

Procedure Title: **Thawing Cryopreserved Stem Cells for Infusion At
Childrens Hospital Los Angeles**

Procedure #: **HSC.B401.01**

I. Specimen Requirements

All units acceptable for thawing must have been maintained at liquid nitrogen vapor temperature until the time of thawing.

II. Reagents, Supplies and Equipment

- A. Sterile syringe (1 ml)
- B. 15 G 1 ½" needle
- C. Sampling site coupler
- D. Povidone - iodine USA prep pad
- E. Alcohol prep pad
- F. Cryogloves
- G. Liquid nitrogen
- H. Water bath

III. Procedure

- A. Preparation for Infusion

Note: These steps can be performed before the day of infusion.

1. Contact the bone marrow transplant unit to confirm the identity of the patient, date, time, location and number of bags to infuse.
2. Retrieve a copy of the Patient Summary Sheet to bring to the infusion
3. Begin filling out the infusion record.
4. Begin preparing a CHLA Progress Notes form (which will be kept in the patient chart).

5. Determine the storage location of the units to be infused.

B. Prepare the Water Bath

1. Measure the water bath temperature with a thermometer. Adjust the water temperature accordingly to achieve a $37^{\circ}\text{C} \pm 2^{\circ}\text{C}$ temperature.
2. When the water bath temperature has stabilized within the appropriate range, record the temperature on the Infusion Record.

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C. Thaw the Frozen Component

1. Remove the frozen unit from vapor storage. Two technicians must identify that the correct patient unit has been removed before thawing begins. Record initials on the Infusion Record.
2. Place the entire cassette into a ziploc bag. Immerse the entire cassette into the water bath.
3. After about 10 seconds, open the cassette and carefully remove the frozen component. Immediately transfer the component to a ziploc bag and continue thawing. Be sure to hold the top of the ziploc bag out of the water to keep the inside of the bag dry.
4. Record the start time of the thaw on the Infusion Record
5. As the cell suspension begins to thaw, visually inspect the integrity of the component and the cryocyte freezing bag.
6. If the component integrity is acceptable, initial in the appropriate column on the infusion record. If component integrity is not acceptable:
 - a. Document on the infusion record and notify the laboratory director or designee.
 - b. Prepare a summary of all procedural steps undertaken based on instructions from the laboratory director or designee and document on the Infusion Record in the comments section.
7. Continue thawing the component until less than 0.5 ml of ice crystals remain. The component should be slightly cool to the touch.

8. Remove the bag from the water bath. Keep at room temperature until the infusion.

D. Obtain a sample for quality testing of thawed component.

Note: Sample for quality testing will be obtained for at least the first and last unit thawed.

1. Open a sampling site coupler.
2. Pull the yellow port cover at the top of the cryocyte bag (that does not have the laminated identification label attached) to expose a port.

Note: Do not remove the second port cover as this will be needed by the infusionist.

3. Wipe the port septum with povidone-iodine, then alcohol to sterilize.
4. Attach a 1 ml sterile syringe to a sterile needle.
5. Remove the cap from the needle. Enter into the cryocyte bag through the port septum.
6. Invert the bag several times to mix the bag contents well.
7. Aspirate 0.5 ml of the sample into a syringe.
8. Remove the syringe with sample from the port.
9. Quality testing for thawed cells must be attended to immediately to minimize the deleterious effects of DMSO.
10. For CFU-GM enumeration, refer to HSC D441

E. Issue the thawed unit to the infusionist

1. Check patient identifiers with a member of the transplant team. Document this patient identification check on the appropriate CHLA Progress Notes form that remains in the medical chart.
2. Document the time the unit was issued to the transplant team on the

Infusion Record.

3. Deliver the patient summary sheet to the attending physician or designee.
4. Repeat sections C and E of this procedure for all units to be infused. For section D, see note.