

## **WB Stim & Smart Tube Stabilizer – Cluster Tubes Cryopreservation**

### **Equipment:**

- Pipette Gun
- p200 micropipette
- Biosafety Cabinet (BSL-2 rated)
- Tube racks
- 37C Water Bath

### **Materials:**

- Heparin Green Top Tube (Fisher, #367874)
- Sterile, filtered, p200 pipette tips (Rainin, #RT L250F)
- 2mL, 5mL, 10mL, 25mL, and 50mL sterile, serological pipettes (Fisher, #356507, #356543, #356551, #356525, #356550 respectively)
- Nunc Cryobank 1.0mL, 2D barcoded, w/Red caps, 10 racks of 96/case (Fisher #374120)

### **Reagents:**

- SmartTube Stabilizer, SmartTube Inc.
- Cytokine Stims (provided)

### **Procedure:**

1. Remove a set of cluster tubes from the freezer and warm them in the 37C water bath for approximately 5 minutes.
2. Invert the 2.5 mL blood tube 5x to mix.
3. Add 200 microliters of blood to each of the cluster tubes, pipetting twice up and down to mix.
4. Incubate cluster tubes for 15 minutes in the 37C water bath.
5. Immediately at the end of the 15 minute incubation, add 350 microliters of Smart Tube Stabilizer (at room temperature) to each cluster tube, pipetting twice up and down to mix. Add Stabilizer in the same order of tubes as when adding blood, so that each tube's incubation time is consistent.
6. Incubate 12 min at room temperature.
7. Place in -80°C freezer.