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## **Comparison of Cord Blood Product Thawing Methods on Cell Recovery and Progenitor Integrity**

**D. M. Regan**<sup>1</sup>, L. M. Grunzinger Nelms<sup>1</sup>, J. D. Wofford<sup>1</sup>, J. F. Alonso, III<sup>1</sup>, M. H. Creer<sup>2</sup>;  
<sup>1</sup>St. Louis Cord Blood Bank @ SSM Cardinal Glennon Children's Hospital, St. Louis, MO, <sup>2</sup>St. Louis University Health Sciences Center, St. Louis, MO.

Umbilical Cord Blood (UCB) products have traditionally been thawed using a conventional washing (CW) method intended to stabilize the cells, reduce DMSO toxicity and remove potentially ABO-incompatible RBC stroma and plasma. Concerns with CW include total nucleated cell (TNC) loss, bag breakage during centrifugation and poor reproducibility by transplant centers unfamiliar with this technique. We compared CW, albumin reconstitution without centrifugation (AR), and direct thaw (DT, no dilution or wash) methods by assessing viability and TNC, CD34 and colony-forming cell (CFC) recovery post-thaw. Ten cryopreserved UCB products were thawed, split equally into three parts, processed by CW, AR and DT methods and post-thaw tests performed at multiple time intervals up to 48 hours. Mean TNC recovery by CW was lower than DT ( $p < 0.01$ ) throughout the 48 hour interval, however, CW and AR recoveries were not different ( $p > 0.05$ ). CD34 and CFC in CW, AR and DT did not differ up to two hours but CFC recovery in DT progressively declined with no CFC recovered at 32 hours. In DT, CD34 recovery declined progressively after eight hours compared to AR and CW ( $p = 0.0009$ ). Throughout the entire evaluation, CW and AR methods performed equally well with no significant differences observed in viability, TNC, CD34 or CFC recovery. Subsequent to the success of this study, AR has been used for 6 patients with no apparent effect on engraftment compared to CW.

We conclude that removing DMSO, RBC stroma and plasma post thaw using CW is not necessary when CB products are RBC and plasma depleted before cryopreservation and have implemented the practice of reconstituting units in our laboratory where environmental conditions are controlled and the infused product characterized. Reconstituting products is safe, easily standardized and comparable to conventional wash in maintaining cellular and progenitor integrity while ensuring recipient safety during infusion.

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International Society for Cellular Therapy Head Office  
570 West 7th Avenue, Suite 402  
Vancouver, BC V5Z 1B3 Canada  
Phone: 1.604.874.4366  
Fax: 1.604.874.4378  
[martha.davis@celltherapy.org](mailto:martha.davis@celltherapy.org)