**Perfusion methods using the 10% Neutral Buffered Formalin**

**Materials:**

1. needle connected tubing
2. peristaltic pump
3. Scissor
4. Pincet
5. Scalpel
6. 1x PBS (can be made from 10X PBS; Cat# PBS01-32R)
7. 10% neutral buffered formalin (Cat#NBF07-32R)
8. Anesthesia (phenobarbital etc.)

**Methods**

1. Anesthetized animal.
2. Cut the skin below the sternum and open the thoracic cavity and inserted the needle which connected the tubing that has PBS onto apex of the heart through the aorta and cut the right atrium to let the blood out.
3. Or you can put the tubing into the femoral artery and cut the femoral vein to let the blood out.
4. Run 1xPBS (approximately 50-60mL) through the animal slow speed to wash the cardiovascular system from the blood.
5. Once the blood appear translucent, change the PBS with 10% NBF and run with the medium speed (1-2 ml/minutes) to match hearth rate of the animal..
6. Perfuse the animal adequately at least 1ml for 1 gram body weight (if you have animals weighted 300 gr, you need to perfused the animal with 300 ml of NFB).
7. Make sure the animal is stiffed post-perfusion, if not you need to correct the position of your needle.