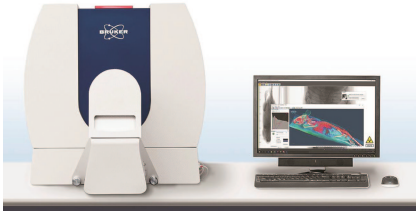
LIFE SCIENCE

NEWSLETTER OF THE UNIVERSITY RESEARCH FACILITY IN LIFE SCIENCES, THE HONG KONG POLYTECHNIC UNIVERSITY / IS06 / FALL 2021

NEW AT THE ULS

Bruker SkyScan 1276 *In Vivo* Micro-CT Scanner

Our new Bruker SkyScan 1276 micro-CT scanner is now open for training and booking. The system is equipped with a high-resolution CCD camera with variable magnification to achieve a wide range of field-of-view to suit biological samples of varying sizes. Other important features include continuous gantry rotation and step-and-shoot modes which could shorten the scanning time, and helical scan to generate distortion-free images. To enable *in vivo* micro-CT scanning, the system emits relatively low doses of radiation during imaging, and is equipped with a physiological monitoring system. The recorded data, including movement, electrocardiography and respiration, may also be utilised in the synchronised scan mode for generating time-resolved tomographic images of an area of interest.



The Bruker SkyScan 1276 micro-CT scanner.

Zeiss Lightsheet 7 Selective Plan Illumination Microscope

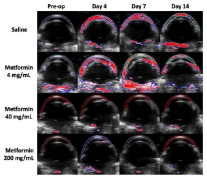
The ULS has upgraded the Zeiss Lightsheet Z.1 selective plane illumination microscope to the latest Lightsheet 7 model. The upgrade includes the acquisition of new imaging chambers that allow for partial imaging of samples as large as 2×2×3 cm. In addition, the new 5× dry and 20× immersion objectives are compatible with "cleared" samples with a wide range of refractive indexes (*i.e.*, from 1.33 to 1.58), facilitating the use of such popular tissue clearing techniques as CUBIC, CLARITY, iDISCO and BABB.

Cytiva ÄKTA Pure 25 M1 Protein Purification System

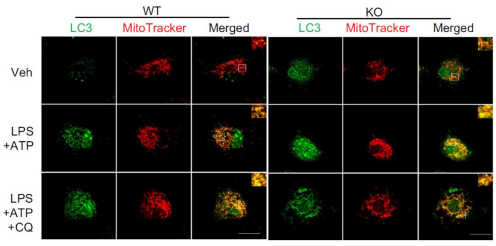
The Cytiva ÄKTA Pure 25 M1 protein purification system recently acquired by the ULS allows fast purification of proteins, peptides and nucleic acids from microgram to gram levels. Flexible purification of target products can be achieved by the 4 buffer inlet valves, system and sample pumps for automated sample loading, and outlet valve and fraction collector for product collection. Besides, UV and conductivity monitors are included for triple wavelength detection and buffer gradient verification, respectively. Overall, the ÄKTA purifier is a useful piece of equipment that facilitates biochemical analysis studies by ULS users.

POLYU RESEARCH

1. Prof. WT Wong's group (ABCT) have demonstrated the use of photoacoustic (PA) imaging to determine the degree of haemodynamics and oxygen saturation in cornea using a rat model, which might prove to be a useful noninvasive method to visualise angiogenesis in a clinical setting. The Fujifilm US/PA imaging system was used in this study.



Above: Overlaid PA and US images visualising the percentage of saturated O_2 in rat cornea. *Biomed. Opt. Express* **12**, 3597 (2021). *Below*: Confocal images showing autophagy (LC3) and mitochondrial markers in wide-type (WT) and *APPL1*-knockout (KO) groups. *Nat. Comm.* **12**, 6637 (2021).



2. Dr Kenneth Cheng's group (HTI) have demonstrated that mitophagy was impaired in mice deficient of APPL1, leading to hyperactivation of NLRP3 inflammasomes in macrophages. The results suggest a possible regulatory mechanism linking the early endosome machinery to the prevention of inflammasome hyperactivation. The Leica SP8 Multiphoton Microscope was used in this study.

METABOLIC CAGES

Tapping into the secret life of laboratory mice

In vivo metabolic studies can be challenging since it requires precise and real-time monitoring of various aspects of the animal model, such as respiration, food and water consumption, as well as physical activities. The Sable Promethion metabolic cage system which is available at the ULS offers users numerous useful features and makes this kind of *in vivo* metabolic monitoring and recording easily accessible **(Fig.1)**.

Figure 1. The Sable Promethion metabolic cage system at the ULS comprises 8 cages for individually collecting metabolic and behavioural data of mice over a desired experimental duration.

The Promethion metabolic cage system allows users to simultaneously monitor different metabolic and behavioural parameters, for example, food intake, energy expenditure, physical activities. All recordings are performed in a highly automated manner, and comprehensive data analysis tools are available for users to organise large sets of collected data into presentable charts and figures with ease.

Energy intake of mice can be calculated by food intake and calorie content of the food consumed. The food intake monitoring system is equipped with high-sensitivity sensors to measure real-time food intake of animals. The computed feeding hopper allows researchers to customise feeding strategies, such as paired feeding, time- and duration-limited feeding, and quantity-limited feeding.

Indirect calorimetry is a useful tool for measuring energy expenditure and examining the energy source of the body. Respiratory exchange ratio (RER), which is defined as the ratio of rates of oxygen consumption (VO_2) to carbon dioxide emission (VCO_2), can be used to determine the energy source being used at the moment (*e.g.*, an RER of 0.7 indicates that fats are being used; 0.8 for proteins and 1 for carbohydrates). Based on these data, energy expenditure (EE) in mice can be calculated using the Weir equation. The Promethion metabolic cage system allows real-time monitoring of these important parameters.

On the other hand, real-time locomotion activities can be recorded by the BXYZ beam break activity monitor. With this activity monitor, various types of vibrations (from shivering to running) can be detected and subsequently translated into an index of locomotion activity.

Examples of data obtainable with the Promethion metabolic cage system is shown below **(Fig. 2)**. In this study, the authors demonstrated that an isocaloric twice-a-day (ITAD) feeding strategy could enhance energy expenditure in daytime and reduce weight, without affecting voluntary locomotion of the mice. The ITAD feeding strategy was achieved by applying the computed feeding hopper to limit the time and duration of feeding. Such a diverse selection of feeding and data acquisition options would without doubt make the Promethion metabolic cage system an essential tool for researchers carrying out energy metabolism studies.

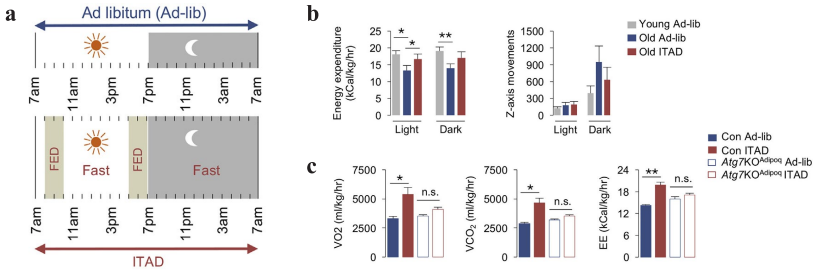


Figure 2. Examples of data obtainable with the Promethion metabolic cage system. **(a)** Schematic representation of the ITAD feeding strategy. **(b)** Energy expenditure and locomotion activity amongst ITAD and control groups. **(c)** The increase in energy expenditure upon ITAD treatment was absent in *Autophagy-related 7* (*Atg7*)-knockout mice. Adapted from Martinez-Lopez *et al.* (2017); *Cell Metab.* **26**, 856.

COMING SOON

An Abberior stimulated emission depletion (STED) super-resolution microscope is being acquired with the support from the RGC Collaborative Research Fund 2020/21. The system is expected to be installed in Q2 2022.

GET IN TOUCH

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ULS EQUIPMENT AT A GLANCE

Mass Spectrometry

- Bruker AmaZon Speed ESI-ion Trap-ETD MS
- Bruker UltrafleXtreme MALDI-TOF-TOF MS
- Agilent 6460 Triple Quadrupole LC/MS
- Agilent 6540 Quadrupole-TOF LC/MS
- SCIEX 6500⁺ LC/ESI-QTrap MS System
- Waters ACQUITY H-Class UPLC with QDa Mass Detector

Fluorescence Microscopy

- Nikon N-SIM/N-STORM/A1 Super-resolution/ Confocal Microscope
- Nikon Eclipse T*i*2-E Live-cell Imaging System
- Nikon SMZ1270i Fluorescence Stereo Microscope
- Nikon NIS-Elements Image Analysis Software
- Leica TCS SPE Confocal Microscope
- Leica TCS SP8 MP Multiphoton/Confocal Microscope
- Zeiss Lightsheet 7 Microscope
- Aviris Vision4D Image Analysis Software
- Imaris 3/4D Visualisation/Analysis Software
- MetaMorph Image Analysis Software

Cellular Analysis

- BD FACSAria III Cell Sorter
- BD Accuri C6/FACSVia Flow Cytometers
- Agilent Seahorse XF^{e}24 Extracellular Flux Analyser
- FlowJo Single-cell Flow Cytometry Analysis Software
- Invitrogen Countess II FL Automatic Cell Counter

Biochemical Analysis

- JASCO J-1500 Circular Dichroism Spectrometer
- JASCO CPL-300 Circularly Polarised Luminescence Spectrometer
- Bio-Rad Bio-Plex 200 Suspension Array System
- Malvern MicroCal PEAQ-ITC Automated Isothermal Titration Calorimeter

Genomics and Molecular Biology

- Agena Bioscience MassARRAY Analyser 4 System
- Applied Biosystems QuantStudio 5 and 7 Flex Real-time PCR Systems
- Roche LightCycler 480 Instrument II Real-time PCR System

Small-animal Research

- Perkin-Elmer IVIS Lumina Series III Pre-clinical *In Vivo* Animal Imaging System
- FUJIFILM VisualSonics Vevo LAZR Multi-modality Imaging Platform
- Bruker SkyScan 1276 *In Vivo* Micro-CT System
- Promethion Metabolic Cage System

General Research

- Drug Formulation Facility
- Bertin Precellys Evolution Homogeniser
- Labconco Refrigerated Vacuum Concentrator
- Sartorius BIOSTAT B-Twin Fermenter
- Logos X-CLARITY Tissue Clearing System
- Cytiva ÄKTA Protein Purification System

• New or upgraded in 2021