TITLE: Polar bears: Blood analyses

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FUNDING SOURCE AND GRANT NUMBER: National Science Foundation, Office of Polar Programs Grant 0732713.

U.S. Geological Survey, Ecosystems and Climate and Land Use Change Mission Areas

DATA SET OVERVIEW:

To establish whether polar bears that follow the pack ice north of the continental shelf experience food deprivation, and to estimate their ability for prolonged adaptive fasting and skeletal muscle protein and strength retention in comparison with land-bound bears, our goal was to sample individuals at the beginning of the ice-retreat period in the summer, and shortly before annual ice is reformed. In the Beaufort Sea, the ice-retreat period extends from late-June to mid-July and new ice forms from mid to late-October.

This dataset contains values of blood chemistry (serum and plasma), hematology (whole blood), quantities of free fatty acids, quantities and profiles of amino acids, serum hormones (cortisol, ghrelin, insulin), and stable carbon and nitrogen isotopes (as well as percent in sample) of blood cells (RBC) and serum. Blood chemistry was measured with Abaxis VS2 Vetscan blood analyzer using "comprehensive chemistry" rotors (www.abaxis.com), and lipid panel discs (Whiteman et al. *in review*). Hematology values were generated with an Abaxis HM5 blood analyzer following manufacture protocols (www.abaxis.com). Serum hormones were assayed with ELISA kits (cortisol -Oxford Biomedical Research, product EA65; ghrelin - Phoenix Pharmaceuticals, product EK-031-50; insulin - Mercodia, product 10-1203-01; absorbance measured on a ThermoScientific MultiSkan spectrophotometer). Free fatty acids were quantified with colorimetric kits (Wako Diagnostics, product 999-34691). Serum amino acid profiles will be quantified via ultra-high performance liquid chromatography (UPLC) at the University of Missouri Agricultural Experiment Station Chemical Laboratories (http://www.aescl.missouri.edu/). Stable carbon and nitrogen isotope values and %C and %N of blood cells and serum were generated with an isotope ratio mass spectrometer connected to an elemental analyzer at the University of Wyoming Stable Isotope Facility (www.uwyo.edu/SIF) following methods described in Ben-David and Flaherty (2012). Stable isotope values are pending as of January 11, 2013.

During our research efforts from August 2008 to May 2010, a total of 110 polar bears were captured and sampled and 29 were recaptured on shore and on the sea ice. Spring captures occurred on the ice between Point Lonely and the US Canadian border within 160 km of shore. Summer captures occurred in the same area on shore. Fall captures occurred in the same area on shore, and on the sea ice from the Alaskan coast to 80°N and from north of Wrangell Island, Russia, to Banks Island, Canada. Ice captures were conducted from the USCG *Polar Sea*.

Project information and updates can be found at www.uwyo.edu/polarbear

INSTRUMENT DESCRIPTION:

Abaxis VS2 Vetscan is an accurate chemistry, electrolyte, immunoassay and blood gas analyzer that generates results from approximately 200µl of whole blood, serum or plasma. For instrument specifications see www.abaxis.co./veterinary/products/vs2.html

Abaxis Vetscan HM5 is an automated cell counter producing values for 22-parameter complete blood count (CBC). For instrument specification see http://www.abaxis.com/veterinary/products/hm5.html

ThermoScientific Multiskan GO UV/V is a microplate spectrophotometer with a wide wavelength selection including the UV area for both 96- and 384-well plates and various types of cuvettes. For instrument specifications see http://www.thermoscientific.com/ecomm/servlet/productsdetail?storeld=11152&categoryld=82178&productId=12706112&ca=multisk anGO

DATA COLLECTION and PROCESSING:

Adult polar bears and their dependent young were captured via helicopter darting following standard immobilization techniques (details in Durner et al. 2011). Blood samples were collected from the femoral artery of all bears kept cool. Hematology analyses were conducted on fresh whole blood. The remaining samples were centrifuged for 15 minutes within 8 hours of collection. Serum and/or plasma were siphoned and separated from blood cells. Blood chemistry analyses were conducted with 12 hours of collection. The remaining samples were stored at -40°C.

Whole blood analyzed for hematology with an Abaxis HM5 blood analyzer and serum was analyzed for blood chemistry with an Abaxis VS2 – vetscan. Each sample was analyzed in duplicate or triplicate until the Coefficient of Variation (CV) was less than 0.1. Average data per sample are reported.

Red blood cells and serum were prepared for stable carbon and isotope signatures following standard protocols (Ben-David and Flaherty 2012) and submitted to the University of Wyoming Stable Isotope Facility (for quality assurance visit www.uwyo.edu/SIF). Each sample was analyzed in duplicate or triplicate until the CV was less than 0.1. Average data per sample are reported.

Stored serum samples were thawed and used for hormonal assays. Each assay was conducted following manufacturer protocols and quality control specifications and absorbance measured with a ThermoScientific Multiskan Go spectrophotometer. Each sample was analyzed in duplicate or triplicate until the CV was less than 0.1. Average data per sample are reported.

Serum amino acids will be generated by ultra-high performance liquid chromatography (UPLC) at the University of Missouri Agricultural Experiment Station Chemical Laboratories (<u>http://www.aescl.missouri.edu/</u>). Data and protocols are not yet available.

DATA FORMAT:

Data file structure: Microsoft Office Excel (.xlsx), Comma delimited ASCII (.csv)

Data format and layout: Each variable is listed in a separate file. Headers provide variable names and units of measurements. To obtain data from multiple files select from the appropriate list.

List of parameters:

Blood chemistry: Serum ALT, Serum ALB, Serum ALP, Serum AMY, Serum CA, Serum CRE, Serum GLOB, Serum GLU, Serum PHOS, Serum K, Serum NA, Serum TBIL, Serum TP, Serum BUN, Serum CHOL, Serum HDL, Serum TRIG, Serum LDL, Serum VLDL, Serum TC/H.

Hematology: Whole blood WBC, Whole blood LYM, Whole blood MON, Whole blood NEU, Whole blood EOS, Whole blood BAS, Whole blood RBC, Whole blood HGB, Whole blood HCT, Whole blood MCV, Whole blood MCH, Whole blood MCHC, Whole blood RDWc, Whole blood PLT, Whole blood PCT, Whole blood MPV, Whole blood PDWc

Serum hormones: cortisol, insulin, ghrelin

Serum fatty acids: Phosopholipids (NEFA) content.

Serum amino acids: hydroxyproline, histidine, serine, arginine, glycine, aspartate, glutamate, threonine, alanine, proline, ornithine, cysteine, lysine, tyrosine, methionine, valine, isoleucine, leucine, phenylalanine

Stable isotopes: RBC δ¹³C, RBC δ¹⁵N, RBC %C, RBC %N, Serum δ¹³C, Serum δ¹⁵N, Serum %C, Serum %N

Description of flags: For data protected under the threatened species status code is "UTSS".

Data version 1.0 date 01/12/13

DATA REMARKS:

Data for serum amino acids and stable nitrogen isotopes for blood cells and serum have not been generated as of January 11, 2013.

To view and manipulate data use Microsoft Excel.

REFERENCES:

Ben-David, M. and E. A. Flaherty. 2012. Stable isotopes in mammalian research: a beginner's guide. Journal of Mammalogy 93: 312-328.

Durner, G. M., J P. Whiteman, H. J. Harlow, S. C. Amstrup, E. V. Regehr, and M. Ben-David. 2011. Consequences of long-distance swimming and travel over deep-water pack ice for a female polar bear during a year of extreme sea ice retreat. Polar Biology 34: 975-984.

Whiteman J. P., N. Frank, K. A. Greller, H. J. Harlow, and M. Ben-David. (*in review*). Characterization of blood lipoproteins and validation of cholesterol and triacylglycerol assays for free-ranging polar bears (*Ursus maritimus*). Journal of Veterinary Diagnostic Investigations.