SUBSTANTIALLY REDUCED RED CELL CONTAMINATION WITH COMPARATIVE NUCLEATED CELL RECOVERY USING SAMPLE SPECIFIC RED CELL & PLASMA REDUCTION TECHNIQUES

(A retrospective analysis of 36489 umbilical cord blood units at Lifecell International)

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ABSTRACT

BACKGROUND: Volume reduction is a widely used procedure in Umbilical Cord Blood (UCB) banking. It concentrates progenitor cells by reducing plasma and red blood cells, thereby optimizing the use of storage space.Today, manual and semi-automated (Sepax & AXP) methods are available for processing cord blood. Automated systems are limited by the presence of high level of Hematocrit in final product, which can be detrimental. Free hemoglobin;

- ✓ Can potentially cause renal failure
- ✓ Interfere with hematopoetic progenitor cells (HPCs) proliferation capacity resulting in lower colony forming units (CFUs) in culture, and delayed engraftment in patient.
- ✓ Attempting to remove free hemoglobin requires washing procedure; which can result in substantial loss of progenitor cells, effectively providing much lower number of cells for transplantation, than initially envisaged. (High cell yields are imperative for transplant success).

MATERIALS & METHODS

- This report outlines the retrospective study for 36489 UCB units processed at LifeCell International, India; with Hydroxy Ethyl Starch (HES) using Density Gradient Centrifugation Technique. The median CBU volume collected was 98 mL and final volume stored was 21.5 mL.

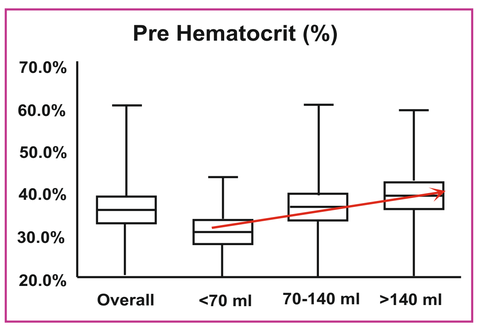
RESULTS

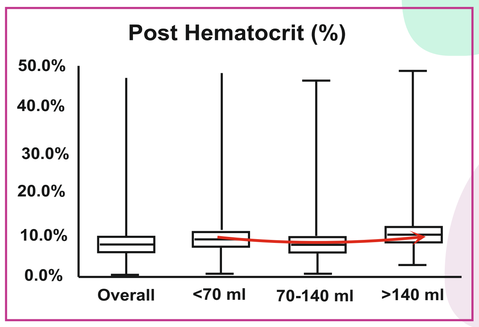
- Median nucleated cell recovery achieved was 78.00 % (comparable to those of automated techniques). Median RBC depletion achieved was 95.4%, Median Hematocrit concentration (post processing) achieved was 7.5% (significantly superior to semi/fully automated techniques.) Median yield for Total nucleated cells (TNC) (excluding NRBCs) and CD34+ cells were respectively 668 million and 2.05 million per CBU.

TABLE 1: HCT

		Pre-Hematocrit (%)		Post-Hematocrit (%)		Hematocrit Reduction (%)		
	n	Median	Mean± SD	Median	Mean± SD	Median	Mean± SD	Max
Overall	36489	36.0%	36.0%±5.0	7.5	8.3%±3.9	95.4%	93.9%±5.5	99.4%
< 70 ml	6469	31.1%	30.9% ±4.3	9.0	10.1%±5.1	89.4%	87.2%±9.3	98.7%
70 - 140 ml	27231	36.7%	36.8%±4.3	7.1	7.8%±3.3	95.8%	95.2%±2.5	99.4%
> 140 ml	2789	39.5%	39.6%±4.8	8.6	9.7%±4.6	97.0%	96.6%±1.6	99.2%

GRAPH 1.1





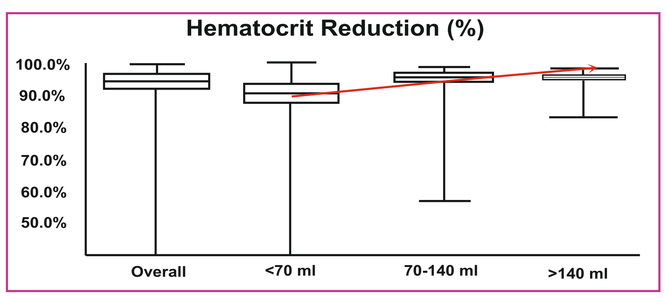
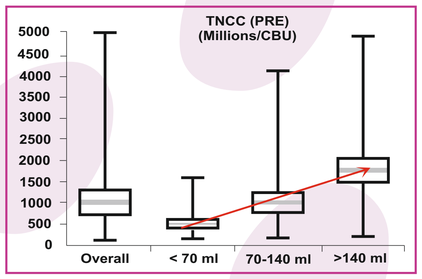
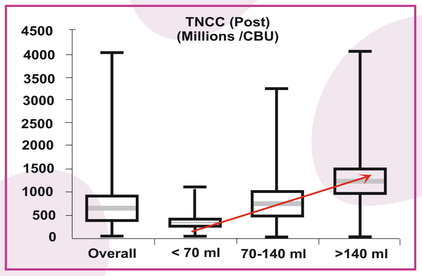


TABLE 2: TNCC

		Pre-TNCC (millions/CBU)		Post-TNCC (millions/CBU)		TNCC Recovery (%)	
	n	Median	Mean ± SD	Median	Mean ± SD	Median	Mean
Overall	36489	906	985 ± 493	668	729 ± 373	78.0%	77.6%±8.0
< 70 ml	6469	442	467 ±187	325	346 ± 145	77.9%	77.5%±7.6
70 - 140 ml	27231	965	1026 ± 394	712	763 ± 309	78.2%	77.8%±8.1
> 140 ml	2789	1706	1785 ± 552	1229	1285 ± 420	75.7%	75.5%±8.1

GRAPH 2.1





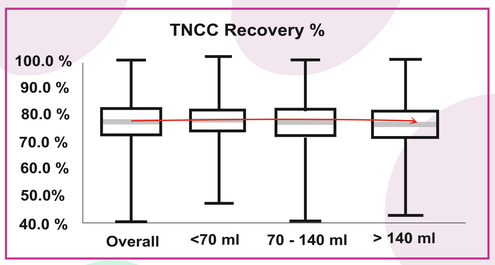
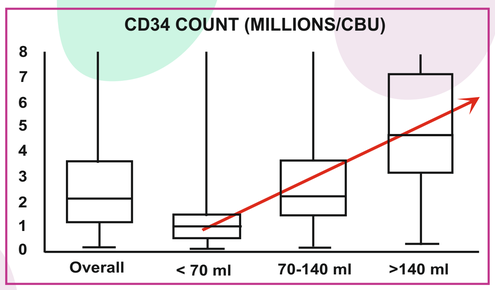
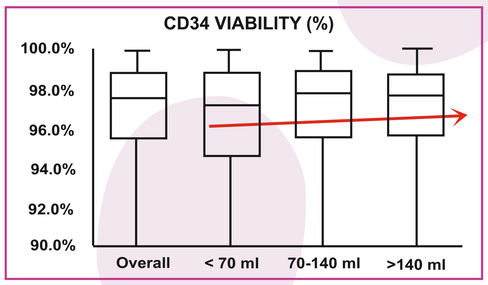


TABLE 3: CD34 COUNT & VIABILITY %

		Total CD34 Count (millions/CBU) Post processing		CD34 Viability %	
	n	Median	Mean	Median	Mean
Overall	36489	2.05	2.74 ±2.4	97.7%	96.6% ± 3.3
< 70 ml	6469	0.87	1.12 ±0.9	97.2%	96.0% ± 4.0
70 - 140 ml	27231	2.24	2.83 ±2.2	97.8%	96.8% ± 3.2
> 140 ml	2789	4.72	5.64 ±3.5	97.7%	96.8% ± 3.0

GRAPH 3.1





CONCLUSION

- Differential gradient, Double centrifugation HES processing in closed triple bag system using sample specific approach yields highly reproducible and equivalent nucleated cell recoveries with substantially reduced hematocrit providing high quality HSC product for long-term storage and transplantation but at substantially lower costs.