

Column-Free T cell Isolation using 50nm Superparamagnetic Beads on MARS® Bar

INTRODUCTION

The therapeutic efficacy of CAR T cells has been firmly established in hematologic malignancies and the key is to develop a low-cost manufacturing process to enable the wide adoption of the therapy. T cell isolation being upstream process for CAR T cell manufacturing has significant impact on the quality of the T cells and the success of the CAR T cell therapy. New technology and products are demanded for high performance T cell isolation.

Conventional micron-sized beads are known for their low efficiency and the need for a de-beading step. In contrast, 50nm superparamagnetic particles offer superior efficiency and eliminate the requirement for removal from cells in downstream applications. Consequently, commercially available 50nm magnetic beads have gained significant traction in clinical settings. However, existing options typically require the use of columns for efficient separation. Here, we introduce T cell isolation utilizing Applied Cells Ingenuity™ 50nm beads with our proprietary column-free technology integrated into the closed automated system MARS® Bar.

INPUT SAMPLE: PBMC

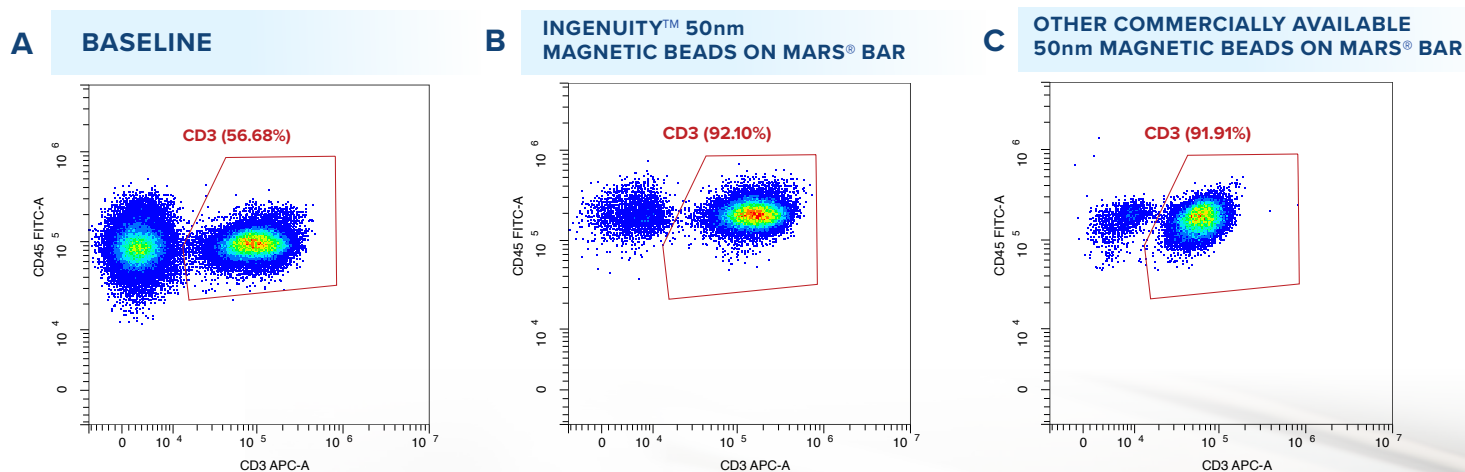


Figure 1. Purity of human T cells isolated using the MARS® Bar platform. (A) PBMC prior to separation, (B) T cells enriched using Ingenuity™ 50nm beads, and (C) T cells enriched using other commercially available 50nm beads. Figures 1(B) and 1(C) reflect the median performance shown in Figures 2(A) and 2(C), respectively.



INPUT SAMPLE: PBMC

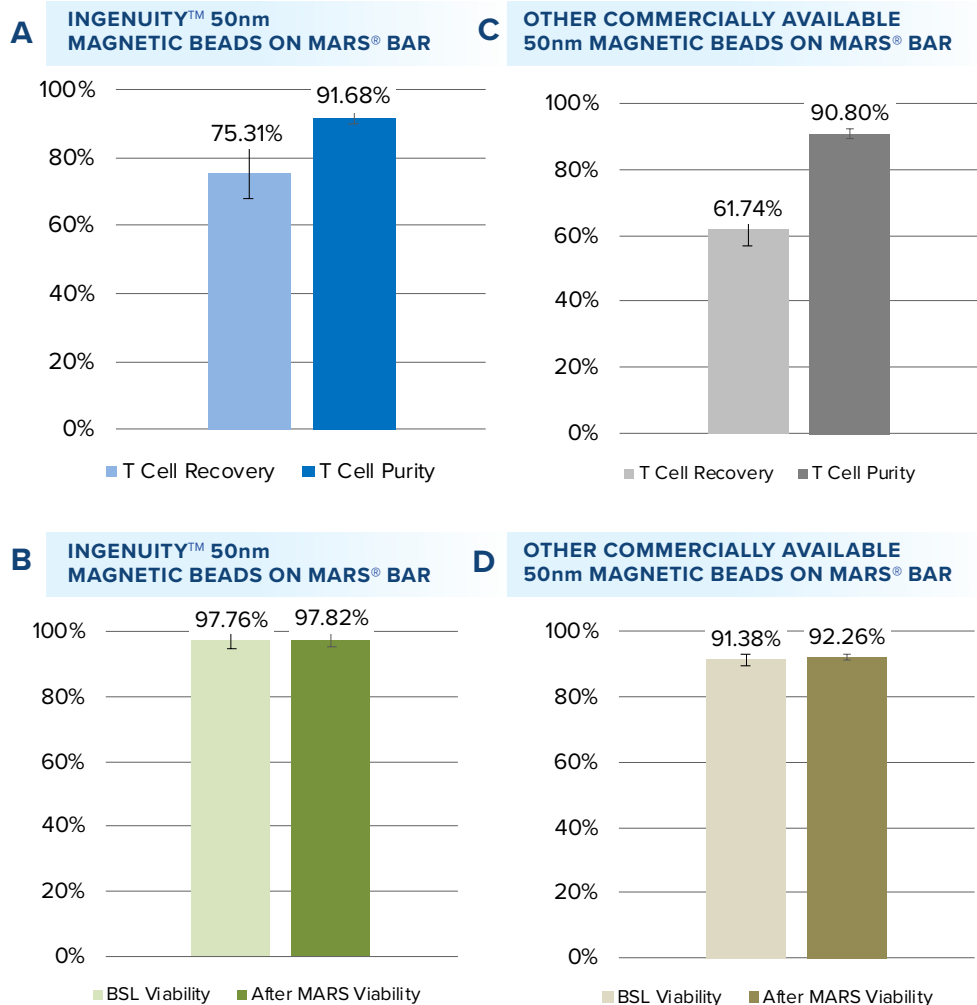


Figure 2. Efficient isolation of T cells from PBMC using the MARS® Bar platform, with no compromise in viability. (A) T cell positive selection using Ingenuity™ 50nm CD4 and CD8 beads on MARS® Bar; average recovery of 75.31% T cells with an average purity of 91.68% from an initial average T cell purity of 49.92%. n= 12 PBMC samples. (B) Viability was not significantly affected before and after positive selection with Ingenuity™ 50nm CD4 and CD8 beads on MARS® Bar (P<0.5). (C) T cell positive selection using other commercially available 50nm CD4 and CD8 magnetic beads on MARS® Bar; average recovery of 61.74% T cells with an average purity of 90.80% from an initial average T cell purity of 47.95%. n= 5 PBMC samples. (D) Viability was not significantly affected before and after positive selection with other commercially available 50nm CD4 and CD8 beads on MARS® Bar (P<0.5).

Applied Cells Ingenuity™ Reagents

- 50nm size superparamagnetic beads
- High safety, biodegradable particles
- High stability, excellent separation efficiency of target cells
- Provide RUO and GMP grade products

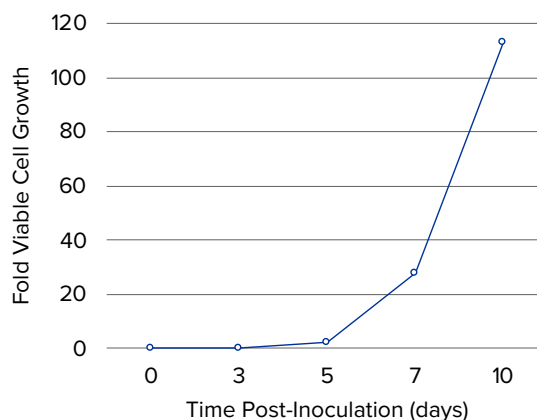


Figure 3. Expansion of T cells enriched by Ingenuity™ 50nm beads with MARS® Bar. The enriched T cells expanded robustly, growing over 112-fold by day 10 post-enrichment in cell culture plates using T cell expansion medium with CD3/CD28 activator.

INPUT SAMPLE: WHOLE PERIPHERAL BLOOD

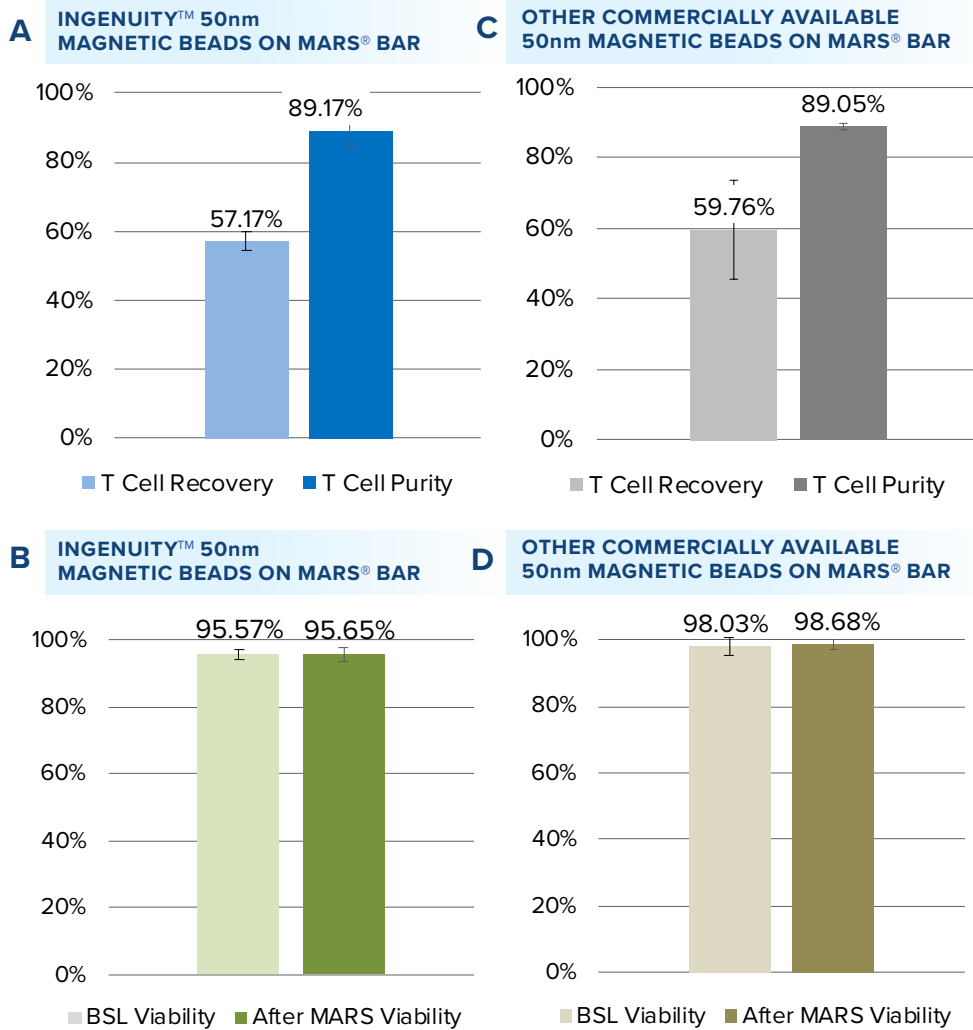


Figure 4. Isolation of T cells from whole blood using the MARS® Bar platform, with no compromise in viability. (A) T cell positive selection using Ingenuity™ 50nm CD4 and CD8 beads on MARS® Bar platform; average recovery of 57.17% T cells with an average purity of 89.17% from an initial average T cell purity of 21.73%. n= 3 healthy donor samples. (B) Viability was not significantly affected before and after positive selection with Ingenuity 50nm CD4 and CD8 beads on MARS® Bar (P<0.5). (C) T cell positive selection using other commercially available 50nm CD4 and CD8 beads on MARS® Bar; average recovery of 59.76% T cells with an average purity of 89.05% from initial average T cell purity of 25.67%. n= 3 healthy donor samples. (D) Viability was not significantly affected before and after positive selection with other commercially available 50nm CD4 and CD8 beads on MARS® Bar (P<0.5).

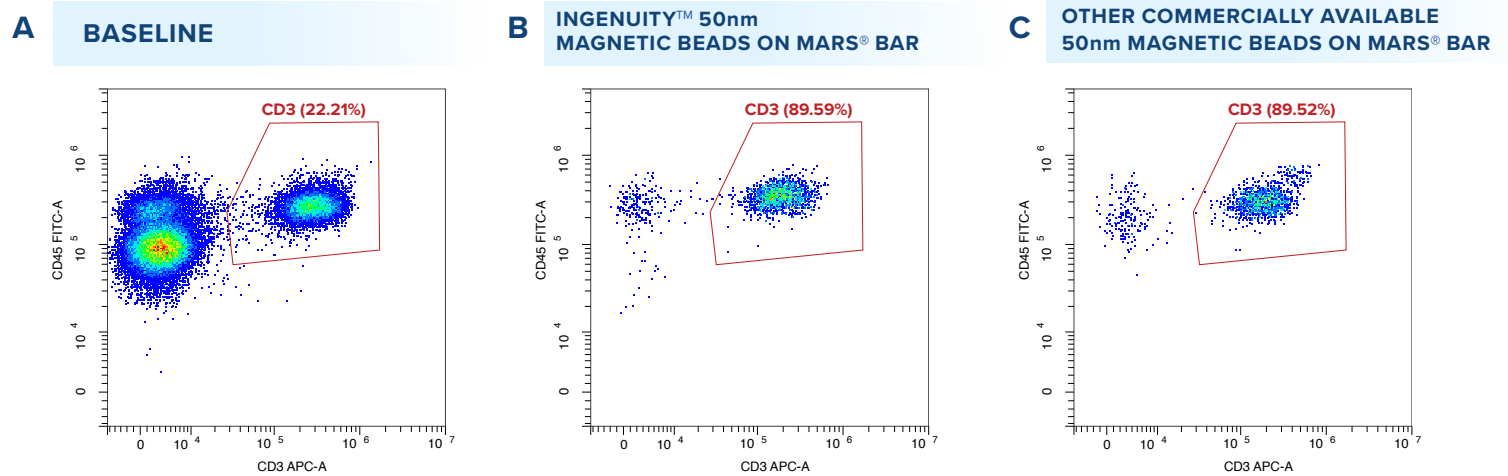


Figure 5. Purity of human T cells isolated by the MARS® Bar platform. (A) Whole blood prior to separation, (B) Enriched T cells using Ingenuity™ 50nm beads, and (C) Enriched T cells using other commercially available 50nm beads. Figures 5(B) and 5(C) represent the median performances of Figures 4(A) and 4(C), respectively.

About MARS® Bar

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MARS® Bar Cell Separation Platform

The MARS® Bar Magnetic Separation Platform is a closed and automated isolation for cell therapy development and manufacturing. Together with fit-for-purpose consumables, it delivers high cell purity, recovery, and viability.

Designed to be used with magnetic beads, MARS® Bar Magnetic Separation Platform uses column-free technology performs in-flow separation at high flow rate to achieve exceptional cell recovery, purity, a wide range of reaction volumes while maintaining cell viability. Together with process flexibility, sterile single-use consumables, and software with 21 CFR Part 11 compliance, this system is designed to help you easily scale from development to clinical and commercial manufacturing.

FLEXIBLE PROTOCOLS
BROAD RANGE OF VOLUMES AND SAMPLE TYPES
FAST AND EASY PROTOCOL OPTIMIZATION

FULLY ENCLOSED
AUTOMATED
ECONOMICAL

MARS® Bar Flex



MARS® Bar



MARS® Bar
ISO13485



Discovery

RUO



Development

RUO



Manufacturing

GMP



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