LAB_083 Use of CaloTreadmill in Mice (Expiry: January 2027)

I. OBJECTIVE

To ensure the safe use of live mice in an indirect calorimetry treadmill (CaloTreadmill, TSE Systems). Mice undergo graded exercise tolerance testing that allows analysis of respiration and running performance.

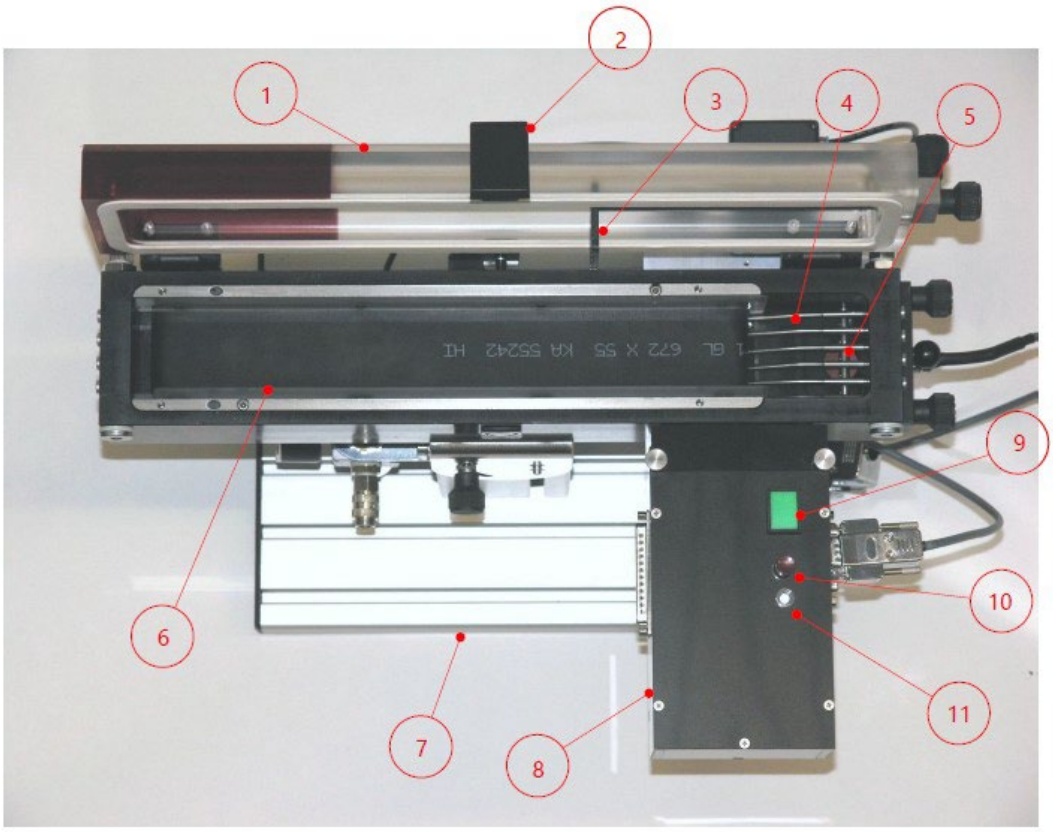

Figure 1: CaloTreadmill records and calculates oxygen consumption (VO_2), carbon dioxide consumption (VCO_2), the respiratory exchange ratio (RER), speed and distance travelled.

1. Cover (for calorimetry measurements)
2. Locking device
3. Pusher
4. Shock grid
5. Air puff
6. Running belt
7. Base
8. Servomotor
9. Start button
10. Emergency off button
11. LED

II. COMMENTS / RECOMMENDATIONS

- Relative to animal ethics applications, when using this SOP, the following must be described in the individual ethics application: any variation to this SOP, test outline and duration within the CaloTreadmill, any specific procedures (e.g. injections or treatments).
- This procedure has been written with specific reference to the Integrated Physiology Facility (IPF). The IPF CaloTreadmill includes both software and hardware (TSE Systems) components and may only be used by an IPF approved experienced operator.

  Warning: failure or improper setup of any one component may lead to malfunction and damage to the system.
- Equipment/software failures and animal escapes need to be reported to the IPF manager immediately.
- Users should further read and understand the associated Risk Assessments prior to operation: 3657 UQBR Handling and restraint of laboratory animals; 3940 Handling rats and mice (available on the UQSafe website).
- This procedure involves mouse handling and appropriate care should be taken, refer to LAB_006 Handling and Restraint in Mice and Neonates.
- Wild type and genetically modified animals must be transported to equipment as per OGTR guidelines and LAB_003 Transportation of Laboratory Rodents.
- The IPF is a shared space with unknown commensal microbial status. Once transported to a shared space it is often not possible, for biosecurity reasons, to return rodents to their original animal facility. Arrangements for transportation and ongoing care of experimental animals must be made with relevant animal facility managers when planning projects that aim to use a shared facility.

III. EQUIPMENT

- PPE

  Minimum PPE in the IPF includes gloves, gown, eye protection, face mask and closed in shoes. Additional PPE may be required based on additional risk e.g., working with infectious animals (P2 fitted mask and viral gown).
- CaloTreadmill automated exercise monitoring system

  Each treadmill can accommodate one mouse at a time.
- Desktop computer
- Scales
- Disinfectant (1-2% Virkon), Ethanol (70%)
- Clinical waste bin

IV. PREPARATION

1. Check AEC approvals to ensure that the correct procedure, personnel and facility are approved to undertake the planned work.

Deviations can occur between approved procedures listed versus what is planned with the animal, check that they match and that the relevant personnel are approved. For example, if dietary modification is intended to occur this must be specified in the AEC application.

2. Check booking dates and details with the animal facility manager.

3. All animal arrivals/departures and euthanasia's must be recorded on the Mosaic movement sheet available in the animal facility.

4. Aged animals or animals with motor impairment should be carefully monitored.

5. Prior to measurement ensure the toggle switch 'Air puff/Shock' on the treadmill control unit is set to the proper position. Settings in the software are used for defining the parameters of shocks or no shocks.

Mice can be encouraged to run by delivering a small electric discharge from the end of the treadmill (Shock intensity Mouse: 0.3 – 0.4 mA). Mice can be prevented from touching the end of the treadmill by using the pusher (Figure 1).

V. PROCEDURE

1. Check with facility staff that the CaloTreadmill has been calibrated and the belt is running at the correct speed.
2. Enter experimental parameters using the treadmill software (e.g. weight of the animal, belt speed, angle of inclination, acceleration, distance, time, number of phases, shock duration/intensity).
3. Place animal onto running surface.
4. Close compartment cover.

Allow mice to rest/freely move for at least 5-10 minutes for equilibration of gas before measurements begin. Basal measurements can be taken at this time. Mice should be monitored the entire time they are housed in the treadmill.

5. Using the software click 'Start Motor' to start the tread drive.

The motor speeds up to the initial speed value set in the profile table.

6. Click the 'Stopped' button at the top of the … and select 'Start Timer'. The display will change from 'Stopped' to 'Running'.
7. If using a speed profile select 'Start Profile'.

If the profile isn't started the running belt retains its initial speed.

8. If using the calorimetry module, select 'Measurement/Start' and open the treadmill dialog box.
9. Interruptions to the light beam sensor are recorded (fatigue), as are active shocks.
10. The treadmill can be stopped at any time by pressing 'Stop'.

Calorimetry measurements can continue into exercise recovery (around 10-15 minutes) if compartment remains closed. This is recommended.

10. Use any project specific score sheets, as approved for the individual model.

If you find an unwell mouse refer to LAB_022 UQBR Veterinary Care Program. Report any Unexpected Adverse Events to the AEC.

11. On completion of your experiment ensure all animals have been removed from the CaloTreadmill and returned to their home cage.
12. Record any movements on the Mosaic movement sheet.
13. Export all relevant data.

VI. REFERENCE INFORMATION

1. Molinero et al., (2017) Role of muscle IL-6 in gender-specific metabolism in mice. PLoS One.
2. Slater et al., (2019) Metformin improves diastolic function in an HFpEF-like mouse model by increasing titin compliance. J Gen Physiol.
3. Hingst et al., (2020) Inducible deletion of skeletal muscle AMPKα reveals that AMPK is required for nucleotide balance but dispensable for muscle glucose uptake and fat oxidation during exercise. Mol Metab.
4. Brynnel et al., (2018) Downsizing the molecular spring of the giant protein titin reveals that skeletal muscle titin determines passive stiffness and drives longitudinal hypertrophy. Elife.

Version #	Reviewing AEC (note: all other relevant AECs ratify the approval)	AEC Review Date	Approval To Date
1	MBS	19/01/2024	19/01/2024

Conditions:

- Investigators named in an animal ethics application, relative to this SOP, must be competent to implement the SOP
- Any variation to this SOP must be described in the relevant animal ethics application
- If this SOP has not been reviewed and approved by a UQ AEC within the last three years it is no longer valid and cannot be used in animal ethics applications until reapproved (see "AEC Reviewed/Approved" date in this document's header).