iron deficiency is the most prevalent micronutrient deficiency disease in the world, affecting up to 2 billion people (46-47). Although iron deficiency occurs at greater rates in developing countries, a significant prevalence continues to be observed in the United States (US), especially among young women. Female military personnel are exposed to intense metabolic and cognitive demands as well as immune challenges, particularly during field training or when operationally deployed. Maintaining optimal iron nutriture in these women is essential, as iron deficiency and its anemia have important health implications. Changes in immune function, cognitive development and behavior, energy metabolism, and work capacity have been described in animals and humans with suboptimal iron status (45,53-54). Although studies examining iron status in female military personnel are limited, suboptimal iron intakes have been reported in female military personnel in both garrison and field training studies (55). In male military personnel, suboptimal iron intakes have been reported during field studies (56), and perturbations in iron status occurred during intense training periods (57-58).

In the present study we will determine the impact of BCT on iron status using a longitudinal design. Furthermore, we will determine whether changes in iron status associated with BCT have a detrimental effect on cognitive function. Overall, this study will provide data contributing to the formulation of nutritionally optimized rations designed to provide levels of dietary iron necessary to support the optimal cognitive function required of the female warfighter.

**Project 6.** The Effect of a Prolonged Period Aboard a Submarine on Bone Strength and Metabolism.

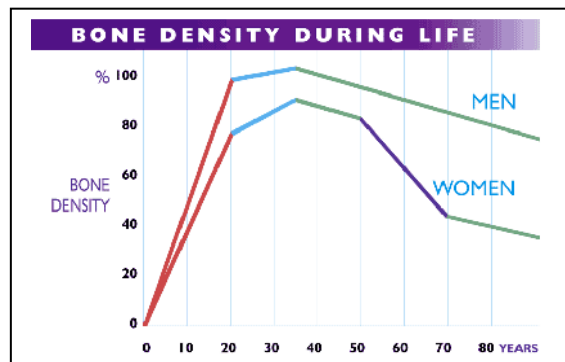
## Background

The Israeli Navy's submarine fleet operates at a considerable distance from Israel's coastline to achieve tactical and strategic goals for the State of Israel. This is a voluntary service which requires the ability to work in cramped conditions, dealing with physical and mental pressures, and maintaining a high level of concentration over a prolonged period of time, all this in an environment unnatural and hostile to man. Much effort and resources are put into improving both mechanical efficiency and human performance in the submarine fleet. To this end, new submarines have been acquired, and improvements made in existing submarines, in the attempt to meet operational demands, as well as the needs of the submariner.

Many countries throughout the world maintain and operate a submarine fleet. Leonardo da Vinci was one of the first to design a submerged vessel with human occupants that would not be visible on the surface of the water. The first submarines were built of wood. They carried a small number of occupants, reaching only the shallowest of depths, and were manually propelled. Their purpose was to approach and sink enemy vessels. In time submarines began to be constructed of metal with an internal combustion engine, which made it possible to increase the submarine's diving depth and carry out longer missions, extending the range of operations and carrying a much greater payload and crew. The advent of the nuclear era led to a significant change in the capabilities and goals of the submarine. Nuclear powered submarines are capable of remaining submerged for an extended period of time due to the immense amount of energy available from the nuclear source. They can thus carry crew and weapons over extreme distances.

A unique range of values determines the composition of the submarine's atmosphere from a physiological point of view. The air inside the vessel is monitored and kept within controlled physiological limits. (The concentration of oxygen is about 18%, CO<sub>2</sub> 0-1.2%; levels of hydrogen, carbon monoxide, cyanide gases and chlorine are also monitored.) Fresh air is introduced into the submarine by "snorkeling" (pumping in air from the surface of the water for the operation of the diesel engines and ventilation of the atmosphere). A supply of air and oxygen is also available from closed cylinders, and the carbon dioxide produced by the submariners is removed by CO<sub>2</sub> scrubbers.

Few studies have been published in the literature regarding the physiological changes resulting from a prolonged period aboard a submarine (59). There are a number of studies from the late 1970's and early 1980's which raise general questions regarding the health of the submariner, and a number of studies from recent years regarding the psychological and psychiatric status of the submariner and the primary care



given by non-physician medical staff aboard the submarine (60,61,62).

Osteoporosis is a disease characterized by low bone density. A reduction in bone density (bone mass per unit volume) diminishes the bone's mechanical strength, thus exposing it to a greater risk of fracture. Remodeling is a process that takes place throughout life, creating a balance between the formation and absorption of bone. Bone mass reaches a peak at the age of 25-30. Bone density in old age is directly dependent on the peak bone mass achieved at age 25-30, and thus if achievement of the peak bone mass is impaired at an early age, this might lead to a deficiency in bone strength in later life and a greater risk of fracture.

The principal risk factors for the development of osteoporosis include genetic background and sex (female), although a lack of physical activity, nutrition deficiency (a lack of calcium, coffee consumption), smoking, pulmonary disease causing respiratory and/or metabolic acidosis, as well as other metabolic diseases, and a lack of exposure to sunlight, represent central risk factors for injury to the bone tissue.

During a prolonged period aboard a submarine, the submariner is exposed to some of these risk factors, but with the addition of risk factors specific to the submarine that are liable to result in injury to the bone tissue. The submarine is a closed system submerged below the surface of the water, which does not permit the exposure to the sunlight required for the production of active vitamin D in the skin. The severe space restrictions aboard the vessel do not permit the movement and physical activity normally performed by the submariners outside the submarine. Nutrition aboard the submarine is not optimal, and some of the crew who work shifts consume large quantities of coffee. CO<sub>2</sub> concentrations are higher than in the open air, ranging from 0.8 to 1.2%.

A number of studies discuss the relation between lack of exposure to sunlight due to indoor confinement, long-sleeved clothing or seasonal sun deprivation, and the need for food supplements such as calcium and vitamin D. Most food products do not contain vitamin D (63). Vitamin D is produced in the body from cholesterol, is activated in the first instance by the liver to become 25-hydroxyvitamin D, and then by the kidneys or the skin via exposure to ultraviolet light to become 1,25-dihydroxyvitamin D. The active derivative of vitamin D, 1,25-dihydroxyvitamin D, keeps serum levels of calcium and potassium within normal limits, ensuring mineralization and strengthening of the skeleton, as well as normal muscle function (64,65,66). It was found that long-sleeved clothing and indoor confinement cause reduced levels of vitamin D and its products (67). One study examined the relation between 25-hydroxyvitamin D levels and levels of PTH and cortical bone density in adolescent girls in Finland. It was found that low vitamin D levels were correlated with high levels of PTH and low bone density (68). Several studies describe an increase in levels of BONE SPECIFIC ALKALINE PHOSPHATASE on exposure to sunlight (69,70). When there is a lack of exposure to sunlight, as in persons who spend a period of time at extreme latitudes, and other such circumstances, a drop is noted in 25-hydroxyvitamin D levels (71,72,73,74) and in the secretion of calcium and potassium in urine and feces (69,75,76).

A unique characteristic that affects the submarine crew is the high concentration of CO<sub>2</sub> in the inspired air. The relation between high CO<sub>2</sub> levels and bone metabolism has been investigated in COPD patients (77,66), in persons working in closed spaces (78), and even in a simulation of the submarine atmosphere in an isolation chamber (79). It was found that high levels of CO<sub>2</sub> in the air within the confined space result in reduced secretion of calcium in feces and urine, accompanied by a slight increase in the absorption and destruction of bone (79,59).

Bone is a metabolically active tissue. In recent years there have been major developments in our ability to obtain quality measurement of metabolites which indicate osteoclast and osteoblast activity. Markers such as Bone Specific Alkaline Phosphatase (BSAP) and C-Telopeptide-1 (CTP1) serve as indices for the formation and destruction of bone, and enable us to gain a better understanding of the pathophysiology of bone disorders and diseases. A



number of studies investigating the lack of exposure to sunlight and its effect on augmented bone metabolism and osteoclast activity have examined these markers (70).

Additional factors that influence bone strength include the insulin-like growth factor (IGF-I), which was found to be correlated with muscle mass and physical fitness and to enhance osteoblasts for bone formation. The opposite effect was observed with interleukin 6 (IL-6), a proinflammatory factor whose level increases in conditions of infection, inflammation, or other stress situations. Interleukin 6 inhibits the activity of IGF-I, impairing the growth of various tissues, such as muscle and bone (80).

## **Hypotheses**

1. A prolonged period aboard a submarine will cause a loss of bone strength.
2. The loss of bone strength will be expressed by a reduction in the marker of bone formation, BSAP, and an increase in the marker of bone destruction, CTP-1, as well as a reduction in the speed of sound conduction through the bone.
3. The changes in the bone will be accompanied by elevation of IL-6 and a decrease in IGF-I.
4. In submariners who maintain a higher level of physical fitness, the loss of bone strength will be less pronounced.

## **Technical Objectives**

The purpose of the proposed study is to investigate, for the first time as far as we know, the influence of a prolonged period aboard a submarine on bone strength (on markers of bone destruction and formation), while also trying to examine variables that are likely to affect these changes (physical fitness, weight, nutrition, stress). We presume that the extraordinary combination of factors prevailing in the submarine, a lack of physical activity, sunlight deprivation, and exposure to high levels of carbon dioxide, will cause significant changes in subjects' bone metabolism and bone strength.

## **Military Relevance**

A combination of factors prevailing in the submarine, a lack of physical activity, sunlight deprivation, and exposure to high levels of carbon dioxide, will cause significant changes in subjects' bone metabolism and bone strength. These changes in bone metabolism and bone strength will lead to decreased physical performance.

**Project 7** - Effects of Physical Training and Iron Status on Body Composition, Muscle Function, and Bone Health in Rats.

## **Background**

Iron is a nutritionally essential trace element known to impart a series of critical biological functions through its incorporation into proteins and enzymes. Perhaps the best known function of iron is through its incorporation into heme-containing proteins, including hemoglobin for oxygen transport, myoglobin for muscle oxygen storage, and cytochromes for ATP production. Iron is also known to incorporate into a series of nonheme enzymes, including NADH dehydrogenase and succinate dehydrogenase, which are required for the electron transport chain and function in energy metabolism (81). Other enzymes catalyzing energy metabolism

that require iron for biochemical function include catalase, aconitase, phosphoenolpyruvate carboxykinase, and ribonucleotide reductase.

Despite the fact that iron metabolism has been well studied and that many of the factors leading to iron deficiency have been identified, iron deficiency continues to be the most common nutritional deficiency disorder worldwide (82-83). Iron deficiency occurs in stages, beginning with the depletion of body iron stores, and eventually resulting in depressed production of hemoglobin, resulting in anemia. Iron deficiency could be of great concern to military personnel because of known effects on immune function, cognitive development and behavior, energy metabolism, and work capacity.

## **Hypotheses**

We will test the hypothesis that increased physical training, independent of dietary iron level, will have a negative impact on iron status. Animals with diminished iron status will display decrements in body composition, bone health and muscle function. Changes in body composition may include reduced accretion of lean body mass and diminished bone mineral content, which may be associated with reduced bone mass and strength. Decrements in muscle function may include reduced contractile force. Furthermore, changes in muscle function will be associated with differential expression of genes sensitive to iron status and physical training.

## **Technical Objectives**

1. To determine the effect of physical training on iron status in animals fed iron adequate and/or iron deficient diets.
2. To assess the effects of physical training and iron status on body composition, muscle function, and bone health.
3. To identify molecular markers of muscle function and iron status in animals subjected to physical training.
4. To compare the differential effects of iron status and physical training on body composition, muscle function, and bone health in male and female rats.

## **Military Relevance**

Iron deficiency is the most prevalent micronutrient deficiency disease in the world, affecting up to 2 billion people (82-83). Although iron deficiency and its anemia occur at greater rates in developing countries, a significant prevalence continues to be observed in the US, especially among young women. The prevalence of iron deficiency and iron deficiency anemia in female military personnel upon initial entry to the US Army is reflective of the American population, although BCT seems to impart a negative impact. In fact, the prevalence of iron deficiency and iron deficiency anemia increase by 2.4- and 3.6-fold, respectively, in enlisted female military personnel immediately following BCT (84). Although the specific causes of diminished iron status in these women have not been identified, it is likely that changes in dietary iron intake, food preference, and physical activity are contributing factors.

In the present study we will determine the effects of iron status and endurance exercise on body composition, muscle function, and bone health in a rat model. Data gleaned from this study will provide insight into the specific independent and interactive effects of iron status and endurance exercise on a number of functional and biochemical measures, thereby contributing to our understanding of these factors during military training. Furthermore, this study will provide data contributing to the formulation of nutritionally optimized rations designed to

provide levels of dietary iron necessary to support the body composition, muscle function, and bone health required of the modern Warfighter.

## **Project 8 - Efficiency of casein-whey drink for optimizing plasma amino acid availability**

### **Background**

Exercise significantly impacts protein metabolism. In general, protein breakdown and amino acid oxidation increase during exercise, with a compensatory increase in protein synthesis during recovery. The increase in protein synthesis during post-exercise recovery improves net protein balance (protein synthesis – protein breakdown), but, is dependent on nutrient availability. Provision of nutrients leads to positive net protein balance, and subsequent accretion of lean body mass (85).

Dietary amino acids (AA), provided via infusion (86-87) or orally (88-91), stimulate skeletal muscle protein synthesis. Furthermore, exercise and hyperaminoacidemia appear to have an additive effect, in that muscle protein synthesis is significantly greater when a bout of exercise precedes amino acid provision, compared to providing amino acids during resting conditions (86). Both essential and non-essential amino acids influence net skeletal muscle protein turnover following resistance exercise, but only the essential amino acids (EAA) are necessary to elicit an increase in protein synthesis (87,90). For practical purposes, EAA are too expensive to include in military rations and impart adverse tastes to food. Our goal is to identify natural proteins that best stimulate synthesis and attenuate degradation, yet are inexpensive, shelf-stable and good tasting. CAS and WP are naturally occurring proteins, containing EAA, and can be isolated from milk into a powder form. These are viable options for the formulation of a supplement.

The metabolic response to consuming CAS and WP differ, with each protein eliciting unique time-dependent changes in AA concentrations in blood due to differences in their digestion from the stomach (e.g. CAS is released slowly, WP empties rapidly) (92-93). WP ingestion produces a fast and large increase in plasma AA concentrations; however, the duration is short (Figure 1). In contrast, CAS exhibits a less pronounced but more prolonged increase in plasma AA. The plasma amino acid responses of these proteins further result in differing rates of protein synthesis and protein breakdown. Protein synthesis is stimulated to a greater degree (68%) with WP, compared to the slight increase elicited by CAS (94). However, CAS induces a marked decrease in protein breakdown (~30%) compared to WP (94). Thus, a single meal of CAS (a “slow” protein) versus WP (a “fast” protein) results in a more positive net protein balance when administered at rest (94).

The administration and subsequent measurement of labeled leucine or phenylalanine concentration changes in blood over time is a common method to assess protein metabolism. Tipton et al (95) reported that leucine uptake by the leg muscle was greater for WP (versus CAS), however WP also results in increased amino acid oxidation (94). Additionally, Tipton et al. (95) administered labeled phenylalanine, since it is not oxidized by the muscle and its disappearance is solely due to incorporation into body proteins. Phenylalanine disposal was ~35% greater for CAS than WP (P=0.06) suggesting CAS may be more effective than WP for stimulating protein accretion after resistance exercise. Although this finding was not statistically significant, it suggests that more research is needed to elucidate whether CAS has advantages versus WP.

While the aforementioned studies examined the response of CAS and WP feeding exclusively, these proteins are normally packaged with other macronutrients in a convenient, eat-on-the-move form. Literature suggests that the addition of carbohydrates and fat affects

digestion and absorption rates, thereby altering the effect on protein utilization (94). When CAS and WP are fed with carbohydrates (0.75 g/kg body weight) and fat (0.13 g/kg body weight), the differences in both protein digestion rate and protein gain are attenuated. However, WP is still more rapidly absorbed, and net protein gain remains more elevated with CAS (94).

Advantages of feeding CAS, versus WP, have also been demonstrated in a practical setting employing a resistance training program. Volunteers consuming CAS supplements as part of a high protein (i.e. 1.5 g protein/kg body weight/day) and hypocaloric diet (75-85% of predicted kcal requirements) exhibited significantly greater fat loss (7.0 kg vs. 4.2 kg), lean mass gains (4.0 kg vs. 1.4 kg), and strength gains (59% vs. 29%) (96). Furthermore, nitrogen retention was improved in volunteers consuming CAS supplements as part of this weight loss program employing resistance exercise (96).

Milk contains a combination of CAS (~80%) and WP (~20%), and is highly digestible and has the highest quality protein score (97). It has been demonstrated that milk produces a unique blood profile that captures the combined effects of WP and CAS. Specifically, milk produced the rapid increase in amino acids seen with WP as well as the abrupt insulinotropic effect; however the magnitude was lower. In addition, milk produced a blood glucose-dependent insulinotropic polypeptide response similar to that produced by CAS which is much lower than WP. Other investigators have further demonstrated that supplementation with milk, results in greater stimulation of AA uptake and net protein deposition compared to hydrolyzed soy protein (98).

Shelf and taste stability issues currently prevent the use of milk in combat rations. However, it would be advantageous to reproduce the amino acid and hormone (i.e. insulin and GIP) responses demonstrated by milk. We believe that the test drink (i.e. composed of 85% CAS and 15% WP hydrolysates) should perform like milk in regards to the proposed outcome measures; however, this has not been experimentally tested. This proposal will compare a carbohydrate beverage containing a combination of CAS-WP to carbohydrate beverages composed of either CAS-only or WP-only.

## Hypotheses

### Phase 1: At Rest

1. CAS-WP's initial (~0-120 min) postprandial areas under the curve (AUC) for amino acids will be lower than WP, but higher than CAS.
2. CAS-WP's latter (~120-240 min) postprandial AUC for amino acids will be higher than WP, but lower than CAS.
3. CAS-WP's initial (~0-120 min) postprandial AUC for insulin will be similar to WP and higher than CAS.
4. CAS-WP's initial (~0-120 min) postprandial AUC for glucose-dependent insulinotropic polypeptide (GIP) will be lower than WP.

### Phase 2: Post-Exercise

1. CAS-WP's initial (~0-120 min) postprandial areas under the curve (AUC) for amino acids will be lower than WP, but higher than CAS.
2. CAS-WP's latter (~120-240 min) postprandial AUC for amino acids will be higher than WP, but lower than CAS.
3. CAS-WP's initial (~0-120 min) postprandial AUC for insulin will be similar to WP and higher than CAS.
4. CAS-WP's initial (~0-120 min) postprandial AUC for glucose-dependent insulinotropic polypeptide (GIP) will be lower than WP.

## Technical Objectives

### Phase 1: At Rest

Evaluate the postprandial effect of three different eucaloric nutritional supplements containing the same macronutrient ratio, but different protein sources (i.e. CAS-WP contains 85% CAS/15% WP, CAS contains 100% CAS and WP contains 100% WP) on plasma amino acids, insulin, blood glucose, and glucose-dependent insulintropic polypeptide (GIP) concentrations.

### Phase 2: Post-Exercise

Evaluate the postprandial effect of three different eucaloric nutritional supplements containing the same macronutrient ratio, but different protein sources (CAS-WP: 85% CAS/15% WP, CAS contains 100% CAS and WP contains 100% WP) on plasma amino acids, insulin, blood glucose, and glucose-dependent insulintropic polypeptide (GIP) concentrations immediately after an exercise bout.

## Military Relevance

This research addresses USARIEM ATO IV.MD.2005.02 (Nutritionally Optimized First Strike Ration). It is necessary to test the scientific efficacy of lightweight, efficient, nutritionally complete, eat-on-the-move ration components that will sustain warrior performance during high intensity missions. The shelf-stability requirements established for combat rations are not met by milk or commercially-available beverage supplements. A supplement developed by the Combat Feeding Directorate (Soldier Systems Center, Natick, MA) meets these rigorous shelf-life specifications but whether it offers the advantages of milk (compared to whey- or casein-only products) remains to be tested. This protocol will evaluate three protein beverage formulations for their ability to produce a blood milieu that is apparently optimal for stimulating protein synthesis and attenuating protein oxidation and degradation.

We provide below the background information for each of the tasks that are funded in this award.

### 1. Background - Task 1: Clinical Laboratory for Human Samples

The clinical laboratory provides support for military nutrition research by providing the following services:

- a. assistance with protocol development
- b. sample collection and processing on-site or in a field setting
- c. sample analysis
- d. new method development
- e. assistance with manuscript publication

The laboratory is accredited by the Health Care Financing Administration (HCFA) and the College of American Pathologists (CAP) and participates in the lipid standardization program of the Centers for Disease Control. Good Laboratory Practices guidelines are being followed in the laboratory. The Clinical Research Laboratory is staffed by licensed medical technologists, phlebotomists, and accessioners.

The laboratory is well-equipped for performing routine and specialized tests on clinical subjects. In 2006 over 250 different analytical procedures involving 625,000 tests were performed by the lab. The laboratory is comprised of 5 departments: chemistry, special chemistry, point of care testing, hematology, and urinalysis. Testing is performed on a variety of specimen types including blood, urine, sweat, saliva, and feces. The Facilities Description in the Appendix provides a complete list of tests that are available in the Clinical Laboratory.

## **2. Background - Task 2: Stable Isotope Laboratory**

The research conducted by the Stable Isotope Laboratory is in the area of energy and water requirements, and changes in body water, of soldiers, often under harsh environmental conditions. The method used to determine energy requirements is the doubly labeled water (DLW) technique, which involves oral administration of water labeled with the stable isotopes,  $^2\text{H}$  and  $^{18}\text{O}$ . Saliva and urine samples are then obtained for periods of four to 14 days, longer with redosing. Water intake can be determined using only the  $^2\text{H}$  labeled water. The use of doubly labeled water for measurement of energy expenditure was developed as a field technique for use in small animals (99)\*. The method is based on the premise that after a loading dose of  $^2\text{H}_2^{18}\text{O}$ ,  $^{18}\text{O}$  is eliminated as  $\text{CO}_2$  and water, while deuterium is eliminated from the body as water. The rate of  $\text{CO}_2$  production, and, hence, energy expenditure, is calculated from the difference of the two elimination rates. The only requirement of subjects is to give urine and saliva specimens before and after drinking an initial dose of  $^2\text{H}_2^{18}\text{O}$ , and then return in one to two weeks to give a final urine specimen. During the period between the two urine and saliva samplings, subjects are free to carry out their normal activities and are not required to maintain extensive diaries. The doubly labeled water method has been extensively validated in humans under controlled settings (100), but there are confounding factors that need to be considered in field studies, particularly in Army Field Studies. Among these are change in location or food and water supply immediately preceding, or during an energy expenditure study. These changes may cause a change in baseline isotope abundance and, therefore, interfere with the accuracy of the energy expenditure measurement. This has occurred in a previous field training exercise involving the study of the MRE and RLW rations (101). This is a particular problem with studies such as the Ranger Training Studies (102), in which soldiers are moved to different parts of the country during the study. Therefore, a group not receiving labeled water must be followed to make any corrections in baseline isotope shifts.

Hydration status is another main focus for some Army studies. Using the cheaper and more readily available deuterium tracer, either changes in total body water (103-104) can be followed during a study, or water turnover (intake) (105-106) can be measured during a study.

One advantage of the DLW method is that it uses stable isotopes so there is no radiation exposure. The method uses two heavy isotopes of water, which are naturally occurring in food and water. There are no known side effects of either isotope at the doses given in DLW studies and has been used extensively to study energy expenditure during pregnancy (107-108) lactating women (109), and infants for measurement of energy expenditure and human milk intake (110-112).

In addition to the DLW studies, the PBRC Stable Isotope lab has provided analytical and technical support to examine protein turnover and gluconeogenesis using stable isotope tracer technology in a clinical protocol examining the effects of protein supplementation during caloric restriction (Project 1). The purpose of the study is two fold: 1) to determine if, during increased energy expenditure, physically fit individuals exhibit a smaller, larger, or equivalent increase in protein requirement as sedentary individuals and 2) to determine if, during periods of heavy

physical activity and inadequate intake, an increase in protein intake will enhance conservation of whole-body protein content.

The lab determined the appropriate methods to quantitate glucose appearance, disappearance (6,6 d<sub>2</sub> glucose) and gluconeogenesis (2-<sup>13</sup>C glycerol). The laboratory also decided on the appropriate tracers to quantitate protein synthesis (<sup>15</sup>N phenylalanine, 2,3,5,6 D<sub>4</sub> tyrosine, and <sup>15</sup>N tyrosine).

### **3. Background - Task 3: Nutrient Database Laboratory**

Assessing dietary intake is essential in determining the soldier's nutritional needs and how those needs interface with other aspects of military performance. PBRC currently participates in field studies planned and conducted by the Military Nutrition Division of USARIEM by providing assistance with and analysis of dietary intakes collected during military field studies. That participation includes the following:

- Support for USARIEM field studies requiring data collection and data entry needs
- Support for PBRC in-house Military Nutrition Tasks
- Continued programming efforts directed toward meeting computer needs of both USARIEM and PBRC Military Nutrition Tasks

The Nutrient Database Integration Laboratory provides essential services for military operations. This Task oversees the operation of MiDAS, the database containing nutrient information for all operational rations, in addition to the USDA Standard Reference Foods and the USDA Food Survey Database food files. The Task provides critical support to studies which seek to improve soldier nutrition in a variety of field settings. The Laboratory is currently integrating all Armed Forces Recipes, special formulations (MREs) and other food formulations merged into one centralized database system at PBRC to be maintained and updated for USARIEM's use. This project is entitled "MRE Nutrient Composition: Integrating MREs into the Army Database Files" and the PI is Dr. Andrew Young.

The Task Leader and her staff are proficient in all aspects of nutritional intake assessment, including the Food Frequency Questionnaire, Food Diary Analysis, and Dietary Recall. The Leader and staff have been trained in the USDA multiple pass methodology. The Task Leader is a trained food scientist and has unsurpassed knowledge of nutrient database operations.

### **4. Background - Task 4: Metabolic Unit Services**

In 1993 the PBRC Metabolic Unit was used for a special inpatient study. A description of this experience serves to illustrate how other studies might be developed to address military problems.

The PBRC served as the site for two research cohorts of U.S. Army Rangers. That project was, "Assessment of Intra- and Inter- Individual Metabolic Variation in Special Operations Forces Soldiers." The PI for the project was Ms. T. E. Jones, affiliated with the Military Nutrition Division at USARIEM. Co-Investigators were C. Gabaree, Lt. Col. T. C. Murphy, Donna Ryan, M.D., and E. Brooks, R.N., M.N.

The purpose of the study was to evaluate a group of Special Operations Forces (SOF) volunteers to determine the metabolic variation during rest, exercise and post-exercise recovery

of the individual soldiers. On June 11, 1993, 10 SOF soldiers arrived to serve as the first cohort for testing. Army personnel at the PBRC included Tanya Jones (PI), Sven Ljamo, M.D. (Medical Monitor), Catherine Gabaree (Exercise Physiologist), Lt. Col. Cliff Murphy (Dietitian) and three civilian spotters for exercise testing. The first cohort of military volunteers and civilians left the PBRC on July 1, 1993. There were minimal complications that occurred in the SOF volunteers (subungual hematomas, muscle soreness, and poison ivy dermatitis). All procedures were carried out safely and satisfactorily. A mid-course correction session at the end of the first cohort stay resulted in minor procedure adjustments. From July 9-24, 1993, 10 members of the SOF from the 10th SFG at Fort Devens, Massachusetts participated in the study. All procedures were carried out safely and satisfactorily.

The Metabolic Unit project demonstrated that carbohydrate loading produced increments in physical performance in SOF soldiers. However, the variation between individual soldiers was not great enough to support developing individualized carbohydrate supplements. As a result of this work, the SOF did not pursue a plan to develop individualized soldier supplements for SOF. Therefore, this lack of metabolic variation does not mean that carbohydrate loading would not be effective and the military will pursue carbohydrate loadings for high intensity exercise operations for our SOF soldiers.

The PBRC Metabolic Unit was also used for studies on sleep deprivation (113-114 ). These included a comparison of tyrosine, against phentermine, caffeine, and d-amphetamine during sleep deprivation with analysis of effects on sleep and on cognitive and motor performance. PBRC has also been a site for a clinical study for the evaluation of minimal or non-invasive methods for field assessment of nutritional and metabolic status. Cpt. Mark Kellogg served as the PI for this project. We have been asked by USARIEM to continue this project's availability for the upcoming grant cycle and have plans to expand our facilities to magnetic resonance spectroscopy studies to accommodate projects in 2006 and beyond.

We are expanding our Metabolic Unit capabilities with a multinuclear magnetic resonance system, installed in this year and described in this report. The system operates at strength of 3.0 Tesla and sets a new standard for state-of-the-art in situ biochemistry and imaging. The system contains multiple expandable channels and high bandwidth receivers that ensure unparalleled image reconstructions from all pulse sequences. The multinuclear system allows for the tracing of hydrogen, carbon, and phosphorus atoms within biological molecules such as glucose and fatty acids.

This instrument is critical for the development of research protocols that will determine the optimal nutrients for military performance. Similarly, this instrument can be used to probe the basic biochemistry of the cell, evaluate ergonomic aids, and identify defects in cellular metabolism that are related to peak physical performance. This technology is not currently available within the DoD research environment and will provide state-of-the-art capabilities in military nutrition research. Peak physical performance is required for the elite warfighter. At its core, physical performance is the utilization of nutrients to perform work. Most of our knowledge of the energy producing mitochondria comes from animal studies or ex-vivo studies. This is obviously an artificial situation. The emerging field of magnetic resonance spectroscopy allows us to probe the biochemistry of the cell in vivo in a non-destructive manner, with no radiation and during the performance of work. These kinds of studies are important, because our understanding of how nutrients influence cellular bioenergetics in-vivo is weak.



## **Key Research Accomplishments**

### **Task 1 - Clinical Research Laboratory for Human Samples**

- Recertification by the College of American Pathologists.
- Continued excellence in the Centers for Disease Control lipid standardization program.
- Completion of blood, saliva, sweat, and fecal testing for Project 1. Specifically, the following tests were completed: glucose, creatinine, blood urea nitrogen, cortisol, growth hormone, insulin, free fatty acids, beta hydroxybutyrate, glucagon, and IGF-1, epinephrine, norepinephrine, urine creatinine, urine total nitrogen, urine urea, urine epinephrine, urine norepinephrine, sweat total nitrogen, and fecal total nitrogen. A total of 2604 assays were completed for this project.
- Completion of blood testing for Project 3. Specifically, the following tests were completed: albumin, bone alkaline phosphatase, calcium, CTP, IL-1B, IL-6, PINP, parathyroid hormone, TNFa, tartrate resistant acid phosphatase, and vitamin D. A total of 6105 assays were completed for this project.
- Completion of blood testing for Project 4. Specifically, the following tests were completed: AST, beta hydroxybutyrate, blood urea nitrogen, free fatty acids, glucose, glycerol, prealbumin, and retinol binding protein. A total of 288 assays were completed for this project.
- Completion of blood testing for Project 5. Specifically, the following tests were completed: iron, total iron binding capacity, transferrin saturation, ferritin, zinc protoporphyrin, and soluble transferrin receptor. A total of 1385 assays were completed for this project.
- Completion of onsite collection and processing at Fort Jackson, SC for Project 6. Completion of blood testing for Project 6. Specifically, the following tests were completed: albumin, bone specific alkaline phosphatase, calcium, CTP, PINP, parathyroid hormone, tartrate resistant acid phosphatase, and vitamin D. A total of 512 assays were completed for this project.
- Completion of blood testing for Project 7. Specifically, the following tests were completed: ferritin, iron, total iron binding capacity, % transferrin saturation and soluble transferrin receptor. A total of 200 assays were completed for this project.
- Partial completion of blood testing for Project 8. Specifically, the following tests were completed: glucose, BHBA, glycerol, free fatty acids, insulin, and GIP. A total of 2292 assays were completed for this project. This project is ongoing and will continue throughout the next year.

## **Task 2 - Stable Isotope Laboratory**

- Completion of saccharic acid, glucose, and amino acid assays for 20 subjects. Work is continuing on the remaining 10 subjects.
- Completion of all assays for project 4. Total body water determination and water turnover studies were performed for 10 subjects.
- Completion of all assays for project 5. Total body water determination and water turnover studies were performed for 26 subjects.

The PBRC has added Jean-Marc Schwarz, PhD as a consultant (see cv in appendix). Dr. Schwarz is providing expertise in using stable isotopes as tracers to measure gluconeogenesis.

## **Task 3- Nutrient Database Laboratory**

- Continued development of MENU and MiDAS applications for Military Nutrition use.
- Further development of digital photography methodology for use in dietary assessment of soldiers' intakes.

Although we have not been asked to support any field studies and have been advised that there are not studies in planning, we continue to update and enhance our database capabilities to support studies, either through participation in the field or utilization of the internet as a more efficient means of access to the MENU and MiDAS databases. We can incorporate the digital photography methodology for dietary assessment of soldiers' intake for use when relevant studies are in the planning stages. Modification of this methodology and work on internet application of the databases and software programs developed for military nutrition efforts has continued.

## **Task 4 – Metabolic Unit Services**

- $^{31}\text{P}$  ATP synthesis rates by phosphocreatine turnover and resting ATPase completed and running.
- Body composition and  $^1\text{H}$  intramyocellular lipid spectra developed and running.

Although there are no currently active metabolic unit studies that we are supporting we continue to update and enhance our capabilities to support future studies through the development of advanced imaging resources. We are also working to further enhance our capabilities in skeletal muscle mitochondrial function through the finalizing and validating the  $^{31}\text{P}$  ATP synthesis rate measurement and have incorporated optical spectroscopy to measure oxy/deoxyHb oxy/deoxyMb [oxygen consumption] in order to directly measure P/O ratios. These are now operational and validation studies are in progress.

We have renovated a space in the outpatient clinical research facility to house a new Echo MRI, which will be installed in 2007. The PBRC imaging facilities now include DEXA, BodPod, echocardiography, a 3T MRI/MRS, and the soon to be installed Echo MRI.

The metabolic kitchen at PBRC has undergone administrative restructuring to incorporate the following functions: delivery of controlled feeding study diets, inpatient unit food service delivery, food intake assessments, and food hedonic assessments.

The PBRC added Timothy Church, MD, PhD to the faculty ( see CV in appendix ). Dr. Church has organized an Exercise Testing Core facility and oversees the exercise facility and the delivery of exercise interventions in that unit.

## **Reportable Outcomes**

### Task 1

#### Manuscripts

Nindl B, Alemany H, Kellogg M, Rood J, Allison S, Young A, Montain S. Utility of insulin-like growth factor-I as a biomarker for assessing short-term metabolic stress in healthy men. *AJCN* (submitted), 2006.

### Task 2

#### Abstracts

1. McClung, H., Karl, J.P., Bathalon, G.P., Smith, T.J., Sigrist, L.D., Rood, J.C., and Young, A.J. Monitoring energy intake and expenditure: A PDA provides accuracy comparable to written records. NAASO, 2006.
2. Sigrist, L.D., McClung, H.L., Smith, T.J., Karl, J.P., Bathalon, G.P., Rood, J.C. and Young, A.J. Use of a PDA to record energy intake: The minimum recording period for reliable assessment. NAASO, 2006.
3. Smith, T.J., Karl, J.P., Bathalon, G.P., McClung, H.L., Sigrist, L.D., Rood, J.C., and Young, A.J. Actical® accelerometer provides similar energy expenditure as doubly labeled water in free living volunteers. NAASO, 2006.
4. McClung, J.P., Karl, J.P., Corum, J.C., Williams, K.W., Rood, J.C., Young, A.J., and Leiberman, H.R. Longitudinal changes in iron status of enlisted female soldiers during basic combat training. FASEB (submitted), 2007.

### Task 3

None

### Task 4

None

## **Conclusions**

### **Task 1**

The clinical research laboratory and food analysis laboratory continue to provide valuable support services to enhance nutrition research in the military. The laboratory plays an important role in furthering the knowledge concerning nutrition in the military by providing routine and esoteric testing, custom method development, assistance with testing and collection protocols, field assistance with blood collection and processing, and collaboration on protocols.

### **Task 2**

The Stable Isotope Laboratory continues to provide valuable support services to enhance nutrition research in the military. The laboratory plays an important role in furthering the knowledge concerning nutrition in the military by providing research expertise and sample analysis in the area of energy and water requirements, and changes in body water. In addition to the labeled water techniques, the laboratory plays a key role in furthering knowledge on protein turnover and gluconeogenesis using stable isotope tracer technology.

### **Task 3**

We continue to position ourselves to expedite efforts on our end to incorporate state of the art technology for use in field trials in the Military Nutrition Division of USARIEM. We have developed a system that can meet the needs of field experiments in a timely manner. We will be ready to support future studies with very little lead time.

### **Task 4**

We anticipate additional development of the MRS system over the next year and becoming fully operational for mitochondrial measurements in about six months.

## **References**

1. Rose MS, Buchbinder JC, Dugan TB, Szeto EG, Allegretto JD, Rose RW, Carlson DE, Samonds KW, Schnakenberg DD. Determination of Nutrient Intakes by a Modified Visual Estimation Method and Computerized Nutritional Analysis for Dietary Assessments. *Technical Report No. T6-88*. Natick, MA, U.S. Army Research Institute of Environmental Medicine. Ref Type: Report, 1987.
2. Hallfrisch JJ, Steele P, Cohen L. Comparison of seven-day diet record with measured food intake for twenty-four subjects. *Nutr Res* 2:263-73, 1982.
3. Johnson RK, Goran MI, Poehlman ET. Correlates of over- and underreporting of energy intake in healthy older men and women. *Am J Clin Nutr* 59:1286-90, 1994.
4. Tomoyasu NJ, Toth MJ, Poehlman ET. Misreporting of total energy intake in older men and women. *J Am Geriatr Soc* 47:710-5, 1999.
5. Conway JM, Seale JL, Jacobs DR, Jr., Irwin ML, Ainsworth BE. Comparison of energy expenditure estimates from doubly labeled water, a physical activity questionnaire, and physical activity records. *Am J Clin Nutr* 75:519-25, 2002.
6. Irwin ML, Ainsworth BE, Conway JM. Estimation of energy expenditure from physical activity measures: determinants of accuracy. *Obes Res* 9:517-25, 2001.
7. Masse LC, Fulton JE, Watson KL, Mahar MT, Meyers MC, Wong WW. Influence of body composition on physical activity validation studies using doubly labeled water. *J Appl Physiol* 96:1357-64, 2004.
8. Livingstone MB, Black AE. Markers of the validity of reported energy intake. *J Nutr* 133 Suppl 3:895S-920S, 2003.
9. Boutelle KN, Kirschenbaum DS. Further Support for Consistent Self-Monitoring as a Vital Component of Successful Weight Control. *Obes Res* 6:219-24, 1998.
10. Boutelle KN, Kirschenbaum DS, Baker RC, Mitchell ME. How can obese weight controllers minimize weight gain during the high risk holiday season? By self-monitoring very consistently. *Health Psychol* 18:364-8, 1999.
11. Institute of Medicine. *Weight Management State of the Science and Opportunities for Military Programs*. Food and Nutrition Board, 2003.
12. Jakicic JM, Clark K, Coleman E, et al. American College of Sports Medicine position stand. Appropriate intervention strategies for weight loss and prevention of weight regain for adults. *Med Sci Sports Exerc* 33:2145-56, 2001.
13. Institute of Medicine. *Weight Management State of the Science and Opportunities for Military Programs*. Food and Nutrition Board, Washington, DC: National Academy Press, 2003.

14. Beaton GH. Approaches to analysis of dietary data: Relationship between planned analysis and choice of methodology. *Am J Clin Nutr* 59:253S-61S, 1994.
15. Bingham SA. Limitations of the various methods for collecting dietary intake data. *Ann Nutr Metab* 35:117-27, 1991.
16. Livingstone MB, Black AE. Markers of the validity of reported energy intake. *J Nutr* 133 Suppl 3:895S-920S, 2003.
17. Schoeller DA. Limitations in the assessment of dietary energy intake by self-report. *Metabolism* 44:18-22, 1995.
18. Nagy K. CO<sub>2</sub> production in animal: Analysis of potential errors in the doubly labeled water method. *Am J Physiol* 238:466-73, 1980.
19. Schoeller DA. Measurement of energy expenditure in free-living humans by using doubly labeled water. *J Nutr* 118:1278-89, 1988.
20. Thompson FE, Byers T. Dietary assessment resource manual. *J Nutr* 124:2245S-317S, 1994.
21. Thompson FE, Byers T. Dietary assessment resource manual. *J Nutr* 124:2245S-317S, 1994.
22. Johnson RK, Goran MI, Poehlman ET. Correlates of over- and underreporting of energy intake in healthy older men and women. *Am J Clin Nutr* 59:1286-90, 1994.
23. Tomoyasu NJ, Toth MJ, Poehlman ET. Misreporting of total energy intake in older men and women. *J Am Geriatr Soc* 47:710-5, 1999.
24. Bandini LG, Schoeller DA, Cyr HN, Dietz WH. Validity of reported energy intake in obese and nonobese adolescents. *Am J Clin Nutr* 52:421-5, 1990.
25. Scagliusi FB, Polacow VO, Artioli GG, Benatti FB, Lancha AH, Jr. Selective underreporting of energy intake in women: magnitude, determinants, and effect of training. *J Am Diet Assoc* 103:1306-13, 2003.
26. Tomoyasu NJ, Toth MJ, Poehlman ET. Misreporting of total energy intake in older men and women. *J Am Geriatr Soc* 47:710-5, 1999.
27. Taren DL, Tobar M, Hill A et al. The association of energy intake bias with psychological scores of women. *Eur J Clin Nutr* 53:570-8, 1999.
28. Goris AH, Westerterp-Plantenga MS, Westerterp KR. Undereating and underrecording of habitual food intake in obese men: selective underreporting of fat intake. *Am J Clin Nutr* 71:130-4, 2000.
29. Pirke KM, Laessle RG. *Obesity Theory and Therapy*. Stunkard AJ, Watson TA (Eds), 2nd ed. New York, NY: Raven Press, Ltd. Ref Type: Serial (Book, Monograph), 1993.
30. Thompson FE, Byers T. Dietary assessment resource manual. *J Nutr* 124:2245S-317S, 1994

31. Thompson FE, Byers T. Dietary assessment resource manual. *J Nutr* 124:2245S-317S, 1994
32. Ainsworth BE, Haskell WL, Whitt MC, et al. Compendium of physical activities: an update of activity codes and MET intensities. *Med Sci Sports Exerc* 32:S498-S504, 2000.
33. American College of Sports Medicine. American College of Sports Medicine *Guidelines for Exercise Testing and Prescription*, 5th edition. Baltimore, Maryland: Williams & Wilkins. Ref Type: Serial (Book, Monograph), 1995.
34. Bray RM, Hourani LL, Rae KL, Dever JA, Brown JM, Vincus AA, Pemberton MR, Marsden ME, Faulkner DL, Vandermaas-Peeler R. *Department of Defense Survey of Health Related Behaviors among Military Personnel, 2003*. Research Triangle Park, NC: Research Triangle Institute. Ref Type: Report, 2002
35. Funk D. Battling the Bulge: Officials plan attack on obesity in military, retirees, families. *Army Times*. Ref Type: Newspaper, February 23, 2004.
36. U.S. Department of the Army. AR 600-9, *The Army Weight Control Program*. AR 600-9. Washington, DC, Headquarters, Department of the Army. Ref Type: Report, June 10, 1987.
37. Sharp MA, Patton J, Knapik J, et al. Comparison of the physical fitness of men and women entering the U.S. Army from 1978-1998. *Med Sci Sports Exerc* 34:356-63, 2002.
38. Bray RM, Hourani LL, Rae KL, Dever JA, Brown JM, Vincus AA, Pemberton MR, Marsden ME, Faulkner DL, Vandermaas-Peeler R. *Department of Defense Survey of Health Related Behaviors among Military Personnel, 2003*. Research Triangle Park, NC: Research Triangle Institute. Ref Type: Report, 2002.
39. Bathalon GP, McGraw SM, Friedl KE, Sharp MA, Williamson DA, Young AJ. *Rationale and Evidence Supporting Changes to the Army Weight Control Program*. Technical Report T04-08. Natick, MA, U.S. Army Research Institute of Environmental Medicine. Ref Type: Report, 2004.
40. Funk, D. *Battling the Bulge: Officials plan attack on obesity in military, retirees, families*. Army Times. Ref Type: Newspaper, February 23, 2004.
41. Montain, SJ, C Koenig, S McGraw. First Strike Ration acceptability: dismounted combat soldiers in Afghanistan. USARIEM Technical Report T06-02 (AD A444070), 2006.
42. Montain, SJ, CJ Baker-Fulco, PJ Niro, A Reinert, J Domitrovich, and BC Ruby. Eat-on-move rations improve actimetry scores during wildland fire suppression. *Med Sci Sports Exerc* 38: S36, 2006b (Abstract)
43. Ruby, BC, TC Shriver, TW Zderic, BJ Sharkey, C Burks, and S Tysk. Total energy expenditure during arduous wildfire suppression. *Med Sci Sports Exerc* 34: 1048–1054, 2002.

44. Tharion WJ, HR Lieberman, SJ Montain, AJ Young, CJ Baker-Fulco, JP DeLany, and RW Hoyt. Energy requirements of military personnel. *Appetite* 44:47-65, 2005
45. Dallman PR: Biochemical basis for the manifestations of iron deficiency. *Annu Rev Nutr* 6:13-40, 1986.
46. DeMaeyer E, Adiels-Tegman, M: The prevalence of anemia in the world. *World Health Stat Q* 38:302-16, 1985.
47. Stoltzfus RJ: Defining iron-deficiency anemia in public health terms: time for reflection. *J Nutr* 131:565S-567S, 2001.
48. Viteri FE, Torun B: Anemia and physical work capacity. *Clin Hematol* 3:609-626, 1974.
49. Basta SS, Soekiman MS, Karyadi D, Scrimshaw NS: Iron deficiency anemia and the productivity of adult males in Indonesia. *Am J Clin Nutr* 32:916-925, 1979.
50. Edgerton VR, Gardner GW, Ohirya Y, Gunawardena KA, Senewiratne B: Iron-deficiency anemia and its effect on worker productivity and activity patterns. *Br Med J* 2:1546-1549, 1979.
51. Davies KJA, Donovan CM, Refino CA, Brooks GA, Packer L, Dallman PR: Distinguishing effects of anemia and muscle iron deficiency on exercise bioenergetics in the rat. *Am J Physiol* 246:E35-E543, 1984.
52. Yip R, Dallman PR: Iron, In: *Present Knowledge in Nutrition*. ILSL Press, Washington, D. C. pp 277-292, 1996.
53. McClung JP, Marchitelli LJ, Friedl KE, Young AJ: Prevalence of iron deficiency and iron deficiency anemia among three populations of female soldiers in the US Army. *J Am Coll Nutr* (In Press), 2006.
54. Haas JD, Brownlie T: Iron deficiency and reduced work capacity: A critical review of the research to determine a causal relationship. *J Nutr* 131:676S-690S; 2001.
55. King N, Fridlund KE, Askew EW: Nutrition issues of military women. *J Am Coll Nutr* 12:344-348, 1993.
56. Cline AD, Tharion WJ, Tulley RT, Hotson N, Lieberman HR: Influence of a carbohydrate drink on nutritional status, body composition and mood during desert training. *Aviat Space Environ Med* 71:37-44, 2000.
57. Singh A, Smoak BL, Patterson KY, LeMay LG, Veillon C, Deuster PA: Biochemical indices of selected trace minerals in men: effect of stress. *Am J Clin Nutr* 53:126-131, 1991.
58. Moore RJ, Friedl KE, Tulley RT, Askew EW: Maintenance of iron status in healthy men during an extended period of stress and physical activity. *Am J Clin Nutr* 58:923-927, 1993.
59. Messier AA, Heyder E, Braithwaite WR, McCluggage C, Peck A, Schaefer KE.



Calcium, magnesium, and phosphorus metabolism, and parathyroid-calcitonin function during prolonged exposure to elevated CO<sub>2</sub> concentrations on submarines. *Undersea Biomed Res.* 1979;6 Suppl:S57-70.

60. Burr RG, Palinkas LA. Health risks among submarine personnel in the U.S. Navy, 1974-1979. *Undersea Biomed Res.* 1987 Nov;14(6):535-44.
61. Jan MH, Thomas TL, Hooper TI. Prescription medication use aboard US submarine during periods underway. *Undersea Hyperb Med.* 2002 Winter; 29(4): 294-306.
62. Thomas TL, Hooper TI, Camarca M, Murray J, Sack D, Mole D, Spiro RT, Horn WG, Garland FC. A method for monitoring the health of US Navy submarine crewmembers during periods of isolation. *Aviat Space Environ Med.* 2000 Jul;71(7):699-705.
63. Fraser DR. Biochemical and clinical aspects of vitamin D function. *Br Med Bull.* 1981 Jan;37(1):37-42.
64. Zittermann A. Vitamin D in preventive medicine: are we ignoring the evidence? *Br J Nutr.* 2003 May;89(5):552-72.
65. Mawer EB, Davies M. Vitamin D nutrition and bone disease in adults. *Rev Endocr Metab Disord.* 2001 Apr;2(2):153-64.
66. Dimai HP, Domej W, Leb G, Lau KH. Bone loss in patients with untreated chronic obstructive pulmonary disease is mediated by an increase in bone resorption associated with hypercapnia. *J Bone Miner Res.* 2001 Nov;16(11): 2132-41.
67. Glerup H, Mikkelsen K, Poulsen L, Hass E, Overbeck S, Thomsen J, Charles P, Eriksen EF. Commonly recommended daily intake of vitamin D is not sufficient if sunlight exposure is limited. *J Intern Med.* 2000 Feb;247(2):260-8.
68. Cheng S, Tylavsky F, Kroger H, Karkkainen M, Lyytikainen A, Koistinen A, Mahonen A, Alen M, Halleen J, Vaananen K, Lamberg-Allardt C. Association of low 25-hydroxyvitamin D concentrations with elevated parathyroid hormone concentrations and low cortical bone density in early pubertal and prepubertal Finnish girls. *Am J Clin Nutr.* 2003 Sep;78(3):485-92.
69. Edwards HM Jr, Elliot MA, Sooncharernying S, Britton WM. Quantitative requirement for cholecalciferol in the absence of ultraviolet light. *Poult Sci.* 1994 Feb;73(2):288-94.
70. Yonei T, Hagino H, Katagiri H, Kishimoto H. Bone metabolic changes in Antarctic wintering team members. *Bone.* 1999 Feb;24(2):145-50.
71. Guzel R, Kozanoglu E, Guler-Uysal F, Soyupak S, Sarpel T. Vitamin D status and bone mineral density of veiled and unveiled Turkish women. *J Womens Health Gend Based Med.* 2001 Oct;10(8):765-70.

72. Diamond TH, Levy S, Smith A, Day P. High bone turnover in Muslim women with vitamin D deficiency. *Med J Aust.* 2002 Aug 5;177(3):139-41.
73. Schmidt-Gayk H, Bouillon R, Roth HJ. Measurement of vitamin D and its metabolites (calcidiol and calcitriol) and their clinical significance. *Scand J Clin Lab Invest Suppl.* 1997;227:35-45.
74. Sack DM, Holick M, Bondi KR. Calcium and vitamin D metabolism in submariners: carbon dioxide, sunlight, and absorption considerations. Groton, CT: Naval Submarine Medical Research Laboratory, 1986; NSMRL Report No. 1037
75. Pettifor JM, Moodley GP, Hough FS, Koch H, Chen T, Lu Z, Holick MF. The effect of season and latitude on in vitro vitamin D formation by sunlight in South Africa. *S Afr Med J.* 1996 Oct;86(10):1270-2.
76. Thomas MK, Lloyd-Jones DM, Thadhani RI, Shaw AC, Deraska DJ, Kitch BT, Vamvakas EC, Dick IM, Prince RL, Finkelstein JS. Hypovitaminosis D in medical inpatients. *N Engl J Med.* 1998 Mar 19;338(12):777-83.
77. Schaefer KE, Pasquale S, Messier AA, Shea M. Phasic changes in bone CO<sub>2</sub> fractions, calcium, and phosphorus during chronic hypercapnia. *J Appl Physiol.* 1980 May;48(5):802-11.
78. Zerath E, Holy X, Gaud R, Schmitt D. Decreased serum levels of 1,25-(OH)<sub>2</sub> vitamin D during 1 year of sunlight deprivation in the Antarctic. *Eur J Appl Physiol Occup Physiol.* 1999 Jan;79(2):141-7.
79. Drummer C, Friedel V, Borger A, Stormer I, Wolter S, Zittermann A, Wolfram G, Heer M. Effects of elevated carbon dioxide environment on calcium metabolism in humans. *Aviat Space Environ Med.* 1998 Mar;69(3):291-8.
80. Ferrucci L, Guralnik JM. Inflammation, hormones, and body composition at a crossroad. *Am J Med* 2003 Oct;115(6):501-2.
81. Dallman PR: Biochemical basis for the manifestations of iron deficiency. *Annu Rev Nutr* 6:13-40, 1986.
82. DeMaeyer E, Adiels-Tegman, M: The prevalence of anemia in the world. *World Health Stat Q* 38:302-16, 1985.
83. Stoltzfus RJ: Defining iron-deficiency anemia in public health terms: time for reflection. *J Nutr* 131:565S-567S, 2001.
84. McClung JP, Marchitelli LJ, Friedl KE, Young AJ: Prevalence of iron deficiency and iron deficiency anemia among three populations of female soldiers in the US Army. *J Am Coll Nutr* 25:64-69, 2006a.
85. Tipton KD, Wolfe RR. Exercise-induced changes in protein metabolism. *Acta Physiol Scand* 1998; 162:377-87.

- 86 . Biolo G, Tipton KD, Klein S, Wolfe RR. An abundant supply of amino acids enhances the metabolic effect of exercise on muscle protein. *Am J Physiol* 1997;273:E122-E129.
- 87 . Volpi E, Ferrando AA, Yeckel CW, Tipton KD, Wolfe RR. Exogenous amino acids stimulate net muscle protein synthesis in the elderly. *J Clin Invest* 1998;101:2000-7.
88. Borsheim E, Tipton KD, Wolf SE, Wolfe RR. Essential amino acids and muscle protein recovery from resistance exercise. *Am J Physiol Endocrinol Metab* 2002;283:E648-E657.
89. Miller SL, Tipton KD, Chinkes DL, Wolf SE, Wolfe RR. Independent and combined effects of amino acids and glucose after resistance exercise. *Med Sci Sports Exerc* 2003;35:449-55.
90. Tipton KD, Ferrando AA, Phillips SM, Doyle D, Jr., Wolfe RR. Postexercise net protein synthesis in human muscle from orally administered amino acids. *Am J Physiol* 1999;276:E628-E634.
91. Tipton KD, Borsheim E, Wolf SE, Sanford AP, Wolfe RR. Acute response of net muscle protein balance reflects 24-h balance after exercise and amino acid ingestion. *Am J Physiol Endocrinol Metab* 2003;284:E76-E89.
92. Boirie Y, Fauquant J, Rulquin H, Maubois JL, Beaufrere B. Production of large amounts of [<sup>13</sup>C]leucinenriched milk proteins by lactating cows. *J Nutr* 1995;125:95-8.
93. Dangin M, Boirie Y, Garcia-Rodenas C et al. The digestion rate of protein is an independent regulating factor of postprandial protein retention. *Am J Physiol Endocrinol Metab* 2001;280:E340-E348.
94. Dangin M, Boirie Y, Guillet C, Beaufrere B. Influence of the protein digestion rate on protein turnover in young and elderly subjects. *J Nutr* 2002;132:3228S-33S.
95. Tipton KD, Elliot TA, Cree MG, Wolf SE, Sanford AP, Wolfe RR. Ingestion of casein and whey proteins result in muscle anabolism after resistance exercise. *Med Sci Sports Exerc* 2004;36:2073-81.
96. Demling RH, DeSanti L. Effect of a hypocaloric diet, increased protein intake and resistance training on lean mass gains and fat mass loss in overweight police officers. *Ann Nutr Metab* 2000;44:21-9.
97. Schaafsma G. The Protein Digestibility-Corrected Amino Acid Score (PDCAAS)—a concept for describing protein quality in foods and food ingredients: a critical review. *J AOAC Int* 2005;88:988-94.
98. Phillips SM, Harman JW, Wilkinson SB. Dietary protein to support anabolism with resistance exercise in young men. *J Am Coll Nutr* 2005;24:134S-9S.
99. Lifson N. Theory of use of the turnover rates of body water for measuring energy material balance. *J Theoret Biol* 12:46-74, 1966.

100. Schoeller DA. Measurement of energy expenditure in free-living humans by using doubly labeled water. *J Nutr* 118:1278-1289, 1988.
101. DeLany JP, Schoeller DA, Hoyt RW, Askew EW, Sharp MA. Field Use of D<sub>2</sub><sup>18</sup>O to Measure Energy Expenditure of Soldiers at Different Energy Intakes. *J Appl Physiology* 67:1922-1929, 1989.
102. Hoyt RW, Moore RJ, DeLany KP, Friedl LE, Askew EW. Energy balance during 62 days of rigorous physical activity and caloric restriction. *FASEB J* 7:A726, 1993 (Abs).
103. Schoeller DA, van Santen E, Peterson DW, Dietz W, Jaspen J, Klein PD. Total body water measurement in humans with <sup>18</sup>O and <sup>2</sup>H labeled water. *Am J Clin Nutr* 33:2686-93, 1980.
104. Lyons TP, Muza SR, Cymerman A, DeLany JP. Total body water and plasma volume responses to high altitude acclimatization and deacclimatization. *FASEB J* 8:A288, 1994 (Abs).
105. Fjeld CR, Brown KH, Schoeller DA. Validation of the deuterium oxide method for measuring average daily milk intake in infants. *Am J Clin Nutr* 48:671-679, 1988.
106. Jones TE, Hoyt RW, DeLany JP, Hesslink RL, Askew EW. A comparison of two methods of measuring water intake of soldiers in the field. *FASEB J* 7:A610, 1993 (Abs).
107. Heini A, Schutz Y, Diaz E, Prentice AM, Whitehead RG, Jéquier E. Free-living energy expenditure measured by two independent techniques in pregnant and nonpregnant Gambian women. *Am J Physiol* 261:E9-E17, 1991.
108. Goldberg GR, Prentice AM, Coward WA, Davies HL, Murgatroyd PR, Wensing C, Black AE, Harding M, Sawyer M. Longitudinal assessment of energy expenditure in pregnancy by the doubly labeled water method. *Am J Clin Nutr* 57:494-505, 1993.
109. Goldberg GR, Prentice AM, Coward WA, Davies HL, Murgatroyd PR, Sawyer MB, Ashford J, Black AE. Longitudinal assessment of the components of energy balance in well-nourished lactating women. *Am J Clin Nutr* 54:788-798, 1991.
110. Roberts SB, Coward WA, Ewing G, Savage J. Effect of weaning on accuracy of doubly labeled water method in infants. *Am J Physiol* 254:R622-R627, 1988.
111. Fjeld CR, Brown KH, Schoeller DA. Validation of the deuterium oxide method for measuring average daily milk intake in infants. *Am J Clin Nutr* 48:671-679, 1988.
112. Butte NF, Wong WW, Patterson BW, Garza C, Klein PD. Human-milk intake measured by administration of deuterium oxide to the mother: a comparison with the test-weighing technique. *Am J Clin Nutr* 47:815-821, 1988.
113. Waters WF, Magill RA, Bray GA, Volaufova J, Smith SR, Lieberman HR, Hurry M, Anderson T, Ryan DH. A comparison of tyrosine, against phentermine, caffeine, and d-amphetamine during sleep deprivation. *Nutritional Neuroscience* 6(4):221-235, 2003.

114. Magill RA, Waters WF, Bray GA, Volaufova J, Smith SR, Lieberman HR, McNevin N, Ryan DH. Effects of tyrosine, phentermine, caffeine, d-amphetamine, and placebo on cognitive and motor performance deficits during sleep deprivation. *Nutritional Neuroscience* 6(4):237-246, 2003.

## **Appendices**

## **CURRICULUM VITAE**

**Timothy S. Church, M.D., M.P.H, Ph.D.**

### **Work Address**

Tim Church, MD, MPH, PhD  
Pennington Biomedical Research Center Louisiana State University System  
6400 Perkins Road  
Baton Rouge, Louisiana 70808-4124

Email: tim.church@pbrc.edu  
Phone: 225-763-2632  
Fax: 225-763-3014

### **Education**

Tulane University School of Public Health and Tropical Medicine  
Degree: Masters of Public Health, Epidemiology Tract  
May 2000

Tulane University School of Medicine, New Orleans, Louisiana.  
Department of Structural and Cellular Biology  
Degree: Ph.D.  
Thesis: Effect of Exercise on Arteriolar Blood Rheology in Health and Cardiovascular Disease  
May 1998

Tulane University School of Medicine, New Orleans, Louisiana.  
DEGREE: M.D.  
JUNE 1996

University of California at Davis, California.  
DEGREE: B.S. ANIMAL PHYSIOLOGY  
JUNE 1991

### ***Appointments***

Professor  
John S. McIlhenny Endowed Chair of Health Wisdom  
Director Laboratory of Preventive Medicine Research  
Pennington Biomedical Research Center Louisiana State University System, Baton Rouge, LA  
September 2006 - present

Adjunct Associate Professor

University of South Carolina  
Department of Exercise Science  
July 1, 2005 - present

Vice-President  
Medical and Laboratory Research  
The Cooper Institute, Dallas, Texas  
November 2002 – September 2006

Senior Associate Director  
Medical and Laboratory Director  
Division of Epidemiology and Clinical Applications  
The Cooper Institute, Dallas, Texas  
July 2000 – November 2002

Chief Resident- Tulane Medical Center Preventive Medicine Residency Program  
Tulane Medical Center, New Orleans, LA  
July - December 1999

Tulane Medical Center Preventive Medicine Residency Program  
Tulane Medical Center, New Orleans, LA  
July 1998 - July 2000

Transitional Medicine Residency  
Presbyterian-St. Luke Hospital, Denver, CO  
June 1997 - June 1998

### **Professional Licensure and Memberships**

**Louisiana State Medical License**  
October 2006 - Present

**Texas State Medical License**  
January 2001 - Present

**Specialist in Public Health and General Preventive Medicine**  
**Certified by the American Board of Preventive Medicine**  
January 2001 - Present

**Member of the American College of Sports Medicine and American Heart Association**

### **Research Grants and Contracts**

Principal Investigator – Health Benefits of Aerobic and Resistance Training in Individuals with Diabetes (HART-D), NIDDK RO-1, 2006-2010, \$1,610,176 direct costs over four years.

Co-Investigator (Barbara Nicklas, PI) Exercise Training and Inflammation Risk Factors for Disability, NIA RO1, 2006-2010

Principal Investigator – Inflammation (C-reactive protein) and Exercise (INFLAME), NHLBI RO1, 2004-2008, \$1,383,642 direct costs over four years.

Co-Investigator (Tuomo Rankinen, PI) – Hypertension and Genetics (HYPGENE), NHLBI RO1, 2003-2007.

Site Director & Investigator- (Marco Pahor, PI) Physical Exercise to Prevent Disability Pilot Study (LIFE), NIH/NIA UO1 AG022376-01 2003-2007

Co-Investigator (Madhukar Trivedi, PI) – Exercise & Depression: Dose, Treatment, Adjunct Effects (TREAD), NIMH RO1, 2002-2007.

Co-Investigator (Kevin Oeffinger, PI) Physical activity intervention in adult survivors of child hood leukemia (ALLIFE), NCI RO1, 2003-2007

Co-Investigator – (Steve Blair, PI) Diastolic Function and exercise in postmenopausal women, NHBLI – Innovative Research Grant Program- 2004-2006.

National Institutes of Health Clinical Research Loan Repayment Program Award Recipient. September 2003 to September 2007.

Co-Investigator – (Jean-Pierre Despres, PI) Fatness, Fitness, and Features of Metabolic Syndrome in Postmenopausal Women: A Dose Response Study of the Effect of Endurance Exercise Training. Canadian Diabetes Association, 2004-2006.

Principal Investigator, Rimonabant in Prediabetic Subjects to Delay Onset of type 2 Diabetes (RAPSODI - Protocol EFC5107); Sanofi-Aventis. 2006

Principal Investigator, Buprion SR plus Zonisamide in the Treatment of Subjects with Uncomplicated Obesity(Protocol ZB201); Orexigen. 2006

Project Director and Co-Investigator (Steven N. Blair, P.I.) – Dose-Response in Postmenopausal Exercise in Women (DREW), NHLBI R01 HL66262, 2001-2005. Completed

Co-Investigator – Coronary artery calcium test and CHD outcome-continuation grant, NHLBI RO-1, 2002-2005. Completed

Principal Investigator – C-reactive protein and exercise in postmenopausal women, NHBLI – Innovative Research Grant Program- RO1 HL071900-01, 2002-2004, \$200,000 direct costs over two years. Completed

Principal Investigator- Identifying effective community based childhood obesity prevention and treatment programs for wide spread dissemination. Michael and Susan Dell foundation, \$250,000. Completed

Principal Investigator - Cooper Complete Vitamin Study- Privately funded-\$340,000 total costs. Completed

Clinical Site-Medical Supervisor and Co-Director, Adolescent Weight-Loss Study of Meridia (Protocol SB238); Knoll Pharmaceutical Company. Completed



Principal Investigator – Heart Rate Variability and exercise in postmenopausal women, American Heart Association-Texas Affiliate Beginning Grant in Aid, 0265140Y, 2002-2004, \$128,000 direct costs over 2 years. Completed

## **Publications**

Kirby, G.S., Church, T.C., Beecherl, E.E., Barron, O.A., Smith, J.L. and Terkildsen, M.W. A rapid response microviscosimeter. *Biorheology*, 35:1 (1998); 89-102.

Church, T.S., Gibbons, L.W., Kampert, J.B., Barlow, C.E., and Blair, S.N. Usefulness of cardiorespiratory fitness as a predictor of all-cause and cardiovascular disease mortality in men with systemic hypertension. *American Journal of Cardiology* 88 (2001); 651-656.

Church T.S., Kampert, J.B., Wilkinson, W.J, Dunn, A.L., Blair, S.N. Evaluating the reproducibility and validity of the Aerobic Adaptation Test. *Medicine and Science in Sports and Exercise* 33:10 (2001); 1770-1773.

Church, T.S., Lavie, C.J., Milani, R.V., Kirby, G.S. Improvements in blood rheology following cardiac rehabilitation and exercise training in patients with coronary heart disease. *American Heart Journal* 143:2 (2002); 349-355.

Church, T.S., Finley, C.E., Kampert, J.B, Gibbons, L.W., Blair, S.N. Relative associations of fitness and fatness to fibrinogen, white blood cell count, uric acid and metabolic syndrome. *International Journal of Obesity Research* 26:6 (2002); 805-13.

Church, T.S., Earnest, C.P. Morss, G. Field-Testing of physiological responses associated with Nordic Walking. *Research Quarterly*, 73:3 (2002);296-300.

Church, T.S., Barlow, C.E., Earnest, C.P., Kampert, J.B., Priest, E.L., Blair, S.N. Associations of cardiorespiratory fitness to C-reactive protein in healthy men. *Arteriosclerosis, Thrombosis, Vascular Biology*, 22(2002); 1869 - 1876

Cheng, YJ, Macera, CA, Church, TS, Blair, SN. Heart rate reserve as a predictor of cardiovascular and all-cause mortality in men *Medicine and Science in Sports and Exercise* 34:12(2002); 1873-1878

Nguyen-Duy, TB, Nichaman, MZ, Church,TS, Blair, SN, Ross R. Visceral fat and liver fat are independent predictors of metabolic risk factors in men. *Am J Physiol Endocrinol Metab* 2003;284(6):E1065-71

Cheng YJ, Lauer MS, Earnest CP, Church TS, Kampert JB, Gibbons LW and Blair SN; Heart Rate Recovery Following Maximal Exercise Testing as a Predictor of Cardiovascular Disease and All-Cause Mortality in Men with Diabetes. *Diabetes Care* 2003 Jul;26(7):2052-7

Cheng YJ, Church TS, Kimball TE, Nichaman MZ, Levine BD, McGuire DK, and Blair SN. Coronary Artery Calcium Detected by Electron Beam Tomography and its Association with Coronary Heart Disease in 17,967 Women and Men. *Am Journal of Cardiology* 2003 Sep 1;92(5):498-503

Earnest CP and Church TS. Complex Multivitamin Supplementation Affects Homocysteine and LDL-C Oxidation. *Journal of American College of Nutrition* 2003 Oct; 22(5):400-407

Church TS, Earnest CP, Wood KA and Kampert JB. Reduction of C-reactive protein concentrations through use of a multivitamin. *Am Journal of Medicine* 2003 Dec 15; 115(9):702-707

Church TS, Cheng YJ, Earnest CP, Barlow, C.E., Gibbons, L.W., Priest, E.L., Blair, S.N. Exercise Capacity and Body Composition as Predictors of Mortality among Men with Diabetes. *Diabetes Care* 2004 Jan; 27(1): 83-88

Wong, S, Katzmarzyk,PT, Nichaman, MZ, Church,TS, Blair, SN, Ross R. Cardiorespiratory fitness is associated with lower abdominal adiposity independent of body mass index in men. *Medicine and Science in Sports and Exercise* 2004 Feb; 36(2): 286-291

Morss GM, Jordan AN, Skinner JS, Dunn AL, Church TS, Earnest CP, Jurca R, Blair SN. The Dose-Response to Exercise in Women Aged 45-75 Years (DREW) Study: *Med Sci Sports Exerc* 2004 Feb; 36(2): 336-344

Earnest CP, Morss GM, Wyatt F, Jordan AN, Colson S, Church TS, Fitzgerald Y, Autrey L, Jurca R, Lucia A. Effects of a commercial herbal-based formula on exercise performance in cyclists. *Med Sci Sports Exerc.* 2004 Mar;36(3):504-9.

Jurca R, Church TS, Jordan AN, Morss GM, Earnest CP. Eight Weeks of Moderate Intensity Exercise Training Increases Heart Rate Variability in Sedentary Postmenopausal Women. *Am Heart Journal* 2004 May;147(5):e21

Katzmarzyk PT, Church TS and Blair SN. Cardiorespiratory Fitness Attenuates the Effects of the Metabolic Syndrome on All-Cause and Cardiovascular Disease Mortality in Men. *Archives of Internal Medicine* 2004 May 28;164(10): 1092-1097

Jackson AS, Kampert JB, Barlow CE, Morrow JR Jr, Church TS, Blair SN. Longitudinal Changes in Cardiorespiratory Fitness: Measurement Error or True Change? *Med Sci Sports Exerc.* 2004 Jul;36(7):1175-1180

Jordan AN, Jurca R, Abraham EH, Salikhova A, Mann JK, Morss GM, Church TS, Lucia A, Earnest CP. Effects of oral ATP supplementation on anaerobic power and muscular strength. *Med Sci Sports Exerc.* 2004 Jun;36(6):983-90

Kuk JL, Nichaman M, Church TS, Blair SN, and Ross R. Liver fat is not a marker of metabolic risk in lean premenopausal women. *Metabolism.* 2004 Aug;53(8):1066-71

Jurca R, LaMonte MJ, Church TS, Earnest CE, FitzGerald SJ, Barlow CE, Jordan AN, Kampert JB, Blair SN. Associations of Muscle Strength and Aerobic Fitness with Metabolic Syndrome in Men. *Med Sci Sports Exerc* Med Sci Sports Exerc. 2004 Aug;36(8):1301-1307

Earnest CP, Jurca R, Church TS, Chicharro JL, Hoyos J, Lucia A. Relationship Between Physical Exertion and Heart Rate Variability Characteristics in Professional Cyclists During The Tour Of Spain. *Br J Sports Med* 2004 Sept;38 568-575

Katzmarzyk PT, Church TS, Janssen I, Ross R, and Blair SN. Metabolic Syndrome, Obesity and Mortality: Impact of Cardiorespiratory Fitness. *Diabetes Care* 2005 Feb; 28(2): 391-397

Lee SJ, Kuk JL, Katzmarzyk PT, Blair SN, Church TS, Ross R. Cardiorespiratory Fitness Attenuates Metabolic Risk Independent of Abdominal Subcutaneous and Visceral Fat in Men. *Diabetes Care* 2005 Apr; 28(4):895-901

Church TS, Willis MS, Priest EL, Lamonte MJ, Earnest CP, Wilkinson WJ, Wilson DA, Giroir BP. Obesity, macrophage migration inhibitory factor, and weight loss. *Int J Obes Relat Metab Disord.* 2005 Jun;29(6):675-81.

Earnest CP, AN Jordan, M Safir, E Weaver, and TS Church. Cholesterol-lowering effects of bovine serum immunoglobulin in participants with mild hypercholesterolemia. *Am J Clin Nutr.* 2005 Apr;81(4):792-8.

LaMonte MJ, Barlow CE, Jurca R, Kampert JB, Church TS, and SN Blair  
Cardiorespiratory Fitness Is Inversely Associated With The Incidence Of Metabolic Syndrome. A 6-Year Prospective Study of Men and Women. *Circulation.* 2005;112; 505-512.

Arden C.I., P.T. Katzmarzyk, I. Janssen, T.S. Church and S.N. Blair. Adult Treatment Panel III Guidelines and cardiovascular disease mortality. *Circulation.* 2005;112; 1481-1488.

A.N. Jordan, Jurca, G.M., Tudor-Locke, C., Church, T.S., and Blair, S.N. (2005). Pedometer Indices for Weekly Recommendations in Postmenopausal Women. *Medicine and Science in Sport and Exercise.* 2005 Sep;37(9):1627-1632

LaMonte MJ, FitzGerald SJ, Church TS, Wright CB, Radford NB, Levine BD, Pippin JJ, Gibbons LW, Blair SN and, Nichaman MZ. Coronary Artery Calcium Score Predicts Coronary Heart Disease Events in a Large Cohort of Asymptomatic Men and Women. *Am J Epidemiol.* 2005 Sep 1;162(5):421-9

Janssen I, Katzmarzyk PT, Church TS, and Blair SN. Development of a Clinical Score Sheet for Predicting All-Cause Mortality in Men: The Cooper Clinic Mortality Risk Index. *Am J Prev Med.* 2005 Oct;29(3):194-203.

Jurca R, Jackson AS, Lamonte MJ, Morrow JR Jr, Blair SN, Wareham NJ, Haskell WL, van Mechelen W, Church TS, Jakicic JM, Laukkanen R. Assessing cardiorespiratory fitness without performing exercise testing. *Am J Prev Med.* 2005 Oct;29(3):185-93

Church TS, LaMonte MJ, Barlow CE, and Blair SN. Cardiorespiratory Fitness and Body Mass as Predictors of Cardiovascular Disease Mortality among Men with Diabetes. *Archives of Internal Medicine.* 2005 Oct 10;165(18):2114-20.

Jurca R, Lamonte MJ, Barlow CE, Kampert JB, Church TS and Blair SN. Association of muscular strength with incidence of metabolic syndrome in men. *Med Sci Sports Exerc.* 2005 Nov; 37(11):1849-55.

Davidson LE, Kuk JL, Church TS, and Ross R. Protocol for Measurement of Liver Fat by Computed Tomography. *J Appl Physiol.* 2006 Mar; 100(3):864-8.

Katzmarzyk PT, Janssen I, Church TS, Ross R and Blair SN. The Importance of Waist Circumference in the Definition of Metabolic Syndrome: Prospective Analyses of All-cause and CVD Mortality in Men. *Diabetes Care.* 2006 Feb; 29(2):404-409.

LaMonte MJ, FitzGerald SJ, Levine BD, Church TS, Kampert JB, Nichaman MZ, Gibbons LW, and Blair SN. Coronary artery calcium, exercise tolerance, and CHD events in asymptomatic men. *Atherosclerosis.* 2006 Jan 23.

Kuk J, Church TS, Blair SN, and Ross R. Does Measurement Site for Visceral and Abdominal Subcutaneous Adipose Tissue Alter Associations with the Metabolic Syndrome? *Diabetes Care.* 2006 Mar;29(3):679-684.

Kuk JL, Katzmarzyk PT, Nichaman MZ, Church TS, Blair SN, Ross R. Visceral Fat Is an Independent Predictor of All-cause Mortality in Men. *Obes Res.* 2006 Feb;14(2):336-341

Church TS, Levine BD, McGuire DK, Lamonte MJ, Fitzgerald SJ, Cheng YJ, Kimball TE, Blair SN, Gibbons LW, Nichaman MZ Coronary artery calcium score, risk factors, and incident coronary heart disease events. *Atherosclerosis.* 2006 Mar 13.

Church TS, Kuk JL, Ross R, Priest EL, Biltoff E & Blair SN. Association of Cardiorespiratory Fitness, Body Mass Index and Waist Circumference to Nonalcoholic Fatty Liver Disease. *Gastroenterology.* 2006 Jun;130(7):2023-30.

Trivedi MH, Greer TL, Grannemann BD, Church TS, Galper DI, Sunderajan P, Wisniewski SR, Chambliss HO, Jordan AN, Finley C, Carmody TI. TREAD: Treatment with Exercise Augmentation for Depression: study rationale and design. *Clin Trials.* 2006;3(3):291-305

The LIFE Study Investigators. Effects of Physical Activity Intervention on Measures of Physical Performance: Results of the Lifestyle Interventions and Independence for Elders Pilot (LIFE-P) Study. *Journal of Gerontology.* 2006, Vol.61A, No. 11,1157-1165

#### Honors, Awards and Peer Review Assignments.

Tulane University Graduate School Tuition Scholarship 1996-1997

Tulane University School of Public Health, Dean's Grant 1998-1999

2nd Place, Scientific Poster Competition, Delta-Omega Honor Society, Eta Chapter. 1999

Travel Award for 10th International Congress of Biorheology and 3rd International Conference on Clinical Hemorheology, Pecs, Hungary. 1999

Invited Fellow, Physical Activity and Public Health 2000: The Postgraduate Course on Research Directions and Strategies. Sponsored by Centers for Disease Control and Prevention and University of South Carolina, Prevention Research Center. 2000

2nd Place, Martti J. Karvonen Young Investigator Award, International 16<sup>th</sup> Puijo Symposium, Koupio, Finland. 2001

Jeremiah and Rose Stamler Research Award for New Investigators, 42<sup>nd</sup> Annual American Heart Association Conference on Cardiovascular Epidemiology and Prevention, Honolulu, Hawaii, April 2002

Member of Physical Activity Committee of the Council on Nutrition, Physical Activity and Metabolism. American Heart Association. 2005 to present

National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK) grant reviewer. Special Emphasis Panel (ZDK1-GRB8-M1) evaluating applications (RO3s) submitted in response to the Program Announcement PAR-01-056 entitled "Small Grants in Digestive Diseases and Nutrition". April 2004, July 2004, Dec 2005

National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK) grant reviewer. Special Emphasis Panel evaluating applications (RO1s) submitted in response to the RFA DK-03-022 entitled "Ancillary Studies to Obesity-Related Clinical Trials". March 2005

National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK) grant reviewer. National Institutes of Health Clinical/Pediatric Research Loan Repayment Program. March 2006

Peer reviewer for Journal of American Medical Association, Circulation, Medicine & Science in Sports and Exercise, American Journal of Preventive Medicine, Journal of Applied Physiology, Obesity Research, American Journal of Cardiology, Preventive Medicine

#### Miscellaneous Grants, Presentations, and Publications

"What We know about Preventing Obesity": A presentation to the Louisiana Department of Health and Hospitals Ad Hoc Committee on Obesity, which was created to develop a scientifically based report on obesity for the Louisiana Legislature. 1999.

Co-Author: Louisiana CVD Grant to build capacity. A successful \$300,000 CDC grant to build capacity to fight cardiovascular disease in Louisiana. 1999.

AUTHOR OF *STATE OF THE HEART REPORT, CVD IN LOUISIANA*: AN AMERICAN HEART ASSOCIATION FUNDED REPORT DESCRIBING THE CVD BURDEN AND EPIDEMIOLOGY IN LOUISIANA. 12,000 COPIES WERE DISTRIBUTED ACROSS LOUISIANA AND A WEB SITE CREATED. 2000.

Invited Speaker, Defense Advanced Research Projects Agency (DARPA). Role of cytokines in extreme human performance.

Invited Speaker, **Congress of the United States**-Committee on Government Reform: Diet, Physical Activity, and Dietary Supplements- the Scientific Basis For Improving Health, Saving Money and Preserving Personal Choice. July 25, 2002

Activities

Two-time finisher of Ironman distance triathlons

## CURRICULUM VITAE

**Name:** Jean-Marc Schwarz

|                 |                                                                                                                         |                                                                                                                                                               |
|-----------------|-------------------------------------------------------------------------------------------------------------------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <b>Address:</b> | Touro University, California<br>1310 Johnson Lane<br>Mare Island<br>School of Osteopathic Medicine<br>Vallejo, CA 94592 | University of California<br>San Francisco General Hospital<br>School of Medicine Endocrinology<br>1001 Potrero Ave Bldg. 30 R 3501<br>San Francisco, CA 94110 |
|-----------------|-------------------------------------------------------------------------------------------------------------------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------|

|                   |                |                |
|-------------------|----------------|----------------|
| <b>Telephone:</b> | (707) 638-5268 | (415) 206-5533 |
|-------------------|----------------|----------------|

### EDUCATION:

|      |                             |                                     |
|------|-----------------------------|-------------------------------------|
| 1985 | BS, Biochemistry/Physiology | University of Lausanne, Switzerland |
| 1990 | PhD, Physiology/Nutrition   | University of Lausanne, Switzerland |

### RESEARCH EXPERIENCE:

|               |                                 |                                                                         |
|---------------|---------------------------------|-------------------------------------------------------------------------|
| 1985-1989     | Research Assistant              | University of Lausanne, Switzerland                                     |
| 1990-1991     | Post-doctoral Fellow            | University of Texas Medical School, Galveston                           |
| 1992-1994     | Post-doctoral Fellow            | University of California, Berkeley                                      |
| 1995-2003     | Assistant Professor             | University of California, Berkeley                                      |
| 1999-2003     | Assistant Adjunct Professor     | University of California, San Francisco                                 |
| 2004- present | Associate Professor             | Touro University, School of Osteopathic<br>Medicine Vallejo, California |
| 2004-present  | Assoc. Research Endocrinologist | University of California, San Francisco                                 |

### HONORS AND AWARDS:

|           |                                                                     |                                     |
|-----------|---------------------------------------------------------------------|-------------------------------------|
| 1985-1989 | Scholarship Xyrofin Ltd.                                            | University of Lausanne, Switzerland |
| 1989      | International Foundation for the<br>Promotion of Nutrition Research | University of Lausanne, Switzerland |
| 1992      | Fonds National Suisse de la<br>Recherche Scientifique               | University of California, Berkeley  |
| 1993      | Fondation Suisse de Bourses<br>en Médecine et Biologie              | University of California, Berkeley  |
| 1995      | Mead Johnson Research Fund                                          | University of California, Berkeley  |

### PUBLICATIONS

#### Papers:

1. **Schwarz J-M**; Y Schutz; F Froidevaux; KJ Acheson ; N Jeanpretre; H Schneider; JP Felber; and E Jequier: Thermogenesis in men and women induced by fructose vs glucose added to a meal. *Am J of Clin Nutr* 49(4):667-74, 1989.
2. **Schwarz J-M**, Y Schutz, V Piolino, H Schneider, JP Felber and E Jéquier: Thermogenesis in obese women: effect of fructose vs glucose added to a meal. *Am J Physiol* 262:E394-E401, 1992.
3. **Schwarz J-M**, KJ Acheson, L Tappy, V Piolino, MJ Muller, JP Felber and E Jéquier: Thermogenesis mechanisms and fructose metabolism in humans. *Am J Physiol* 262:E591-E598, 1992.
4. Hellerstein MK, RA Neese, and **J-M Schwarz**: Model for measuring absolute rates of hepatic de novo lipogenesis and re-esterification of free fatty acids. *Am J Physiol* 265:E814-E820, 1993.
5. Romijn, JA, D Chinkes, **J-M Schwarz**, and RR Wolfe: Lactate-pyruvate interconversion in blood: implications for in vivo tracer studies. *Am J Physiol* 266:E334-E340, 1994.
6. Hellerstein, MK, NL Benowitz, RN Neese, **J-M Schwarz**, R Hoh, P Jacob, J Hsieh and D Faix: Effects of cigarette smoking and its cessation on lipid metabolism and energy expenditure in heavy smokers. *J Clin Invest* 93:265-272, 1994.
7. **Schwarz J-M**, RA Neese, D Dare, SM Turner, and MK Hellerstein: Short-term alterations in carbohydrate energy intake in humans. *J Clin Invest* 96:2735-2743, 1995.
8. Neese RA, **J-M Schwarz**, D Faix, S Turner, A Letscher, D Vu and MK Hellerstein: Gluconeogenesis and intrahepatic triose phosphate flux in response to fasting or substrate loads. *J Biol Chem* 270(24):14452, 1995.
9. Hellerstein MK, A Letcher, **J-M Schwarz**, D Cesar, CHL Schackleton, S Turner, RA Neese, K Wu, S Block, and S Kaempfer: Measurement of rate of appearance of hepatic UDP-glucose in vivo in rats: relation to glycogen deposition and labeling patterns. *Am J Physiol* 272:E155-E162, 1997.
10. Hellerstein MK, RA Neese, **J-M Schwarz**, S Turner, D Faix , and K Wu: Altered fluxes responsible for reduced hepatic glucose production and gluconeogenesis by exogenous glucose in rats. *Am J Physiol* 272:E163-E172, 1997.
11. Gastaldelli, A, **J-M Schwarz**, E Caveggion, , L Traber, D Traber, J Rosenblatt, G Toffolo, C Cobelli and R Wolfe: Modeling in physiology: Glucose kinetics in interstitial fluid can be predicted by compartmental modeling. *Am J Physiol* 272:E494-E505, 1997.
12. Tappy L, **J-M Schwarz**, P Schneiter, C Cayeux, J-P Revely, CF Fagerquist, E Jequier, and R Chiolero: Effects of isoenergetic glucose-based or lipid-based parenteral nutrition on glucose metabolism, de novo lipogenesis, and respiratory gas exchanges in critically ill patient. *Crit Care Med* 26 (5):860-867, 1998.
13. Fagerquist CK and **J-M Schwarz**: Gas-phase acid/base chemistry and it's effects on mass isotopomer abundance measurements of biomolecular ions. *J Mass Spectrom* 33:144-153, 1998.
14. Tappy L, MM Berger, **J-M Schwarz**, M McCamish, J-P Revely, P Schneiter, E. Jequier and R Chiolero: Hepatic and peripheral glucose metabolism in intensive care patients receiving continuous high or low carbohydrate enteral nutrition. *Jpen-Parenter Enter* 23: 260-268, 1999.
15. **Schwarz J-M**, R Chiolero, J-P Revely, C Cayeux, P Schneiter, E Jequier, T Chen, and L Tappy: Effects of enteral carbohydrates on de novo lipogenesis in critically ill patients. *Am J of Clin Nutr*. 72(4):940-5, 2000



16. Loe YC, N Bergeron, N Rodriguez, and **J-M Schwarz**. A gas chromatography mass spectrometry method to quantify blood hydroxycitrate concentration. *Anal Biochem* 292 (1): 148-154 2001.
17. Noor M, JC Lo, K Mulligan, **J-M Schwarz**, R Halvorsen, M Schambelan, and C Grunfeld. Metabolic effects of indinavir in healthy HIV seronegative men. *Aids* 15 (7): F11-F18, 4 2001
18. Battilana P, K Ornstein, K Minehira, **J-M Schwarz**, K Acheson, P Schneiter, J Burri, E Jequier, and L Tappy. Mechanisms of action of  $\beta$ -glucan in postprandial glucose metabolism in healthy men. *Eur J Clin Nutr* 55 (5): 327-333, 2001.
19. Lo JC, K Mulligan, M Noor, **J-M Schwarz**, RA Halvorsen, C Grunfeld, and M Schambelan. The effects of recombinant human growth hormone on body composition and glucose metabolism in HIV-infected patients with fat accumulation. *J Clin Endocrinol Metab* 86:3480-87, 2001.
20. Minehira K, Novel-Chaté V, **Schwarz J-M**, Gillet M, Darioli R, Chioléro R, Tappy L. Hepatic de novo lipogenesis after liver transplantation. *J Parenter Enter Nutr* 25:229-235, 2001.
21. **Schwarz J-M**, Mulligan K, Lee J, Lo JC, Noor M, Wen M, Grunfeld C, Schambelan M. The effects of recombinant human growth hormone on hepatic lipid and carbohydrate metabolism in HIV-infected patients with fat accumulation. *J Clin Endocrinol Metab* 87(2): 942-945, 2002.
22. Trimmer JK, **J-M Schwarz**, GA Casazza, MA Horning, N Rodriguez, and GA Brooks. Measurement of gluconeogenesis in resting and exercising men by mass isotopomer distribution analysis (MIDA) *J Appl Physiol* 93: 233-241, 2002.
23. Noor MA, Seneviratne T, Aweeka F, Lo JC, **Schwarz J-M**, Mulligan K, Schambelan M, Grunfeld C. The HIV protease inhibitor indinavir acutely inhibits insulin-stimulated glucose disposal: A randomized, placebo-controlled study. *AIDS* 16(5):F1-F8, 2002.
24. Minehira K, L. Tappy, R Chioléro V Vladomira, MM Berger, J-P Revelly, and **J-M Schwarz**. Fractional hepatic de novo lipogenesis in healthy subjects during near-continuous oral nutrition and bed rest: a comparison with published data in artificially fed, critically ill patients. *Clin Nutr* 21 (4): 345-350, 2002.
25. **Schwarz J-M**, PA Linfoot, D Dare, and K Aghajanian. Hepatic de novo lipogenesis in normo and hyperinsulinemic subjects consuming high-fat/low-carbohydrate and low-fat/high-carbohydrate isoenergetic diets *Am J of Clin Nutr* 77 (1): 43-50, 2003.
26. Lee G, Seneviratne T, Noor MA, Lo JC, **Schwarz J-M**, Aweeka FT, Mulligan K, Schambelan M, Grunfeld C. The metabolic effects of lopinavir/ritonavir in HIV-negative men. *AIDS* 18:641-649, 2004.
27. Lee GA, Mafong DD, Noor MA, Lo JC, Mulligan K, **Schwarz J-M**, Schambelan M, Grunfeld C. HIV Protease inhibitors increase adiponectin levels in HIV-negative men. *J Acquire Immune Defic Syndr* 36:645-647, 2004.
28. Lo JC, Mulligan K, Noor MA, Lee GA, **Schwarz J-M**, Grunfeld C, Schambelan M. The effects of low dose growth hormone in HIV-infected men with fat accumulation: a pilot study. *Clin Infect Dis*. 39(5):732-5, 2004.
29. **Schwarz J-M**, Lee GA, Park S, Noor MN, Lee J, Wen M, Lo JC, Mulligan K, Schambelan M, Grunfeld C. Indinavir increases glucose production in healthy HIV-negative men. *AIDS* 18(13):1852-4, 2004.
30. Faeh D, Minehira K, **Schwarz J-M**, Periasamy R, Park S, Tappy L. Effect of fructose overfeeding and fish oil administration on hepatic de novo lipogenesis and insulin sensitivity in healthy males. *Diabetes* 54:1907-1913, 2005.

### Reviews and Book Chapters:

Hellerstein MK, **J-M Schwarz**, and RN Neese: Regulation of hepatic de novo lipogenesis in humans. *Annual Review of Nutrition* 16:523-57, 1996.

Tappy L and **J-M Schwarz**. Metabolic Disorders. *Clinical Nutrition*, 5:76-84, 2005. The Nutrition Society Textbook Series. Published by Blackwell Science.

### Selected Abstracts (since 1995):

1. Schwarz J-M, RA Neese, D Dare, SM Turner, MK Hellerstein. Effect of carbohydrate overfeeding and underfeeding on de novo lipogenesis and insulin levels. *Proceedings of the Nutrition Society*. 54 (1): 47A, 1995.
2. Hibbert J, J-M Schwarz, DC Gore, RA Neese, MK Hellerstein. De novo lipogenesis and energy expenditure before and after food restriction in morbid obesity. *Proceedings of the Nutrition Society*. 54 (1): 48A, 1995.
3. Schwarz J-M, A Letscher, P Linfoot, D Cesar, S Bock, RA Neese. A tracer method for simultaneous measurement of gluconeogenesis and glucose production using stable isotopes. *Diabetes* (Suppl. 2) 45: 321A, 1198, 1996.
4. Letscher A, S.M. Turner, D Faix, J-M Schwarz, MK Hellerstein. Metabolic mechanisms by which IV glucose reduces hepatic glucose production and gluconeogenesis in rats. *Diabetes* (Suppl. 2) 45: 162A, 593, 1996.
5. Linfoot P, RA Neese, J-M Schwarz, S Turner, H Sauerwein, E Dekker, MK Hellerstein. Absolute gluconeogenesis in humans by mass isotopomer distribution analysis: isotopic design and analytic requirements. *Diabetes* (Suppl. 2) 45: 326A, 1218, 1996.
6. Aghajanian K, CK Fagerquist, J-M Schwarz. Effect of fructose on de novo lipogenesis and hepatic glycogen in rats. *Faseb J* 11 (3):A381, 2208, 1997.
7. Neese R, P Linfoot, S Turner, J-M Schwarz, M Christiansen, MK Hellerstein. Hepatic gluconeogenesis fluxes and glycogen turnover during fasting in human. *Diabetes* (Suppl. 1) 46: 243A, 930, 1997.
8. Schwarz J-M, R Linfoot, RA Neese, D Dare, MK Hellerstein. Effect of diet on leptin: a signal of surplus dietary fat in lean and obese men. *Diabetes* (Suppl. 1) 46: 245A, 942, 1997.
9. Quilici A, J Reilly, J King, B Braun, J-M Schwarz. Effects of glucose infusion on glucose production and gluconeogenesis: a new method to assess hepatic insulin resistance. *Diabetes* (Suppl. 1) 46: 246A, 943, 1997.
10. Trimmer JK, BC Bergman, AL Friedlander, GA Casazza, MA Horning, J-M Schwarz and GA Brooks: Mass Isotopomer Distribution Analysis of gluconeogenesis during hard exercise using [2-<sup>13</sup>C]glycerol and [3-<sup>13</sup>C]alanine. *FASEB J*, 12(5):A737 (#4276) 1998.
11. Mills ML, J Chen, RA Neese, MK. Hellerstein, J-M Schwarz. Mass isotopomer distribution analysis for conditions of hyperglycemia in streptozotocin diabetic rats. *Diabetes* (Suppl 1) 47:287A: 1112, 1998.
12. Fagerquist CK, J-M Schwarz, MK Hellerstein, SC Davis, AA Makarov, JD Hughes. A supersonic molecular beam source for mass isotopomer abundance measurements of biomolecular ions. *Am Soc Mass Spectrom* : 41, 1998.
13. Schwarz P, R Linfoot, L Liu: Effect of fructose infusion on gluconeogenesis, glucose production and hepatic lipogenesis in obese hyperinsulinemic subjects. *Diabetes* (Suppl 1) 48: A299: 1307, 1999.
14. Chen TW, LR Lee, V NG, J Chen, JM Schwarz. Relationship between hepatic lipogenesis and malonyl-CoA concentration in a rat model. *Diabetes* (Suppl 1), 48: A452: 2006, 1999.

15. Schwarz JM, TW Chen, R Linfoot. Effect of hydroxycitrate on hepatic de novo lipogenesis, gluconeogenesis and glucose production in obese hyperinsulinemic subjects. *Circulation* Suppl 1, 100; 18: I-196, A1015, 1999.
16. Loe YCN. Rodriguez, J.M. Schwarz. New mass spectrometric method to quantify blood hydroxycitrate concentration in human subjects. *FASEB J.* 14 (4), A158.1, 2000.
17. Noor M, Lo J, K Mulligan, JM Schwarz, R Halvorsen, M Schambelan, C Grunfeld. Metabolic effects of indinavir in healthy HIV seronegative subjects. 2nd International Workshop on Adverse Drug Reactions and Lipodystrophy in HIV, Toronto, 2000.
18. Lo JC, K Mulligan, M Noor, JM Schwarz, C Grunfeld, M Schambelan. The effects of recombinant human growth hormone (GH) on glucose metabolism and body composition in HIV-positive subjects with fat accumulation (FA) syndromes. 2nd International Workshop on Adverse Drug Reactions and Lipodystrophy in HIV, Toronto, 2000
19. Schwarz JM, Linfoot P, Rodriguez N, Aghajanian K. Effect of insulin resistance and dietary carbohydrate content in hepatic de novo lipogenesis in men. *Circulation* Suppl 2, 102; 18: II-702, A3394, 2000.
20. Hsieh E, J Lee, A Young, N Bergeron, M Wen, JM Schwarz. GC/MS determination of lipoprotein lipid composition and concentration. *FASEB J* 15 (4): A292-A292 Part 1 2001.
21. Loe YC, N Bergeron, J Phan, M Wen, J Lee, JM Schwarz. Time course of hydroxycitrate clearance in fasting and fed *FASEB J* 15 (4): A632-A632, Part 1 2001.
22. JM Schwarz, K Mulligan, J Lee<sup>1</sup>, JC Lo, M Wen<sup>1</sup>, MA Noor, C Grunfeld, M Schambelan. Effects of growth hormone on hepatic lipid and carbohydrate metabolism in HIV-infected patients with fat accumulation (3rd International Workshop on Adverse Drug Reactions and Lipodystrophy in HIV, 2001)
22. Noor MA, S Park, GA Lee, J-M Schwarz, J Lee, M Wen, JC Lo, K Mulligan, M Schambelan, C Grunfeld. Indinavir increases hepatic glucose production in HIV-negative men. *Antiviral Therapy* 8:L9, 2003.
23. Lo JC, Mulligan K, Havel PJ, Noor MN, Lee GA, Schwarz JM, Grunfeld C, Schambelan M. The effect of recombinant human growth hormone treatment on circulating leptin and adiponectin concentrations in patients with HIV-associated fat accumulation. *Antiviral Therapy* 9:L4, 2004.
24. Lee GA, Mafong DD, Noor MA, Lo JC, Mulligan K, Schwarz J-M, Schambelan M, Grunfeld C. HIV protease inhibitors increase adiponectin levels in HIV-negative men. *11<sup>th</sup> Conference on Retroviruses and Opportunistic Infections*, San Francisco, p. 325, 2004.
25. Lee GA, Mafong DD, Lo JC, Schwarz JM, Aweeka FT, Mulligan K, Schambelan M, Grunfeld C. Ritonavir acutely induces insulin resistance in healthy normal volunteers. *Antiviral Therapy* 9:L6, 2004.
26. Lo JC, Mulligan K, Havel PJ, Noor MN, Lee GA, Schwarz JM, Grunfeld C, Schambelan M. The effect of recombinant human growth hormone treatment on circulating leptin and adiponectin concentrations in patients with HIV-associated fat accumulation. *Antiviral Therapy* 9:L4, 2004.
27. Schwarz JM, Wen MJ, Park S, Periasamy R, Bergeron N, Dare D, Tai V, Linfoot P. Effect of fish oil vs olive oil supplement on de novo lipogenesis and glucose production in normoinsulinemic lean and hyperinsulinemic obese subjects. *Diabetes* (Suppl. 1) 54: A10, 38, 2005.

## Current Support

R01 DK63640 NIH/NIDDK (PI: Schambelan): Leptin Treatment of HIV-Associated Lipodystrophy  
 9/02-7/06.  
 Role: Co-Investigator

R21 DK69185 NIH/NIDDK (PI: Schambelan): Can IGF-I/IGFBP-3 Reverse Central Fat Accumulation? 7/04-6/06.  
Role: Co-Investigator

R01 DK66999 NIH/NIDDK (PI: Grunfeld): HIV Antiretroviral Drugs and Glucose Metabolism 3/04-12/07  
Role: Co-Investigator

### **Pending Grants**

American Heart Association (Grant in Aid) Application submitted 1-5-06  
Effect of high fructose diet on liver fat production and accumulation in lean healthy subjects  
Role: Principal Investigator  
American Diabetes Association (Clinical grant) Application submitted 1-15-06  
Effect of low-fructose and low-lipogenic diet on liver fat content and production in overweight hyperinsulinemic subjects  
Role: Principal Investigator

R21 AT003374-01A NIH/NCCAM (PI: Schambelan) Application pending 4/1/06-3/31/09  
Uridine Supplementation, Mitochondrial Function, and Glucose Metabolism in HIV  
Role: Co-Investigator

### **Previous Grants**

NIH/CRcFF GCRC: Effects of High Fructose Diet on Liver Fat Production and Accumulation 9/04-12/05  
Role: Principal Investigator

R01 DK54615 NIH/NIDDK (PI: Mulligan): Metabolic Effects of Protease Inhibitors in HIV Disease 9/98-08/05  
Role: Co-Investigator

American Diabetes Association (011770): Effect of Fish Oil on Hepatic Lipid and Carbohydrate Metabolism in Hyperinsulinemic Obese Subjects 1/00-12/03  
Role: Principal Investigator

USDA NRI (99 35200 8605): Effect of Fish Oil on Hepatic Lipid and CHO Metabolism in Healthy Human Subjects. 12/01-11/02  
Role: Principal Investigator

R01 DK45833 NIH/NIDDK(PI: Schambelan): Anabolic Therapies & Their Metabolic Effects in AIDS 12/97-05/04.  
Role: Co-Investigator

American Heart Association: Inhibition of de novo lipogenesis: Effect on glucose and VLDL output 1/98-12/01

Role: Principal Investigator

ILSI: Modulation of Hepatic de novo Lipogenesis: Effect on Glucose and VLDL Output 7/98-6/00

Role: Role: Principal Investigator

Hewlett Packard Award #010625: Measurement of isotope ratio of polymers using HP's 8/98-7/99.

Role: Principal Investigator