

## Hi-C Service Sample Preparation Protocol

Customers may submit 3-10 million cells or 50-100 mg of frozen tissue for Active Motif's Hi-C service. If cells are limited, we can accept a lower limit of 1 million cells. Prepare cell pellet(s) or tissue according to one of the protocols below.

### I. Cell Pellet Preparation

#### Protocol A: Adherent Cells

1. Detach cells using dissociation method specific to your cell type/line.
2. Following dissociation method, transfer cells to a new 15 ml or 50 ml conical tube.
3. Spin at 500 x *g* in a refrigerated centrifuge (4°C) for 5 minutes to pellet cells.
4. Discard supernatant and resuspend cells in 10 ml 1X ice-cold PBS.
5. Spin at 500 x *g* in a refrigerated centrifuge for 5 minutes to pellet cells.
6. Discard all supernatant.
7. Immediately snap freeze pellet in liquid nitrogen or on dry ice.
8. Store cells at -80°C prior to shipment.

#### Protocol B: Cell Suspension (including FACS sorted cells)

1. Transfer desired number of cells to (vial/tube).
2. Spin at 500 x *g* in a refrigerated centrifuge (4°C) for 5 minutes to pellet cells.
3. Discard supernatant and resuspend cells in 1X ice-cold PBS.
4. Spin at 500 x *g* in a refrigerated centrifuge for 5 minutes to pellet cells.
5. Discard supernatant.
6. Immediately snap freeze pellet in liquid nitrogen or on dry ice.
7. Store cells at -80°C prior to shipment.

### II. Tissue Preparation

#### Consumables

- Cryogenic vial(s) **or** 2 ml low-bind microcentrifuge tube
- Liquid Nitrogen
- Dry ice

**Protocol A: Liquid Nitrogen**

1. Excise the tissue from the animal and place in a cryogenic vial or microcentrifuge tube.
2. Immediately submerge tube in liquid nitrogen for 2 minutes.
3. Store at -80°C.

**Protocol B: Dry Ice**

1. Excise the tissue from the animal and place in cryogenic vial or microcentrifuge tube.
2. Immediately place tube on dry ice for 15 minutes.

**Best practices for sending cell pellets to Active Motif**

- Avoid scraping cell off plate. Scraping cells can cause damage and induce biological changes
- Seal top of the tube with parafilm to avoid tube from opening during transit
- Ensure that there is enough dry ice in the package for transport and any potential shipping delays
- Avoid shipping over the weekend or for Saturday delivery
- Ship samples Monday through Wednesday
- Ensure that a completed sample submission form is included in shipment

**Best practices for sending tissue samples to Active Motif**

- Avoid overfilling tubes with tissue as this makes it very difficult to extract samples from tube
- Seal top of tube with parafilm to avoid tube from opening during transit
- Ensure that there is enough dry ice in package for transport
- Avoid shipping over a weekend or for Saturday delivery
- Ship samples Monday through Wednesday
- Ensure that a completed sample submission form is included in the shipment