Materials and Equipment

- Leukopak collected at HemaCare Donor Center
- HypoThermosol
- Washing buffer: PBS with 0.5% human serum albumin (HSA) and 2mM EDTA
- PBS without calcium or magnesium
- Sterile conical centrifuge tubes 250mL
- Sterile 1.8mL cryogenic vials
- Labels compatible for use in freezing and liquid nitrogen
- Transfer pack container (for processing &/or storage of blood or blood components, 300ml, 600ml or 1000 ml, Fenwal)
- 60 ml syringe
- Hemostats
- Biosafety cabinet (BSC)
- Pentra XL-80
- Flow Cytometer (Optional)
- Centrifuge
- Heat-sealer

Method

1: Record the detail information of the leukopak and the total cell count by Pentra XL-80
2: Add Hypothermosol to ensure that the white blood cell (WBC) concentration is at the optimal concentration.
3: Calculate final volume for HypoThermosol Exchange
4: Print labels with the Catalog number and donor information on the Transfer pack container.
5: Spray the leukopak with 70% alcohol, wipe dry with paper towel and place into the BSC
6: Transfer the leukopak material into 250 ml conical tube(S).
7: (Optional) Mix the leukopak material and take 1 ml sample for phenotyping study (CD3, CD4, CD8, CD14, CD16, CD19, CD56, CD45, PI).
8: Spin down the leukopak material at 600xg for 10 min at RT.
9: Spray bottles of HypoThermosol with 70% alcohol, wipe dry with paper towel and transfer HypoThermosol into BSC.
10: Transfer the conical tubes with leukopak material from centrifuge to BSC.
11: Discard the supernatant; loosen the cell pellet by gently tapping the tube.
12: Re-suspend the cell pellet with washing buffer.
13: Spin down the leukopak material in washing buffer at 600xg for 10 min at RT.
14: Discard the supernatant; loosen the cell pellet by gently tapping the tube.
15: Re-suspend the cell pellet with the amount of HypoThermosol calculated as Step 3.
16: Transfer the syringes and Transfer pack container into the BSC.
17: Fill the transfer pack container with the leukopak cell suspension in HypoThermosol using the syringe.
18: Generate retention vials by transferring cell suspension in CryoStorCS10 into cryogenic vials (1ml to each cryogenic vial)
19: Triple seal the transfer pack container’s tubing port with heat-sealer.
20: Send the HypoThermosol-Leukopak to shipping department for inspection and transportation
21: Transfer the cryogenic vial with cell suspension into CoolCells and transfer the CoolCells into -80°C freezer
22: Complete and sign production records and forms.

To minimize the risk of injury or cross contamination, always wear personal protective equipment, including gloves, lab coat and goggles. For questions, please contact our scientists at (877) 944-4362.