

<b>University Animal Care Committee Standard Operating Procedure</b>		
<b>Document No:</b> 7.27	<b>Subject:</b> Hypoxic Studies in Mice	
<b>Date Issued:</b> October 25 <sup>th</sup> , 2021	<b>Revision:</b> 2	<b>Page No:</b> 1

**Location:** Queen's University

**Responsibility:** Principal Investigators, Research Staff, Veterinary Staff

**Purpose:** The purpose of this Standard Operating Procedure (SOP) is to describe hypoxic studies in mice.

**1. Introduction and Definitions:** The purpose of this Standard Operating Procedure (SOP) is to describe the method for inducing pulmonary hypertension by exposing mice to hypoxia.

**Abbreviations:** Animal Care Services **ACS**, Principal Investigator **PI**, subcutaneous **SC**, intravenous **IV**, intraperitoneal **IP**, intramuscular **IM**, per os **PO**, per rectum **PR**

## **2. Materials:**

- ProOx Sensor
  - Calibrated to maintain the level of oxygen at 10% oxygen within the chamber. The alarm should be calibrated to allow for +/- range of 2% which, if exceeded, will cause the alarm to sound and the gas to be turned off.
- CO<sub>2</sub> Sensor
  - Measurements of CO<sub>2</sub>, O<sub>2</sub>%, temperature, and humidity will be recorded daily.
- CO<sub>2</sub> Absorbable Granules (soda lime)
- Desiccant beads (eg. DRIERITE tm)- Anhydrous indicating desiccant or equivalent
- Cages
- Water Bottle
- Nitrogen Gas

## **3. Procedures:**

- Submit a special request for supplies needed (ie. caging ,diet, water bottles) and include the study timeline.
  - The ProOx sensor will be calibrated and set to maintain an internal oxygen percentage of 10%. The sensor connection will be checked to ensure a secure contact with the controller. A loose connection can give an erroneously high oxygen reading, causing the chamber to be flooded with nitrogen, asphyxiating the mice.
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- Mice will be transferred into a clean cage with adequate food and water.
- The cage will be placed into the hypoxic chamber.
- The gas will be turned on and will fill the chamber until the ProOx sensor reads between 8-12%.
- Once the chamber is steady at the set point, the alarm will be turned on to prevent the chamber from deviating from the desired oxygen percentage (+/- 2%).
- The O<sub>2</sub>%, CO<sub>2</sub> level, temperature and humidity will be recorded daily.
- The mice will be monitored daily by looking into the chamber and assessing overall wellness.
- The chamber will be opened, and mice will be transferred into a new, clean cage weekly at the minimum and these changes will be tracked on the door sheet. The gas and alarm will be turned off when the chamber is opened.
- The new cage will be placed back into the chamber, and the gas will be turned on.
- Each time the chamber is opened, the silenced alarm must be reset once the internal O<sub>2</sub>% has stabilized.

#### **4. Complications:**

- Failure of the sensor may result in a reading of internal O<sub>2</sub>% ≤ 8% or ≥ 12%. At this point the alarm will sound. For the low O<sub>2</sub> alarm, gas will automatically shut off. Research staff will assess the sensor and nitrogen tank and determine the reason for failure before turning the gas back on.
  - The absorbable granules will be monitored for a colour change from white to purple as a measure of CO<sub>2</sub> absorption within the chamber. If the scrub is found to indicate high CO<sub>2</sub> absorption (purple colour) this will result in the chamber being opened and the scrub being changed. An additional container of absorbable granules can be added to help maintain the level of CO<sub>2</sub> within the chamber.
  - The desiccant will be monitored for a colour change as per the manufacturer's instructions as a measure of internal humidity. Change of colour will result in the chamber being opened and the desiccant being changed.
  - Cages will be monitored to ensure that the mice have adequate food, water and they will also be monitored for water leakage and flooding. For any of these events, the chamber will be opened, and the specific issues resolved.
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**References: Dr. Archer and Dr. Ormiston Laboratories**

**SOP Revision History:**

Date	New Version
7/22/2024	Updated layout and changed some language as per the labs running this procedure.