T400-Series Surgical Protocol

Rat Ascending Aorta: Chronic Blood Flow Measurement (Sternal Approach)

**APPLICATION BASICS**

<table>
<thead>
<tr>
<th>Site:</th>
<th>Ascending Aorta</th>
</tr>
</thead>
<tbody>
<tr>
<td>Species:</td>
<td>Rat</td>
</tr>
<tr>
<td>Body Weight:</td>
<td>277 - 300 grams</td>
</tr>
<tr>
<td>Duration:</td>
<td>Chronic</td>
</tr>
<tr>
<td>Vessel Diameter:</td>
<td>2.5 mm</td>
</tr>
</tbody>
</table>

**PROBE**

| Size:          | 2.5 mm                           |
| Reflector:     | JS                               |
| Connector:     | 4-pin                            |
| Cable Length:  | 12 cm                            |
| Catalog #:     | MC-2.5PSB-JS-WC12-CA4S-GC        |

**FLOWMETER**

TS420 Perivascular Module

**Flow Ranges Observed**

90-100 ml/min in a 300 gram conscious rat.

**Application**

This protocol was developed to evaluate systemic hemodynamic effects of selective inhibitors of cyclic nucleotide phosphodiesterase in the conscious rat model.

**Sternotomy**

A mediasternal approach can allow better visualization into the thoracic cavity for Probe placement on the aorta. Though this approach would seem more invasive, the procedure can be performed more quickly and animals may recover faster from a more expedient surgery. It also has allowed Probe placement in younger, smaller rats (150 grams).

**Advantages of Sternal Approach**

1. Least stressful to the rat for healing
2. Easy to visualize the aorta and heart
3. Fast
4. Low risk of damaging the lungs
5. Allows implants on young (150 gm) rats

**Anesthesia and Surgical Preparation**

1. Anesthetize with pentobarbital (60 mg/kg). Administer 0.1 mg/kg atropine simultaneously. Once the rat is anesthetized, administer 2 mg gentamicin.
2. Intubate with PE-190.
3. Place rat in dorsal recumbency.
4. Scrub surgical area of rat with 1.0% betadine solution and place a surgical drape over the rat.

**Fig. 1: Schematic of sternum separated to xyphoid.**

**Fig. 2: Site and orientation of Probe on the ascending aorta.**

Flowprobe Implantation
1. Make a 3.5 cm. midline muscle and skin incision over the sternum using a #11 scalpel blade.
2. Attach tracheal tube to the respirator.
3. Lift the sternum with rat-toothed forceps. Using a #11 scalpel blade, make a puncture just lateral to the midline on the left side. Puncture should be mid-thoracic.
4. Insert one blade of a 5” straight scissor into the puncture and cut on the midline of the sternum 1.25 cm rostral and caudal. Caudal extent should not exceed 1 cm above the diaphragm. Keep pleural membrane on right side intact.
5. Retract with a microretractor to expose the thymus and heart. Use a retractor with short, blunt prongs to maintain an intact right pleura.
6. Divide the thymus into the left and right lobes to expose the aorta.
7. Gently separate the ascending aorta from the pulmonary artery with a pair of curved 4” micro-dissecting forceps.
8. Use these forceps to clear 3 mm along the aorta for subsequent placement of the J-reflector of the Flowprobe.
9. Place a 5” piece of 3-O silk suture around the aorta.
10. Position the Probe with the open side of the J-reflector toward the aorta and the back side facing the pulmonary artery.
11. Using the suture, gently guide the aorta into the J-reflector, close the J-reflector, and remove the suture (Fig. 1).
12. Remove the retractor and position the cable to exit at approximately a 45 degree angle cranially such that the aorta is parallel with the J-reflector. The position of the heart and aorta within the chest are such that the Probe naturally assumes this position (Fig. 2).
13. Placing one suture on each side of the cable, suture cable in place by closing sternum rostrally.

Restoration of Negative Pleural Pressure
1. From the midline incision, the skin on the left side of the thorax is pulled away by blunt dissection to expose the pectoralis minor muscle.
2. Place a 3-O silk purse string suture in the pectoralis minor muscle 1.5 cm lateral to midline.
3. Brace the inside of the ribcage with a pair of 4” straight dissecting forceps. Make a puncture wound between the ribs inside the purse string using a pair of 4” curved micro-dissecting forceps (Fig. 3).
4. Insert a 5 cm silastic tube (0.03in. x 0.065 in) 1.5 cm into the chest cavity, through the puncture wound, and tie loosely.
5. Complete the closure of the sternal incision and muscle in two layers.
6. Using a 3 cc syringe and 19-gauge needle, draw a negative pressure through the chest tube.
7. Check sternal incision for leaks.
8. Tighten purse string while removing the chest tube.
9. Retract skin flap over purse string.
10. Close skin caudally and leave enough open rostrally for tunneling the connector.

Connector Placement
1. Using a curved hemostat, tunnel from the base of the neck, above the left shoulder, to the cranial end of the incision.
2. Grasp the connector and pull through to base of the neck.
3. Close the sternal skin incision.
4. Place connector into plastic connector plate.
5. Suture connector plate into the neck muscles cranial to scapulae.

Weaning Off Respiration
1. Apply positive end-expiratory pressure by placing exhaust line of respirator in 2 cm of water for 1 minute.
2. Disconnect respirator from tracheal tube and check for spontaneous breathing. This may require up to 1 minute to occur.
3. Extubate when the rat exhibits a strong gag reflex.

Probe Recommendations for Animal Weight & Protocol

<table>
<thead>
<tr>
<th>Weight (Grams)</th>
<th>Probe Size</th>
<th>Probe Model</th>
<th>Cable Orientation</th>
<th>Surgical Approach</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt; 250</td>
<td>2 mm</td>
<td>2 PSB</td>
<td>Back</td>
<td>Medium sternotomy</td>
</tr>
<tr>
<td></td>
<td></td>
<td>2.5 PSL</td>
<td>Lateral</td>
<td>Right thoracotomy at 3rd intercostal</td>
</tr>
<tr>
<td>250 - 350</td>
<td>2.5 mm</td>
<td>2.5PSS</td>
<td>Side</td>
<td>Right thoracotomy with CA4S micro-connector exited at 2nd intercostal</td>
</tr>
<tr>
<td>400 - 500</td>
<td>3 mm</td>
<td>3PSL</td>
<td>Lateral</td>
<td>Right thoracotomy at 3rd intercostal</td>
</tr>
<tr>
<td></td>
<td></td>
<td>3PSB</td>
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ACKNOWLEDGEMENT
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