**1. Title Page**

Insights into space medicine from polar bears and
insights into global climate change from the Arctic

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## 2. Abstract

Polar bears spend most of their time on Arctic sea ice, where they hunt seals. In the summer, sea ice retreats north and most polar bears follow it, but some remain on shore. In response to warm temperatures and lack of seals, these bears likely fast and reduce their activity to conserve energy. In most mammals, reduced activity and lowered food intake lead to atrophy of skeletal muscle (e.g. loss of muscle size and strength). However, polar bears may enter a state of "walking hibernation" which includes mild hypothermia and prevention of muscle atrophy while the animal remains functional and cognizant. Humans in space also experience muscle atrophy, due to reduced mobility and the low-gravity environment, and this can jeopardize NASA missions; thus, the Exploration Systems Mission Directorate seeks to understand the process of atrophy. To investigate walking hibernation in polar bears as a model of the avoidance of muscle atrophy, I will capture bears on shore and on retreating sea ice in early and late summer. During the first capture I will collect a muscle sample, implant a body temperature logger, and attach a radio collar with an activity monitor. During the second capture I will remove the logger and collar and repeat the muscle sampling. I will compare molecular indicators of muscle atrophy and quantify patterns of activity and body temperature between the two captures. Additionally, climate change is reducing Arctic sea ice, likely forcing more bears to spend summer on shore and reducing food availability for bears that follow the sea ice north. By investigating these implications of climate change, this project also contributes to the Earth Science project of the NASA Science Mission Directorate.

## 3. Project Proposal

Description of Proposed Research

Polar bears (*Ursus maritimus*) are strongly dependent upon Arctic sea ice for traveling and hunting seals (Amstrup 2003). Of nearly 15,000 polar bear locations gathered by satellite telemetry in northwestern North America from 1985-2001, all but 7% were on sea ice (Amstrup 2003). In the summer, sea ice retreats north and many bears follow it; however, in some areas bears remain on shore during summer (Regehr et al. 2007; Schliebe et al. 2008). Recent loss of Arctic sea ice due to warmer temperatures has been particularly profound in summer months (Meier et al. 2007), forcing more bears to spend this period on shore.

Polar bears on shore likely find little to eat and face warm temperatures. To avoid starvation and hyperthermia, it is thought that polar bears can enter a state of "walking hibernation" in which they actually reduce their body temperature to a mild hypothermia, reduce their activity, and conserve protein while catabolizing body fat (Nelson et al. 1980). In most animals, long periods of reduced activity and food deprivation cause loss of muscle size and strength (Dudley et al. 1989) and a decrease in slow-twitch endurance fibers in muscles (Baldwin and Haddad 2001). Thus, polar bears in walking hibernation would need physiological adaptations to allow them to maintain integrity of skeletal muscles despite reduced activity. Such adaptations have been described in black bears (*U. americanus*) and brown bears (*U. arctos*) hibernating in winter dens (Harlow et al. 2001; Hershey et al. 2008). In contrast to these species, polar bears in walking hibernation are thought to retain some activity and to remain cognizant and functional (Nelson et al. 1980). Humans in space often experience muscle atrophy which can interfere with their mission (ECP 2009); thus NASA will benefit to understand if and how polar bears enter walking hibernation and avoid atrophy while retaining some activity and function.

To evaluate walking hibernation in polar bears, I will work in collaboration with ongoing projects by US Fish and Wildlife Service (USFWS) and US Geological Survey (USGS) to capture and sample polar bears on shore in northern Alaska at the beginning and the end of summer. Bears will be darted with immobilizing drugs from a helicopter, and using sterile field techniques I will implant a body temperature logger and remove a small tissue biopsy from the biceps femoris, a hindlimb muscle that supports standing (Hershey et al. 2008). Standard measurements such as bear weight, length, and assessment of body fat deposits will be made and each bear will be fitted with a collar carrying a GPS unit, a radio unit, and an activity monitor. Collars will record the location and activity of the bear, and will be removed upon recapture. Necessary approval and permits have been granted by the University of Wyoming Animal Care and Use Committee (approval #A-3216-01) and the USFWS (the current USGS permit for polar bear captures has been amended to include this project). Following the same protocol, I will also sample bears that follow the sea ice north during the summer. I expect that these bears will experience cooler temperatures and have more access to food because seals will be present on the sea ice, allowing the bears to remain fully active and avoid the need for walking hibernation. However, it is possible that because climate change is causing sea ice to retreat further north during the summer, the bears will be north of the productive waters of the continental shelf and will be in areas where seals are scarce. In that situation, I expect bears on the sea ice will conserve protein and catabolize body fat, similar to bears on shore.

I will compare daily patterns and extent of activity, and changes in body temperature, between bears on shore and bears on sea ice. I will use multiple assays to evaluate strength in muscle samples collected from bears. Assays will include: protein content, which decreases with muscle atrophy; DNA:RNA ratio, which is an indicator of protein synthesis; and expression of RNA and of different types of proteins, as indicators of loss of slow-twitch endurance fibers. In addition, portions of muscle samples will be sectioned, stained, and examined with a microscope for the estimation of muscle fiber size, which decreases with atrophy. Finally, portions of the muscle biopsy will be submitted to the University of Wyoming Stable Isotope Facility for quantification of $^{15}N$, which is known to increase in food-deprived mammals that are not adapted to conserve protein. Carbon and nitrogen content will also be quantified as an index of maintained muscle performance.

Relationship to goals of the National Aeronautics and Space Administration (NASA)

A sustained human presence on the moon and the eventual use of human crews for interplanetary spaceflight are explicit goals of the Exploration Systems Mission Directorate of NASA (ESMD 2009). However, even short periods of reduced activity and exposure to low-gravity environments in space cause muscle atrophy in humans (Baldwin 1996), which can endanger the health of crew members and jeopardize NASA missions (ECP 2009). Within the Exploration Systems Mission Directorate, the Human Health Countermeasures Project (HHC 2009) seeks to understand the process of muscle atrophy and to find means of reducing its occurrence and severity. However, the study of atrophy in humans during spaceflight is very difficult due to logistics; instead, ground-based studies that do not involve low-gravity environments provide crucial models of atrophy. For example, humans or animals in states of reduced activity or muscle unloading (e.g. limb immobilization due to casting) experience atrophy similar to that caused by spaceflight, and study of these models provides key insights into the atrophy process (Adams et al. 2003).

Hibernating bears provide an animal model in which the effects of atrophy are reduced or avoided. Black bears that were inactive during winter hibernation for about 4 months lost about 23% of their strength in the tibialis anterior, a hindlimb muscle; this stands in contrast to a predicted strength loss of 90% in humans experiencing the same conditions (Figure 1; Harlow et al. 2001). Polar bears that reduce their activity during summer months and enter walking hibernation can make an additional unique contribution to space medicine and NASA's goals, by providing a model in which muscle atrophy is avoided, while the bears retain some activity and a functional capacity in their environment. This model is particularly relevant to the goal of avoiding atrophy in human crews in space, who likewise must remain functional during missions.

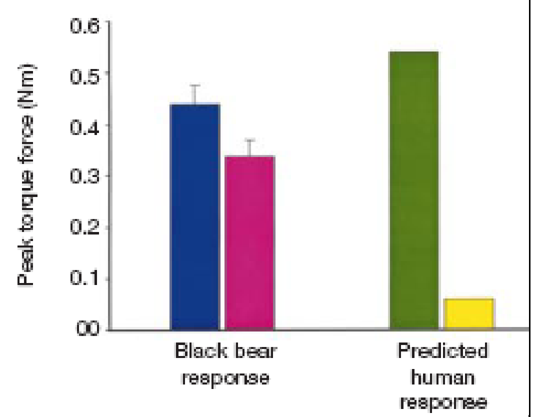


Figure 1. Muscle strength in the tibialis anterior muscle of hibernating black bears. Bears were tested in early hibernation (blue bar, left) and late hibernation (pink bar, right), representing about 4 months of inactivity between tests. At the right is the predicted human response to the same period of inactivity, showing a more dramatic loss of strength. From Harlow et al. 2001.

This project will also contribute to the Science Mission Directorate by investigating the effects of climate change and the loss of Arctic sea ice on animal physiology. Warming temperatures in the Arctic are contributing to a rapid loss of sea ice, which varies in extent by location and time of year (Meier et al. 2007). Within the Science Mission Directorate, the Earth Science project seeks to understand the consequences of changes in the Earth System (NSMD), and the Climate Variability and Change project (CVC 2009) focuses on understanding and modeling the loss of Arctic sea ice, enabling predictions of future ice conditions. Because polar bears rely heavily on sea ice (Amstrup 2003), they can serve as useful indicators of how changing conditions may affect Arctic organisms. Climate change has already been implicated in a decline in a Hudson Bay population of polar bears (Regehr et al. 2007).

This project also aligns with the missions of the National Space Grant College and Fellowship Program. The study of muscle atrophy in polar bears will provide insight into atrophy in humans, a transfer of practical knowledge that will contribute towards the aerospace goal of long-term human spaceflight (NSGCFP, mission statement #2). This study will also integrate human biomedical research and wildlife ecological research, achieving the mission of interdisciplinary programs (NSGCFP, mission statement #5).

Finally, performing field work in northern Alaska provides a unique opportunity to interact with remote communities and disseminate and discuss scientific news regarding the Arctic environment. Towards that goal, a website has been built (www.uwyo.edu/polarbear/) that provides general information on polar bears, a description of this project, and data collection updates; this website will be advertised in northern Alaskan communities. I am also collaborating with US Fish and Wildlife Service and US Geological Survey to participate in community meetings and share scientific developments regarding the Arctic environment and to learn the viewpoint of Arctic residents. Lastly, through the Arctic Research Consortium of the US (www.arcus.org) my advisors and I are paired with a high school teacher from East Palo Alto Academy, California. She will participate in field work in fall 2009 and we will work with her to create lesson plans and presentations focused on our research, which she will bring back to her school and community. These outreach activities achieve the mission of contributing to public scientific literacy (NSGCFP, mission statement #6).

## Products

I will present the findings of this research at appropriate national or international conferences. Possibilities include the 51st annual meeting of the Society for Integrative and Comparative Biology in January 2011, or the 20th International Conference on Bear Research and Management in 2011. Results will also be submitted for publication in a peer-reviewed journal; possibilities include Journal of Applied Physiology, and Physiological and Biochemical Zoology. Data from this project will be shared with collaborators USFWS and USGS.

## Timeline

Summer and Fall 2008: Necessary approvals and permits were granted and a pilot capture season occurred in August. Twenty-nine polar bears, including cubs, were captured on the northern shore of Alaska, and muscle samples were taken from 13 adults who were also fitted with collars. A pilot recapture season occurred in October. Of the 13 adults fitted with collars, 7 were recaptured for muscle sampling and collar retrieval. Based on preliminary data analyses, at least

1 adult substantially reduced activity and did not travel between captures, indicating potential muscle atrophy.

Spring 2009: Muscle samples from pilot work are stored in the University of Wyoming Macromolecular Core Facility and analyses have begun, starting with protein content assays. Community outreach meetings may occur in spring or fall 2009. Collaboration with high school teacher has begun.

Summer 2009: Current graduate student funding from the University of Wyoming Program in Ecology ceases in July; NASA Graduate Fellowship period of support begins in August (Fellowship plus 1 month of requested summer stipend). Capture season begins in May, as the sea ice begins retreating north for the summer. This will include capture of bears that will spend the summer on shore and capture of bears that will spend the summer on ice. An additional capture season is planned for August, to increase sampling of bears on shore. Assays of muscle samples continue.

Fall 2009: Recapture season begins in October, as temperatures decrease and the sea ice begins to advance south. Bears that have spent the summer on shore will be recaptured in a shore-based operation similar to 2008; bears on the sea ice will be recaptured from the US Coast Guard Icebreaker “Polar Sea” stationed near the sea ice. After recaptures, samples will be returned to the University of Wyoming and assays of muscle samples will continue. Activity and temperature data will be downloaded and analyzed.

Spring 2010: Assays of muscle samples will be completed and results organized into manuscripts for inclusion in dissertation, presentations, and peer-reviewed publications.

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## 4. Award Request / Statement of Cost Share / Current and Pending Funding

Award Request

| | |
|---|---|
| PhD graduate fellowship | $18,000 |
| 1 month of summer stipend support | $2,000 |
| Total | ($20,000) |

Statement of Cost Share

Not included below are personnel and equipment from US Fish and Wildlife Service (USFWS) and US Geological Survey (USGS) that will participate in polar bear capture work regardless of this project.

Table 1: Equipment and logistics

| Description | Cost | Source | Federal |
|---|---|---|---|
| Helicopter, housing, travel, and other logistical support for captures | $300,000 | National Science Foundation (NSF) support for grant #0732713, USGS, USFWS | Yes |
| Polar Sea icebreaker logistical support | $1,500,000 | NSF support for grant #0732713, US Coast Guard | Yes |
| Collars and GPS/radio/activity units, temperature loggers | $120,000 | NSF grant #0732713 | Yes |
| DNA and RNA extraction kit, protein content assay kit | $300 | NSF grant #0732713 | Yes |
| Lab supplies for quantifying fiber types, and RNA and protein expression | $1,500 | NSF grant #0732713 | Yes |
| UW Stable Isotope Facility fees | $400 | NSF grant #0732713 | Yes |
| Conference and journal publication costs | $2,000 | NSF grant #0732713 | Yes |

| Table 2: Labor | | |
|---|---|---|
| Name | Source | Amount |
| Dr. Henry Harlow | advisor at UW | 40 hr/wk × 5 wk = 200 hr<br>1 hr/wk × 35 wk = 35 hr<br>total = (235 hr) |
| Dr. Merav Ben-David | co-advisor at UW | 40 hr/wk × 5 wk = 200 hr<br>0.5 hr/wk × 35 wk = 17.5 hr<br>total = (217.5 hr) |

Current and Pending Funding

National Science Foundation (NSF) grant #0732713, “Adaptive long-term fasting in land- and ice-bound polar bears: coping with ice loss in the Arctic” was awarded to principal investigators Dr. Henry Harlow and Dr. Merav Ben-David in 2008. The grant covers the equipment in Table 1 and the time commitment in Table 2 in support of the muscle atrophy project, and the costs of concomitant investigations of polar bear ecology in the summer (e.g. summer diet, population modeling). As the PhD student, it is my role to conduct projects supported by the NSF grant and to seek necessary additional funding and support.