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## User's Guide

### CD31<sup>+</sup>/ CD45<sup>-</sup> Endothelial Colony Forming Cells

The following describes the preferred methods for the thawing and subsequent handling of CD31<sup>+</sup>/ CD45<sup>-</sup> Endothelial Colony Forming Cells as described by Ingram *et al.* (1).

1. As shipped, the cells are suspended at a concentration of  $1 \times 10^6$  cells in 1.0 ml 95% FBS/ 5% DMSO.
2. Recommended Thawing and Handling Procedure:
  - Preheat water bath to 37°C.
  - Place vial in the water bath and until the suspension media has melted.
  - Before opening, wipe the outside of the vial with 70% ethanol.
  - Aseptically pipette the cells into a collagen coated T-75 flask containing 9.0 ml of EGM-2 media.
3. Change the media after 3 to 12 hours in culture (37°C, 5% CO<sub>2</sub>)

#### References:

1. Ingram, D. *et al.*, Blood, 2004, 104(9), 2752.

Dynacell Life Sciences, LLC

P.O. Box 213  
Springhouse, PA 19477  
P: (215) 813-8775 F: (512) 727-1868  
[www.dynacellsciences.com](http://www.dynacellsciences.com)  
[techservice@dynacellsciences.com](mailto:techservice@dynacellsciences.com)