CAUTION: US Federal law restricts the sale and use of this device by or on the order of a physician.

Disclaimer of Warranties

OriGen Biomedical warrants that reasonable care has been used in the manufacture of this device and that it was free from defects in workmanship or materials at the time of shipment from OriGen. OriGen's sole obligation shall therefore be to repair or replace any device which it determines was defective at the time of shipment. Because no product is completely effective under all circumstances, and because the actual use and handling of this device is beyond our control, OriGen cannot warrant for a good effect or against a bad effect in the application and use of this device. The buyer therefore assumes all liability arising from any cause for damages resulting from use, misuse or resterilization of this product. OriGen therefore gives no warranty of merchantability or fitness for a particular purpose. OriGen shall not be liable for incidental or consequential loss, damage or expense resulting from the use or application of this product. This warranty is in lieu of all other warranties, whether implied, express, oral or written, and no individual has the authority to vary the terms of this warranty.

OriGen Biomedical, Inc.
4020 S. Industrial Dr. #160
Austin, TX, USA 78744
Tel: +1 512 474 7278
Fax: +1 512 617 1503

OriGen Biomedical Europe
Drottninggatan 13
25280 Helsingborg
Sweden
Tel: +46 46 772 433
Fax: +46 42 146 074

CE 0473

www.origen.com

1. Day, John and McLelann, Mark Cryopreservation and Freeze-drying protocols *Cryopreservation of Animal and Human Cell Lines*, Humana Press New Jersey. 1995 (p179-188)

Cryostore Freezing Bag

Instructions for Use: Caution: These instructions should be read and understood by all personnel before using the device.

Intended Use: The Cryostore bag is intended to be used for blood component freezing.

Sterile: For single use only. The fluid path is sterile and non-pyrogenic.

Warning: Do not use if port and tubing closures are not in place. Port closure sterility is maintained through one freeze/thaw cycle.

Product Code	Freeze volume, ml, Max. - Min.	Nominal volume, ml
CS 50	20 - 10	50
CS 250	70 - 30	250
CS 500	100 - 55	500
CS 750	190 - 80	750

The freeze volume recommendation is based on horizontal freezing in a standard cassette (internal thickness of approximately 0.37 inches (0.9cm). If the bag is frozen in another manner, the optimum fill volume must be determined by the user.

Fluid Transfer:

Use aseptic technique.

1. Close roller clamps on transfer tube, if needed.
2. Remove luer cap from the appropriate lead and attach to transfer tubing
3. Open clamp(s) and allow blood components to enter freezing bag . Add cryoprotectant according to institutional protocol. The product should be cooled before adding cryoprotectant.
4. **Caution**: Remove as much air as possible from the bag. After air has been removed, close clamp.

5. Using a dielectric or RF sealer, heat seal donor tubing in the first 5 mm of the tube. A second seal above the first is suggested for additional safety in case of a sealer malfunction. Ensure that the sealed tube stub is shorter than the spike port covers to help prevent damage from interference. **Caution: Do not use metal clip to seal tubing.**

Label Placement:

Caution: Do not write on bag or adhere label stickers to bag as this may cause the bag to burst on re-warming.

1. The lot label attached to the hanger hole has a self-adhesive backing, and we recommend that you attach your own label to this backing.
2. The pouch pocket can be used for storing aliquots or additional labels. The pouch pocket may be intermittently sealed with a conventional heat or RF sealer. Do not seal this pocket completely closed, but leave a portion of it open.

Freezing Precautions

Caution: Always follow your Institutions' protocol for freezing. The following is recommended as a starting point only.

1. **Exterior Dry**: Before freezing, ensure that the bag and cassette surfaces are completely dry. Moisture may cause frost adhesion between the bag and the cassette making removal of the bag from the cassette difficult.
2. **Aliquots:** It is recommended that the donor tube be divided into samples by sealing it into segments. Leave the aliquots attached to ensure matching to the bag contents.
3. **Freezing Protocol**: When using rate-controlled freezing, follow institutional protocol to maintain a consistent freezing profile. A controlled freezing rate of -1 to-3°C/min. is recommended (1). When using a constant temperature freezer, an insulating sleeve can help reduce rate of freezing and provide an even shape to the frozen bag.
4. **Use Cassette**: The bag is fragile and easily abraded when frozen, handle with care when outside of the cassette. The use of a protective cassette during storage is always recommended.
5. **Thermal Cycling**: The bag is suitable for freezing in mechanical freezers or in the vapor phase of Liquid Nitrogen (LN). When stored in the vapor phase of LN, ensure that the storage temperature does not fluctuate, as thermal cycling of the bag may cause breakage of the bag when frozen.
6. **Breakage Precaution**: Storage at Liquid Nitrogen temperatures causes the Cryostore bag to become rigid, and while rare, breakage can occur from storage conditions and many other factors. If breakage is a problem in your institution, using an Origen FEP overwrap or switching to the Origen permalife bag is recommended. FEP remains flexible in LN and is completely inert. Contact Origen for mor information.
7.

Thawing recommendations

Caution: Always follow your Institutions protocol for thawing. The following is recommended as a starting point only.

1. **Re-warming burst danger**: If the bag has been stored in the liquid phase of the Liquid Nitrogen (LN) tank, LN seeping inside the bag may cause the bag to burst during thawing. If the bag has been stored in liquid phase, move the bag in the vapor phase of the LN tank or a mechanical freezer for 12 hours, if possible, before completion of thawing.
2. Remove the bag from the cassette prior to thawing and inspect it for breakage or leaks. If the bag does not easily release from the cassette, warm the cassette before removing the bag.
3. Thaw the bag in a 37°C to 40°C water bath with gentle agitation (1). Observe the container carefully during thawing. If the bag begins to swell, it may indicate that liquid nitrogen has seeped into the bag during storage. If this occurs, aseptically puncture the spike port with a needle or spike to vent the pressure.
4. DMSO can be toxic to cells at room temperature. Remove the bag from the water bath as soon as the sample liquefies. Process the sample and begin re-infusion as soon as possible after thawing