

Scale-up and comparability study of T-cell depletion in single inlet – single outlet QMS

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Pre-clinical scale T-cell depletion (2×10^8 processed cells) using a single inlet – single outlet, pump controlled quadrupole magnetic flow cell sorter (QMS) was under investigation to show reproducibility of published results in a process scale-up. Buffy coats from haemochromatosis patients were spiked with cultured Kg-1a cells to mimic peripheral progenitor cells. The target T-cell fraction was immunomagnetically labeled in a one or two step labeling protocol without washing and depleted by negative selection at sorting speeds of up to 3.3×10^6 cells per second. A mean \log_{10} T-cell depletion of 2.9 (range 2.13 to 3.74, n=4) for a one-step labeling strategy and a mean \log_{10} T-cell depletion of 3.1 (range 2.58 to 3.59, n=2) for a two-step labeling strategy showed a reasonable agreement with published data describing $1/10$ as many cells processed compared to this study. The depletion experiments show high total cell recovery ($92.7 \pm 7.7\%$) and high viability ($95.0 \pm 3.8\%$) and recovery of the spiked cell fraction ($81.7 \pm 14.7\%$). The study also revealed challenges and limitations for a scale-up into clinical scale T-cell depletions ($> 1 \times 10^9 - 1 \times 10^{10}$). The current goal is a 4 \log_{10} T-cell depletion. The composition of different cell fractions for an ideal depletion is shown in the Figure.

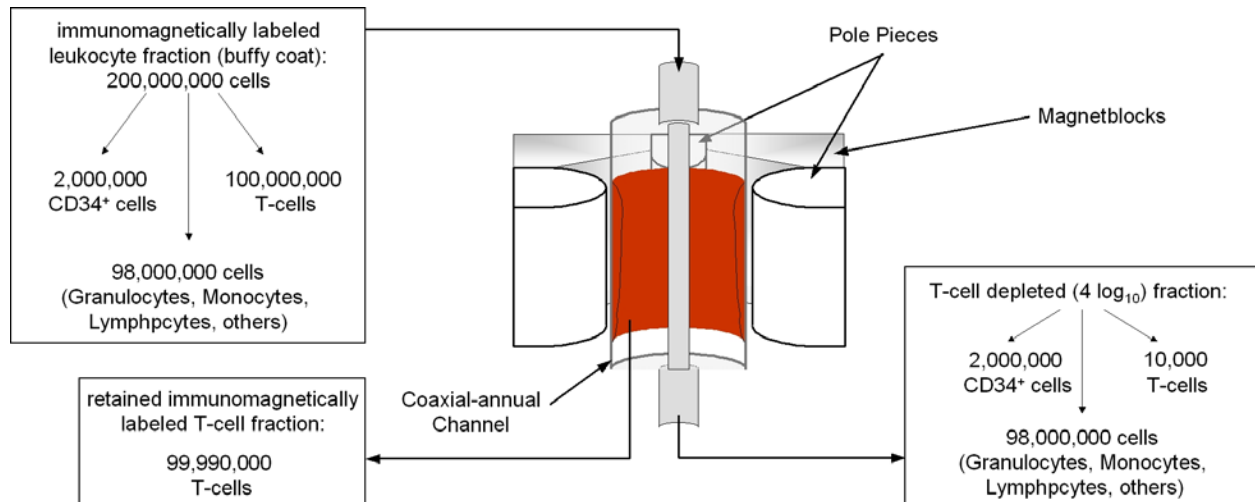


Figure. Idealized 4 \log_{10} T-cell depletion by QMS.