

STEM CELL LABORATORY (STCL)



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DOCUMENT TITLE:				
Flow Diagram for Processing Fresh Cord Blood Samples Using STEMvision™ Analyzer				
DOCUMENT NOTES:				

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Fresh Cord Blood Samples are delivered to HPCA and time stamped



Allow Reagents come to Room Temperature (for ~ 15 minutes)



Verify CBU sample ISBT# with the worksheet and the extra barcodes provided



Log Specimen and Assign each specimen a letter from the alphabet (i.e. A, B, C etc.)



Label a 0.5mL micro centrifuge tube with CB ISBT barcode label



Dispense 150 microliters of PBS into the micro centrifuge tube



Vortex CB Sample for 3-5 seconds and then add 50 microliters into the micro centrifuge tube



Add 40 microliters of HetaSep to the tube in the step above and mix by pipetting (5X)



Seal the micro centrifuge tube & incubate at 37° C for 20 minutes (Do NOT exceed 30 minutes)



Transfer the top layer to a clean microcentrifuge (without disturbing the red cell pellet).



Your 10X suspension (calculated HetaSep-treated CB volume and IMDM) should total 1.0 mL



Transfer 400 microliters of this 10X suspension into a 4 mL aliquot of MethoCult media



Vortex for <u>8-10 seconds</u> then let sample sit undisturbed for <u>5 minutes</u>



Dispense <u>1.1 mL</u> of this suspension into each well & Incubate at 37°C in 5 % CO₂ for 14-16 days; ADD water as recommended during that time frame.

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Signature Manifest

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All dates and times are in Eastern Time.

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