TITLE: Intraperitoneal Injection in Mice

OBJECTIVE: Intraperitoneal injections are used to administer solutions into the intraperitoneal cavity of the mouse (the cavity that encloses the stomach, kidneys, liver and other abdominal organs). An intraperitoneal injection should avoid injecting the organs.

RISK STATEMENT:

The following risks have been identified with this procedure and precautions advised to avoid injury/illness:

<table>
<thead>
<tr>
<th>PROCEDURE</th>
<th>RISK</th>
<th>RISK LEVEL</th>
<th>PRECAUTION</th>
</tr>
</thead>
<tbody>
<tr>
<td>Handling of Mice</td>
<td>Bite/scratch</td>
<td>Low</td>
<td>Effective training and precaution in handling of animals</td>
</tr>
<tr>
<td>Use of needles</td>
<td>Needle Stick injury</td>
<td>Medium</td>
<td>Training in the safe and correct use of needles</td>
</tr>
</tbody>
</table>

PPE: The following personal protective equipment MUST be used during this procedure:

- Gown
- Gloves
- Mask
PREREQUISITES:

The animal to be injected must be older than 6 weeks of age. Unless the protocol has been approved by the AEEC for mice less than 6 weeks of age
Competent in the use of needles and syringes
Competency in the restraint of mice
Competency in cervical dislocation
Competency in the use of a mouse restrain device
Training in this procedure

EQUIPMENT REQUIRED:

0.5ml or 1ml syringe with needle attached(13-25mm 25-27 gauge needle)
An appropriate disinfectant e.g. virkon
Solution to be injected
Sharps bin
Paper towel
PPE – gloves, gown, mask

BACKGROUND INFORMATION:

POSSIBLE PROBLEMS

1. Aspirate:
   - Greenish-brown aspirate indicates needle penetration into the intestines
   - Yellow aspirate indicates needle penetration into the bladder

   Note: If material from these organs is aspirated the needle needs to be disposed of as injecting either of these contents could have the potential of causing a bacterial or chemical infection in the abdominal cavity of the mouse.

2. Bulge in skin

   Injection has been done subcutaneously, absorption will be slow and in some cases you must not re-inject because of the risk of overdosing

NEEDLE SAFETY

- Gloves must be worn.
- Ensure the size of the syringe is appropriate for the volume being drawn up.
- Do not uncap the needle until ready.
- Once a needle is uncapped it is never to be recapped.
- Only dispose of needles in a sharps container.
Do not leave uncapped needles lying on the bench after they have been used, place them straight in the sharps bin.

Do not hold a needle unless you are directly using it.

**PROCEDURE**

1. Have your needle ready with the solution you need to inject drawn up.
   - Ensure there are no air bubbles present in the syringe.
   - Air bubbles can be removed by flicking the needle a few times with your finger or tapping it on a bench until all of the bubbles have reached the top.
   - Then you draw the solution back and then forward slowly till the solution fills the top of the needle. This may need to be done a few times to remove all the bubbles.

2. Restrain mouse using correct technique (refer to the ABS restraint procedure). Be sure to hold enough skin so that it cannot bite or kick.

3. Locate the midline (this is a line of fur that runs down the centre of the mouse, refer to diagram).
4. Prepare to inject on the left or right side of the mouse, half way in between the midline and the top of the hind leg.

5. Hold the mouse at a slightly downward angle
   - To allow the organs to drop towards the chest cavity. This reduces the risk of accidentally injecting into the abdominal contents
   - Ensure that you can read the graduations on the needle to determine the quantity of solution you are injecting is correct.

6. Insert the needle approximately 0.5cm (bevel up) into the abdominal cavity in the lower right quadrant to avoid the cecum and the bladder.
   - Be sure to have a steady hand injecting and do not wiggle the needle around inside the mouse as this could lacerate the organs.
7. The needle is injected at a small angle (15 – 20 degrees).
   - One practice is to place the ring finger on the stomach of the mouse and it
     acts as a measure for the angle.

8. Aspirate the needle to ensure that you are not in any organs.
   - Please see above in Possible Problems

9. If the aspiration test is negative inject the solution slowly and at a constant rate.

10. Pause for a couple seconds so that the solution does not seep out.

11. Remove the needle and place the mouse down gently

12. If requested to inject the solution on both sides repeat steps 4 -11 on the opposite side

13. Dispose of needle in the sharps bin.

**COMPETENCY**

1. Be able to restrain with minimal stress to the animal

2. Be able to understand the importance of safety around needles, syringes, and injectables and competently practice needle safety while training

3. Be able to locate the correct injection site and understand the reason why this site is preferred

4. Be able to understand and recognize aspiration material.

5. Be able to consistently perform successful intraperitoneal injections on 10 mice on two consecutive occasions, performing autopsies to confirm whether
the injection was successful.

- Injection must be dispersed evenly through the abdominal cavity from the injection site/s.
- This is determined in an internal examination of the abdomen after euthanasia.
- A maximum of two injection sites are present with no organs lacerated
SUMMARY:

- Needle safety must be followed during this procedure
- Mice must be a minimum of 6 weeks of age unless AEEC approval has been given otherwise
- Watch for incorrect needle placement into organs or skin