

## HEALTH AND METABOLISM RESEARCH CORE (HaMR Core) Facility and Equipment Description

### Environment – Contribution to Success:

The UAF houses the only dedicated MR (magnetic resonance) imaging suite in Alaska located in the Margaret Murie Life Sciences building on the West Ridge Campus. This facility provides imaging capability for Alaska's unique Arctic animal and human subjects research currently unavailable anywhere in the U.S.

The [Molecular Imaging Facility](#) (MIF) operates the following equipment:

1. [300 MHz NMR](#) (Nuclear magnetic resonance spectroscopy)
2. [600 MHz NMR](#) Used to examine tissues and other semi-solids as well as for quantitative NMR for drug stability and to quantify and characterize molecules in liquids, tissues and other semi-solids, including powdered solids up to 600 MHz.
3. [1.5T MRI](#) (Toshiba, 1.5 T Excelart/Vantage magnetic resonance imaging) equipped with an assortment of available coils to accommodate subjects of various sizes with adjustable lighting and ventilation features.
4. [DXA](#) (Dual-energy X-ray absorptiometry) measures bone density, lean body mass, and adipose tissue.

### FACILITIES:

***The Molecular Imaging Facility*** provides access to state-of-the-art magnetic resonance (MR) techniques for in-vivo and in-vitro studies using both human and animal subjects. The facility is divided into two suites located on the northwest edge of the main Fairbanks Campus. The first suite is a 570 sq ft space, established in 2011, that houses two NMRs, located in the Paul B. Reichardt Natural Sciences Building. The second space is a 2120 sq ft imaging suite in the Margaret Murie Life Sciences Building devoted to magnetic resonance imaging (MRI) and dual-energy x-ray absorptiometry (DXA).

The NMR suite of the facility houses two NMR spectrometers (Fig. 3); one is only for solution-state at 300 MHz and the other, in addition to solutions, can be used to examine tissues and other semi-solids, as well as powdered solids all at 600 MHz. Space is also available in the NMR suite for the preparation of samples that require magic angle spinning.

The imaging suite consists of four main areas: a prep/anesthesia room, a DXA scan room, a clinical lab, and an MRI scan room. The prep/anesthesia room contains all the supplies necessary to induce and maintain animals under gas anesthesia for imaging purposes. The equipment here includes a non-magnetic gurney for large animal transport and a gas anesthesia machine that can be used in the scan room near the magnet. The Clinical Lab is available for clinical trial research that makes use of the MIF instruments.

***The Animal Instrumentation*** group includes technical support and expertise in methodologies for monitoring metabolism, via respirometry and biotelemetry, in hibernating species and laboratory rodents. The equipment used for measuring metabolism and physiology is dispersed between 1) the laboratory animal holding facilities within the Biological Research and Diagnostics Facility on the UAF West Ridge Campus, 2) a specialized bear hibernation facility located in an isolated, undisturbed spruce forest within the UAF Biological Reserve, and 3) a vivarium facility in the Integrated Sciences Building on the UAA main campus. The HaMR core supports project PIs and other Center-TRIM investigators in the collection & processing of metabolic and physiological data using animal instrumentation.

### **UAF Vivarium:**

#### **Small Animal**

UAF's animal facility, administered by the Animal Resource Center, is housed in the Biological Investigation Research and Diagnosis Building (BiRD), a 42,000 sq ft animal facility, designed for animal care and holding, with adjacent procedure as well as facilities for necropsy, histology and surgical and veterinary diagnostic

procedures. The BiRD Facility, connected to the Margaret Murie Life Sciences Building via an underground tunnel, allows personnel to travel, and animal-subjects to be transported, between animal housing and the imaging suite in controlled environmental conditions. The BiRD Facility is staffed by veterinarians, veterinary technicians, and Animal Resources personnel, and houses the facilities for laboratory animal holding and care, as well as surgical and necropsy suites. The Animal Resource Center assumes responsibilities for animal care with oversight by UAF's IACUC as specified in UAF's Animal Welfare Assurance on file with OLAW and in accordance with the PHS Policy for Human Care and Use of Laboratory Animals.

The BiRD building contains 8 animal suites, each with two or more animal holding rooms and two or more procedure rooms. Three of these suites contain two, 15 x 20 ft environmental chambers for a total of 6 chambers for housing hibernating ground squirrels. Four chambers are capable of maintaining ambient temperature of +30 to -20C and two can maintain temperatures of +30 to -40C. Respirometry and telemetry equipment is located adjacent to environmental chambers for monitoring hibernating animals. One procedure room in each suite with environmental chambers contains all surgical equipment for implantable telemetry.

### **Large Animal**

*The large animal hibernation facility* is part of the Animal Resource Center, but is described here to illustrate how instruments described above can be used for the study of large hibernators including black bear. The facility is located in an isolated, undisturbed location in a spruce forest within the UAF Biological Reserve, with no exposure to artificial light or human activity (Fig. 1). It consists of four 3 x 1.8 x 2 m or 2.5 x 2 x 2 m steel bar/heavy duty fencing wire enclosures, each with an artificial den (91 x 97 x 98 cm internal dimensions, 865 L). The dens are constructed from welded 2.5 cm thick HD-polyethylene and insulated with 5 cm Styrofoam® protected by a 2.5 cm thick outer plywood shell. The front opening (46 x 46 cm) has a double-layered breakaway door sealed with weather stripping and is kept in place by screen door clamps. Heating pads in the walls can heat the dens to 40°C above outside winter temperatures and are equipped with safety thermostats. Dens function as respirometry chambers and are instrumented with activity detectors, temperature sensors, thermostats, pressure sensors for qualitative total body plethysmography, video cameras inside and outside the dens and antennas for telemetry. All instruments except the proximity sensor are mounted in sealed penetrating tubes for easy replacement. Air is drawn from the back of the dens through heat taped tubes contained in an Arctic pipe with 10 cm insulation to prevent frost buildup in the tubes. Leakage of respiratory air is prevented by using small openings in the seals of the doors leading into the dens as air inlets.

The summer holding facility, located in the 134-acre UAF Large Animal Research Station, contains six 10.5' X 7' chain link cages arranged in groups of three. The cages are connected with tunnels so that one bear at a time can be let into the middle cage, which contains a hanging tire and functions as an enrichment cage. It can also be used for weighing the bears with a T.R.U.E Test load bar dedicated to the facility. Each cage is equipped with food and water trays, and each of the four permanent holding cages in the corners is equipped with a high density polyethylene nest box. The facility is designed with a second chain link barrier to prevent escape, and has access to water and a septic system. An instrumentation room houses a fully automated respirometry system.

### **Surgical Suites**

Surgical procedure rooms are equipped with downdraft tables, ventilators, surgical microscopes, temperature-controlled blankets, isoflurane vaporizers, pulse oximeters and all surgical equipment and supplies needed to implant a variety of transmitters. One surgical suite houses a GEM 3000 blood-gas analyzer. A separate surgical suite for large animals is located centrally to veterinary diagnostics within the BiRD building. This suite contains all equipment necessary to implant transmitters in large animals (e.g., black bear), including a standard monitoring equipment with ECG, pulse oximetry and temperature probes, cauterization equipment, ultrasound imaging, temperature-controlled blankets, ventilator, and closed circuit isoflurane vaporizers.



*Bear Hibernation Facility (Figure 1)* Hibernation facility showing two of the four cages, dens with instrumentation and instrument hut.

Den sites have breakout boxes for power, thermostats, and instrumentation, and dedicated raceways from each den to protect signal cables. The nearby 16'x24' instrument hut contains all necessary instrumentation, including a fully automated respirometry system with data acquisition computers, breakout instrumentation panels, base station for telemetry with receivers and EMG filters/integrator/activity counters. It is equipped with a 7kW UPS/voltage stabilizer, and connected to the UAF network by fiber optic cable. Thus all signals can be monitored remotely from the UAF network. The facility is approved for holding 4 bears in a hibernating state.

#### *Arctic Ground Squirrel (AGS) Breeding Facility (Figure 2)*

The recently renovated 5,000 sq. ft outdoor AGS Breeding Facility, also located on the UAF Large Animal Research Station, provides breeding and husbandry for AGS to support the research needs of the Center for TRiM and other investigators for biomedical research purposes. Previously, investigators had to rely on live-trapped AGS for laboratory investigations that can be confounded by the presence of pathogens including Arctic Squirrel Hepatitis and *Salmonella* spp., both of which have been found endemic to Alaska's wild AGS population. The facility-bred animals will be screened for hepatitis by the Director of the Virology Unit of the Alaska State Public Health Laboratory, in collaboration with the UAF Animal Resource Center (ARC).

Proposed for completion in Summer 2021, the space includes one large open-area facility with six safe and compliant 500 sq. ft encapsulated pens built to withstand the rigors of Fairbanks climate conditions on grounds similar to those found north of the Brooks Range where wild AGS live. This outdoor facility will reliably and sustainably produce virus-free AGS and AGS tissues for experimental use which is preferred over inside

laboratory breeding conditions with care provided by the ARC to ensure compliance with Animal Welfare Regulations and the ADA.



**FIG. 2. AGS Breeding Facility alteration and renovation in progress, Summer 2020.**

#### **UAA Vivarium**

UAA Laboratory facilities include a vivarium housed within the ConocoPhillips Integrated Science Building (CPSB). The vivarium is a 5925 sq. ft, fully staffed facility located on the first floor of the CPSB designed and constructed for research and husbandry of small vertebrate animals (up to 5 kg). It consists of 14 animal holding rooms, 4 common rooms (surgery/necropsy, central cage wash, storage, quarantine), 2 hibernation chambers, 2 procedure rooms, a staff office with associated locker room, toilet and shower, a janitorial closet and a room for storage of feed/bedding storage. The hibernation suite for AGS is 602 sq. ft and contains 1 holding room, a central work area, and the 2 hibernation chambers noted above. The vivarium has capacity to hold up to 90 euthermic squirrels and up to 60 hibernating squirrels simultaneously in the hibernation suite. Additional euthermic squirrels can be accommodated in other animal holding rooms. Equipment in the vivarium includes VetScan VS2 (Abaxis, Inc), autoclave, automated cage washer, and sufficient cages, tubs and racks and assorted supplies for holding arctic ground squirrels during both the hibernation and active seasons. Animal monitoring equipment is also located in the hibernation suite, including: body temperature (DataSystems International, DataQuest) and metabolic rate/respirometry (Sable Systems) equipment.

#### **Computers:**

In addition to instrument-dedicated computers, the Molecular Imaging Facility also maintains four computers for use as processing workstations for the convenience of the researchers and students making use of the Facility. Each of the stations has licenses for a variety of software including Topspin and Mestrenova for the processing of spectra and their inclusion in reports, presentations, and posters.

The imaging suite also includes a workspace with computers available for the storage and analysis of MRI imaging. The computers include a Dell running windows 7 and an Apple computer running OS X. Software licenses include OsiriX and 3D Slicer for the interpretation of a variety of images and data.

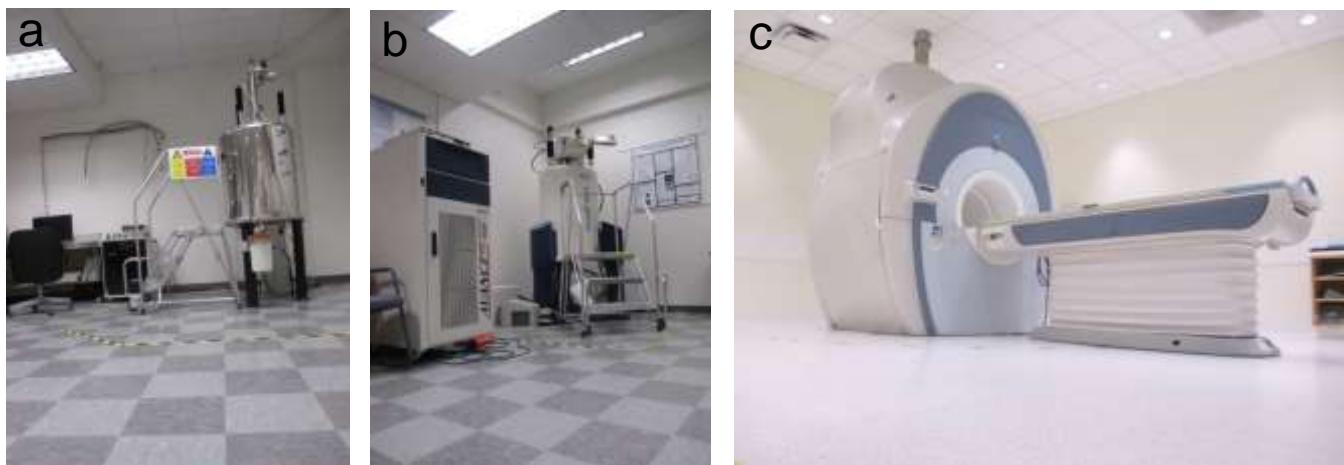
#### **Office:**

The Animal Instrumentation Manager has office space across the street in the Arctic Health Building. The Molecular Imaging Facility Manager also has desk space within the NMR room, with workspace available in the Imaging suite.

**EQUIPMENT:**

**HaMR Core - Molecular Imaging Facility - NMR Suite, Paul B. Reichardt Natural Science Building**

The **Varian Mercury 300 MHZ NMR** is capable of one and two dimensional NMR experiments, and in addition to its role in research, is used by several undergraduate teaching labs. The instrument is equipped with a quad-nucleus probe designed to readily detect the most commonly requested nuclei; hydrogen, carbon, fluorine, and phosphorus. The probe can be manually tuned for experiments where nitrogen, vanadium, or other nuclei is desired.



**Fig. 3 The Molecular Imaging Facility's a) 300MHz and b) 600MHz NMRs within the NMR suite in the Paul B. Reichardt Natural Sciences Building at UAF. c) The MRI Room within the facility's Imaging suite in the Margaret Murie Building.**

The **Bruker Avance III 600 MHz NMR** is primarily used by researchers, but is also occasionally used for upper-division lab classes. In addition to the liquids probe which can acquire on most nuclei, this system also has a High-Resolution Magic Angle Spinning (HR-MAS) probe which can acquire hydrogen and carbon spectra on tissues and other semi solid samples. The NMR suite also includes a Cross-Polarization Magic Angle Spinning (CP-MAS) probe providing solids capabilities to the system.

Both of these spectrometers are able to perform a wide range of modern NMR techniques, and are used by researchers in several departments in areas as diverse as small peptide structure determination, organic teaching chemistry support, metabolomics, and environmental samples.

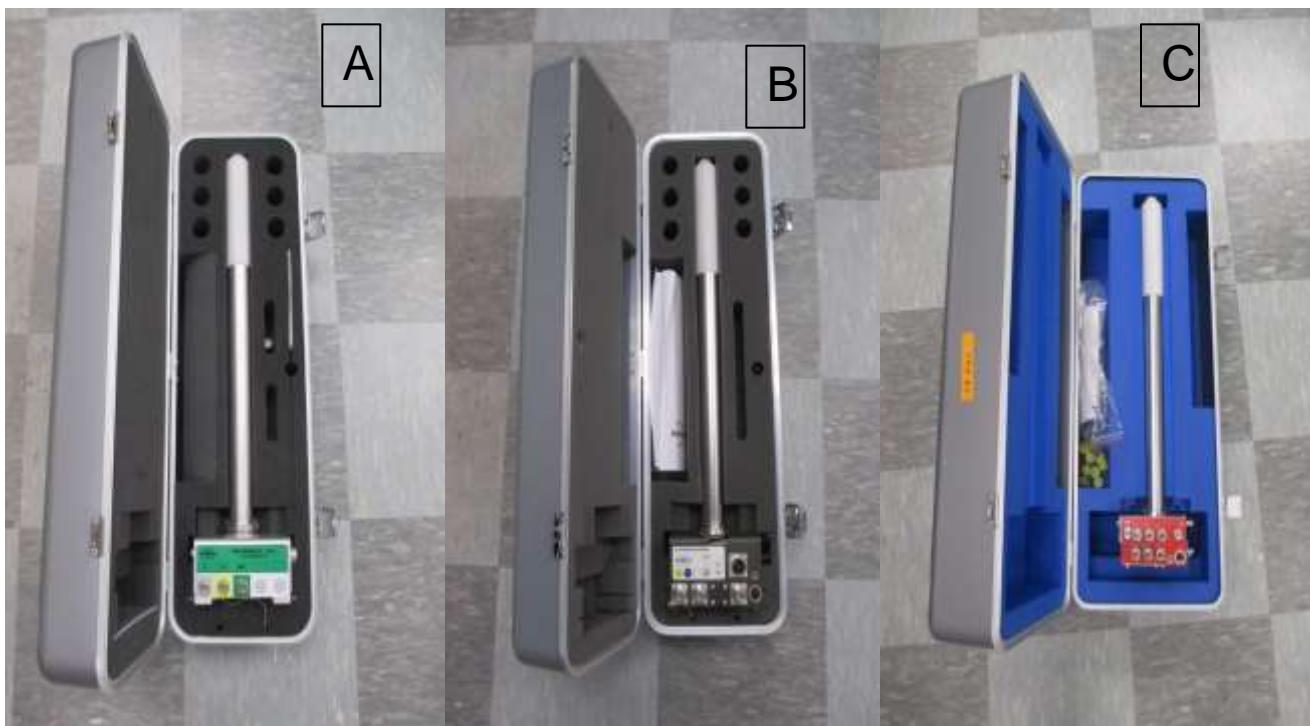


Fig. 2 NMR Probe for the 600 MHz Bruker Avance-II NMR A) Liquids, B)HR-MAS, C) CP-MAS.

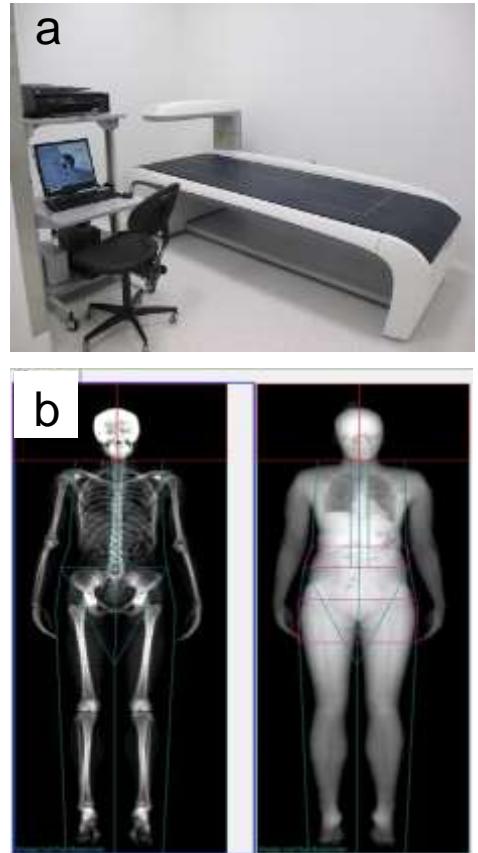
#### HaMR Core – Molecular Imaging Facility – Imaging Suite – Margaret Murie Life Science Building

The **1.5 Tesla Toshiba Excelart/Vantage MRI** is equipped with a short, 1.4 meter magnet with a 65.5 cm aperture large enough to accommodate humans, black bears, muskox and large dogs. The instrument has a quiet pianissimo technology that minimizes stress on the animals. The instrument acquires images of high homogeneity (<2ppm, 50cm FOV) and is capable of multi-planar imaging, direct visualization of arterial anatomy (MRA/MRV), Gradient Echo/Fast Field Echo for quantification of blood flow, and hydrogen Magnetic Resonance Spectroscopy (MRS). The MRI provides researchers a full range of standard experiments including T1,T2, and proton density weighted imaging, 3D-isometric imaging, and MR Spectroscopy available down to 1 cm<sup>3</sup> voxel. Additionally, ECG, respiratory, and peripheral gating options are available. The imaging facility also possesses a **non-magnetic gurney** for large animal transport, and a **gas anesthesia machine** that can be used in the MRI scan room near the magnet. The MRI conditional anesthesia machine interfaces directly with the oxygen and waste anesthesia gas disposal lines that were installed throughout the MRI suite during the construction. The machine can be readily switched between a re-breath circuit, a Bain non-re-breath circuit for small animals, and ventilator operation. In addition to the ECG, respiratory, and peripheral gating options standard with the Toshiba MRI software, the facility also contains MRI safe small animal monitoring equipment from Biopac (**MP150** data acquisition and analysis system). This equipment provides the capability to monitor

the condition of animals while they are in the MRI. The available parameters include pulse rate, percent blood oxygen, surface temperature, rectal temperature, respiration, ECG, and EEG.

The MIF also possesses a variety of coils which allow for higher resolution imaging of specific body regions: The 65.5 cm diameter **QD Whole Body Coil** is built into the MRI magnet and can be used for most experiments on any region of the body. As this coil is stationary during couch movements, the whole-body coil can be used for full-body scans without having to move the subject. The **QD Head Coil** is a transmit and receive coil designed for brain imaging that can be used on anything up to a 25 cm diameter. The **QD Head speeder coil** with neurovascular attachment can perform any of the experiments available on the head coil while adding the neck and upper spine to the available scanning area. The **QD C/T/L Spine Array** is designed for imaging the 2-3 inches of space closest to the coil surface. The coil is separated into six regions enabling the operator to choose which part of the spine to image by selecting a combination of two to four adjacent elements. The total available active area of the coil is ~72 centimeter allowing for complete spine imaging without having to change the position of the subject. The **QD Torso Speeder** is a two-part coil that can be placed above and below the subject for detailed images. In combination with the use of ECG leads, this coil allows for a full array of cardiac scans to be performed. The **QD Knee coil** is also a transmit and receive coil, but with a smaller diameter than the head coil (~20 centimeters). The design of this coil allows for easy switching between the left and right sides for imaging the knees. The **Shoulder array coil** is shaped to match the contour of the shoulder enabling imaging of any large curved regions, such as the shoulder, that may be awkward to image using other coils. The **GP flex coil** is a highly flexible and adjustable coil that is useful for a wide variety of applications (<20cm) including elbows and hands. Finally, the facility has **two 70mm circular coils** that can be used either individually, as surface coils, or in combination to image larger volumes. The use of the mounting platform enables temporomandibular joint (TMJ) imaging on both sides of the head in one acquisition.

The imaging suite of the Molecular Imaging Facility also contains a **General Electric iDXA Lunar Scanner** that was acquired through a prior COBRE grant and installed in the imaging suite in early 2016. The system allows for rapid body scans (7-10 min) and can be used for the measurement of lean body mass, adipose tissue, bone mass, and bone mineral density. The scanner utilizes the enCORE Software Platform for performing the analysis, and can work in both soft tissue mode and bone scan mode. An example showing bone and tissue scan modes is shown in figure 7. While mostly used for total body scanning the DXA has the options available to scan specific regions including the spine, and other long bones that are most likely to suffer from osteoporosis.

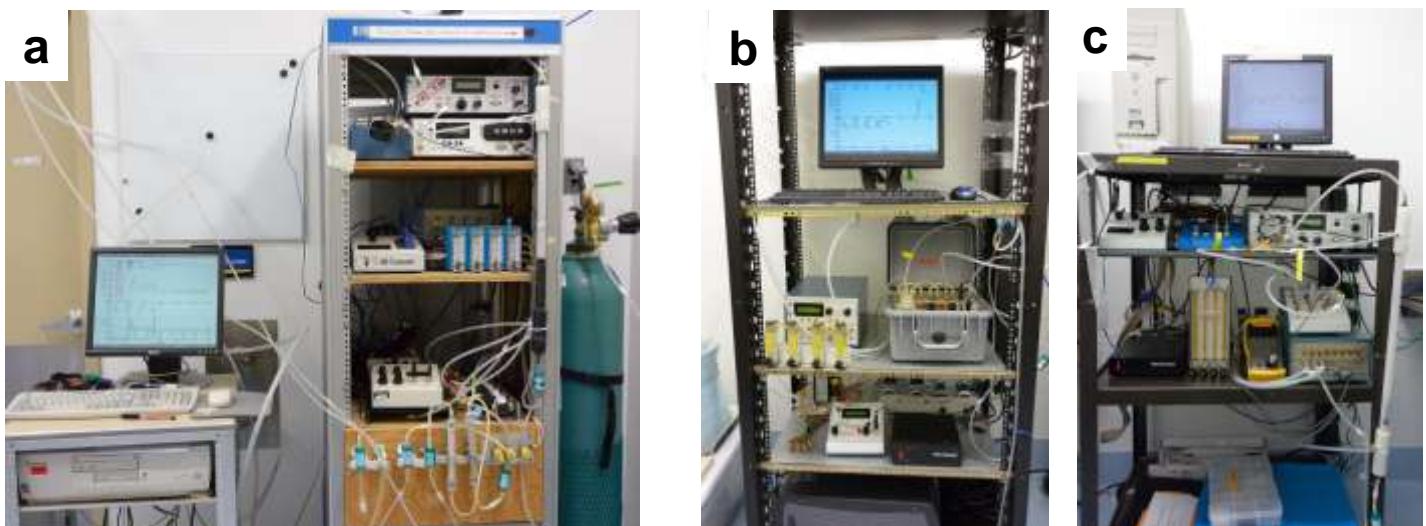


**Fig. 3a)** General Electric iDXA Scanner and **b)** example bone and soft tissue images taken using the scanner (used with permission).

## HaMR Core – Animal Instrumentation – Animal Resource Center – UAF Biological Research and Diagnostics Building

### ***Three different fully automated small animal respirometry systems:***

- a) The first system consists of a single channel FC-1B O<sub>2</sub> analyzer, a CA-2A CO<sub>2</sub> analyzer, a RM-4 gas multiplexer, a MFC2 controller, 2 Sierra mass flow controllers, a gas sample pump and KNF subsample pumps, a TC-1000 thermocouple meter, a subsampler, a UI2 data acquisition device (all from Sable Systems International), and a Nafion dryer used in reflux mode to dry gases. Additional components have been added to monitor flow and switch calibration gas.
- b) The second system is based on a Sable Systems FoxBox O<sub>2</sub> and CO<sub>2</sub> analyzer, and includes the addition of a MFC2 controller, 2 Sierra mass flow controllers, a modified RM-4 gas multiplexer with additional channels to switch calibration gases, KNF sample and subsample pumps, a Nafion dryer, and rotameters for subsample flow control. The Foxbox is used as the data acquisition device on this system.
- c) The third system is based on a Sable TurboFox O<sub>2</sub> and CO<sub>2</sub> analyzer, a RM-8 gas multiplexer with channels modified to switch calibration gases, a MFC2 flow controller, 2 Sierra massflow controllers, KNF sample and subsample pumps, rotameters for flow controls, a Nafion dryer, a TC-1000 thermocouple meter, and a UI2 data acquisition device. This system can also be run as an O<sub>2</sub> only system running a FC-1B analyzer in place of the TurboFox analyzer.



**Fig. 4 Fully Automated respirometry systems located in the Animal Resources Center at UAF.**

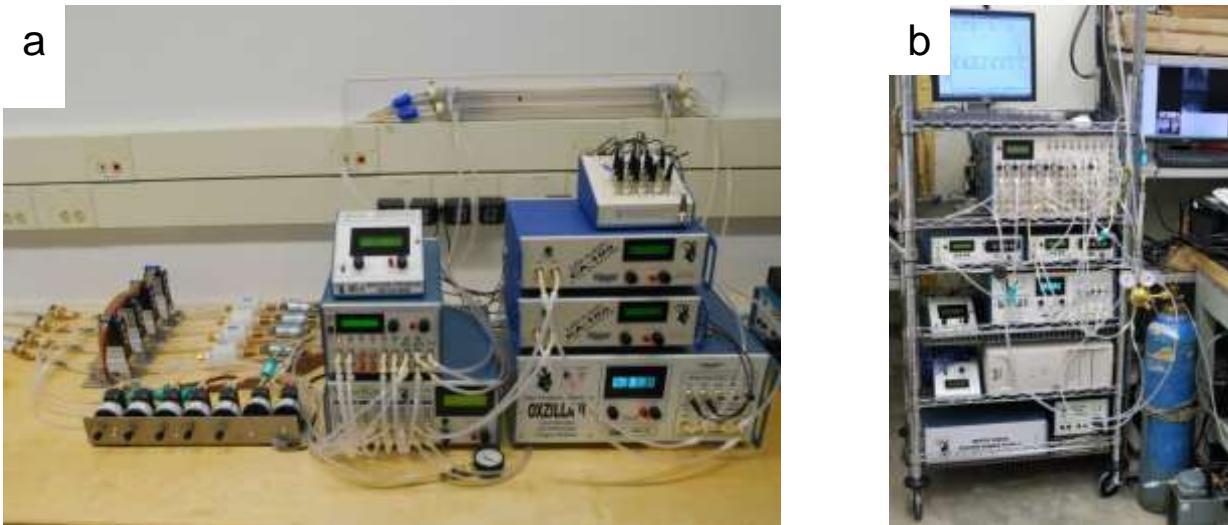
**Implantable Telemetry Systems:** Two DSI Physiotel systems are located in the Animal Resources Center in the Biological Research and Diagnostics building on the UAF Campus. One system at UAF runs on Ponemah 6.32 software (DSI) and has the capacity to simultaneously measure pressure (arterial, left ventricular, ocular, bladder, intra-cranial pressure transmitters), biopotentials (ECG, EMG, EEG, EOG), or core body temperature, from up to sixteen individuals when paired with the appropriate implanted transmitter; this system could also be upgraded for blood glucose measurements with an additional software package. The second system at UAF includes 65 receiver plates (i.e., capacity for simultaneous measurement from 65 animals) but runs on older Dataquest ART software (DSI) which is no longer supported. Funds have been budgeted to upgrade this system to the newer DSI Ponemah 6.32 Software, which also requires upgrading some hardware components (DSI Matrices). This upgrade will result in compatibility across the three acquisition systems and greatly enhance the capacity for metabolic studies

by permitting the measurement of additional physiological variables in high resolution, including ECG, EEG, and blood glucose levels in rodent models, including hibernating ground squirrels.

### HaMR Core – Animal Instrumentation – Bear hibernation and summer housing – Biological Research and Diagnostics Building

#### ***Two additional fully automated large animal 4-channel respirometry systems:***

- a) The first 4-channel system consists of dual filters, four Brooks 5861E mass flow meters 0-50L/min, four Gast sample pumps, KNF NMP830 subsample pumps, a MFC4 controller, a modified Flowbar8, a modified Mux-3 multiplexer, a 2-channel Oxzilla 2 oxygen analyzer, and two CA-10A CO<sub>2</sub> analyzers, a UI2 data acquisition device (all from Sable Systems International Inc.), and custom-built Nafion dryers used in reflux mode. The system is controlled using Dr. Tøien's data acquisition software LabGraph, calculates results on-line, and may run, if needed, unattended for months at a time. Remote monitoring is possible though network file sharing, with the remote computer running a special monitoring version of LabGraph.
- b) The second fully automated 4-channel respirometry system is functionally similar to the system described above, but uses a custom-built unit for flow control, subsample pumps and Nafion dryers (Sable Systems International Inc.); the two CO<sub>2</sub> analyzers are Sable Systems model CA-2A. Currently two Hastings flow meters are available, and one Brooks 5861E mass flow meter. In addition, 4 Brooks Massflow controllers can be used with this system to record metabolic rate of rat or Arctic ground squirrel sized animals.



**Fig. 5. Fully automated respirometry systems in the bear (a) hibernation facility and (b) summer holding facility.**

### HaMR Core – Animal Instrumentation – Vivarium – UAA

A **fully automated 4-channel system**, designed by Dr. Tøien, is located in the vivarium at the University of Alaska Anchorage and is capable of metabolic rate measurements in rat to Arctic ground squirrel sized animals both in an euthermic and hibernating state (Fig. 6). The system is principally similar to the one in the bear facility with the addition of valves that allow automatic switching to low flow during hibernation and different sample pumps. This 4-channel system consists of filters, four Brooks 5851E mass flow controllers 0-5L/min, four KNF N86 sample pumps, 4 BL-1 baselining units to switch flow, KNF NMP830 subsample pumps, an MFC4 controller, a modified Flowbar8, a modified RM-8 multiplexer, a 2-channel Oxzilla 2 oxygen analyzer, and two CA-10A CO<sub>2</sub> analyzers, a UI2 data acquisition device (all from Sable Systems International Inc.), and 24" Nafion dryers used in reflux mode. The system is controlled using Dr. Tøien's data acquisition software

LabGraph, calculates results on-line, and can run unattended for weeks at a time, if needed. The software is capable of switching between high and low flow rates using different flow controllers/flow meters and pumps depending on the O<sub>2</sub> concentration coming from the animal chamber. This allows for accurate unattended measurements to be achieved safely both in a hibernating and euthermic state.

**Implantable Telemetry Systems:** A third system housed in the Vivarium in the Integrated Sciences building on the UAA Campus. This system includes receivers for simultaneous measurement on 26 animals. Funds have been budgeted to upgrade this system to the newer DSI Ponemah 6.32 Software, which also requires upgrading some hardware components (DSI Matrices). This upgrade will result in compatibility across the three acquisition systems and greatly enhance the capacity for metabolic studies by permitting the measurement of additional physiological variables in high resolution, including ECG, EEG, and blood glucose levels in rodent models, including hibernating ground squirrels.



*Fig. 6 A fully automated respirometry system located in the Integrated Sciences Building at UAA.*

## CENTER FOR TRANSFORMATIVE RESEARCH IN METABOLISM (“Center-TRIM,” “TRIM,” or “the Center”)

The UA Center for Transformative Research in Metabolism was established in 2020 with a mission to bring together existing research activities related to hibernation, metabolism, and metabolic health through multidisciplinary research teams with the long-term goal of discovering basic mechanisms to reversibly manipulate metabolic, homeostatic, or related processes and to develop therapies and diagnostics to treat metabolic disease such as sarcopenia, diabetes, obesity, and cardiovascular disease to promote healthy aging across the life span.

The Center is housed within the Institute of Arctic Biology with access to administrative services within the Institute as well as other centralized pre- and post-award services within the research infrastructure at the University of Alaska Fairbanks. The Center supports research within its mission by bringing together researchers within and outside of the University of Alaska for journal club and seminar presentations, collaborative projects, student mentoring, investigator mentoring, sharing of resources, and sharing of technical expertise. The Center supports one Administrative Core devoted to supporting research activities for Center faculty and two technical cores, the Health and Metabolism (HaMR) Core and the Advanced Instrumentation in Microbiome Studies (AIMS) Core. The Center offers funding for technique development, developmental projects and pilot projects building on the Center’s theme and resources.

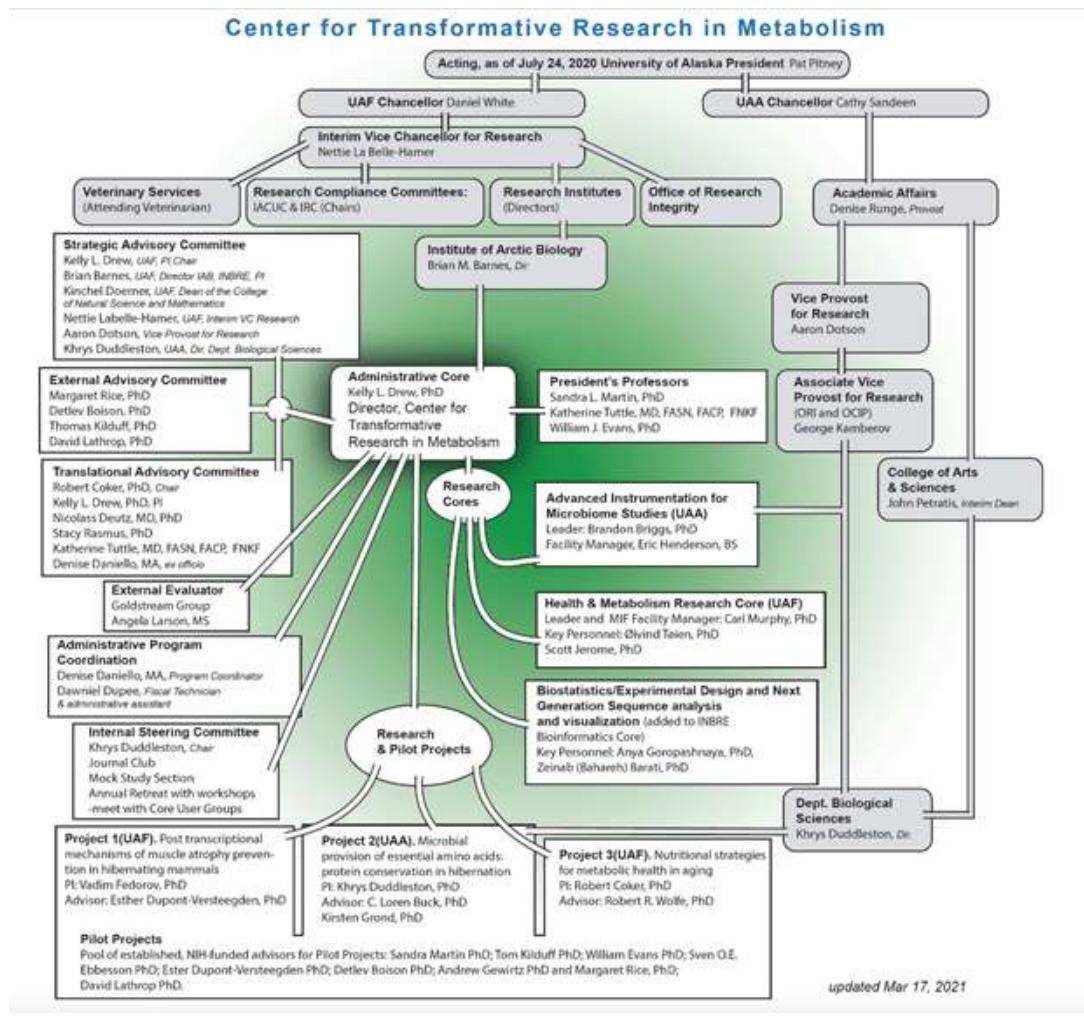


Figure 1: Organizational Structure for the Center for TRIM

Figure 1 shows the broad functional organizational structure of the Center for TRiM. The HaMR Core is an important regional resource, identified as a clinical research center site within the Northwest Participant and Clinical Interactions Network (NW PCI). HaMR Core services include technical support and expertise in methodologies for monitoring metabolism, via open-flow respirometry and biotelemetry, in hibernating species and laboratory rodents.

For more information about the Center for TRiM or the HaMR Core or to request a copy of this document in Word or google doc format, please contact:

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