

Ready·Sep·Go

Fast and Easy Isolation of T Cells



 **STEMCELL**[™]
TECHNOLOGIES

Ready·Sep·Go

Isolate T Cells In As Little As 25 Minutes

Isolate whole T cell populations as well as various T cell subsets with high purity and recovery using the fast and easy T cell isolation products from STEMCELL Technologies. Isolate T cells directly from virtually any sample source, including human whole blood, buffy coat, leukapheresis samples, or fresh or previously frozen peripheral blood mononuclear cells (pages 7 - 9), and from mouse spleen or other tissues (pages 10 - 11).



+EasySep™

Fast and Easy Immunomagnetic Cell Separation (page 4)

EasySep™ isolates T cells quickly and easily without the use of columns in as little as 25 minutes. With a simple pour, highly purified functional cells are immediately ready for downstream use.



⊕RoboSep™

Fully Automated Immunomagnetic Cell Separation (page 4)

RoboSep™ fully automates all T cell labeling and separation steps of the EasySep™ procedure, minimizing sample handling and freeing up technician time.



⊕RosetteSep™

Unique Immunodensity Cell Separation (page 5)

RosetteSep™ isolates highly purified T cells directly from human whole blood during density gradient centrifugation, reducing your T cell isolation workflow to a single step.

FAST AND EASY. T cells are ready in as little as 25 minutes with no columns.

VERSATILE. Isolate T cells directly from virtually any sample source, including whole blood and leukapheresis samples.

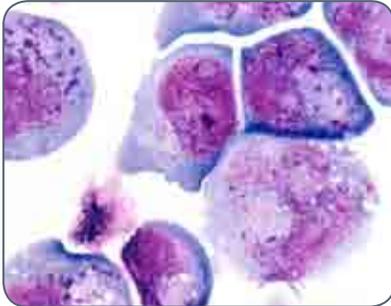
GENTLE. Column-free cell separation yields highly purified, viable T cells that are immediately ready for functional and biological studies.

FRONT COVER: Immunofluorescent light micrograph of cultured T cells from a patient with large granular lymphocytic leukemia. Nancy Kedersha/Science Photo Library

Research and Applications

Fast and Easy T Cell Separation Solutions

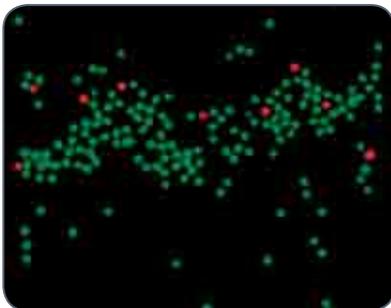
The fast and easy column-free cell separation platforms **EasySep™**, **RoboSep™** and **RosetteSep™** are gentle on cells to preserve T cell viability, ensuring that isolated T cells are suitable for downstream functional and biological studies. Negatively enriched T cells are unlabeled by EasySep™ reagents, while positively selected T cells are labeled with small EasySep™ magnetic particles that are flow cytometry-compatible.



Infectious Disease and Vaccine Research

Isolated T cells are highly functional and have been used to study:

- The effect of HIV infection on T cell expansion¹ and response^{2,3}
- T cell stimulation for adoptive immunotherapy⁴
- The role of T cells in vaccine-mediated protection⁵



HLA and Chimerism Analysis

The HLA T cell isolation products are optimized for HLA assays and are suitable for:

- Serology-based assays
- Flow Cytometry Crossmatch (FCXM)
- Chimerism analysis



Assay Development

Large volumes of highly purified T cells can be isolated directly from leukapheresis samples and are ideal for:

- Drug development
- Immunoassay development
- Vaccine development and testing
- Companion diagnostics assay development

Want to know more?

To learn more about how STEMCELL Technologies supports T cell research and read the latest publications, please visit us at www.stemcell.com/tcells.



Fast and Easy Immunomagnetic T Cell Isolation

EasySep™ is a quick and easy way to get highly purified T cells from whole blood, PBMCs, buffy coat, leukapheresis samples, and mouse spleen or other tissue samples. Cells are cross-linked to EasySep™ magnetic particles using the Tetrameric Antibody Complex (TAC) technology, and then separated from unwanted cells with an EasySep™ magnet. Isolated T cells are immediately ready for downstream applications.

EasySep™ Human CD4⁺ T Cell Enrichment Procedure

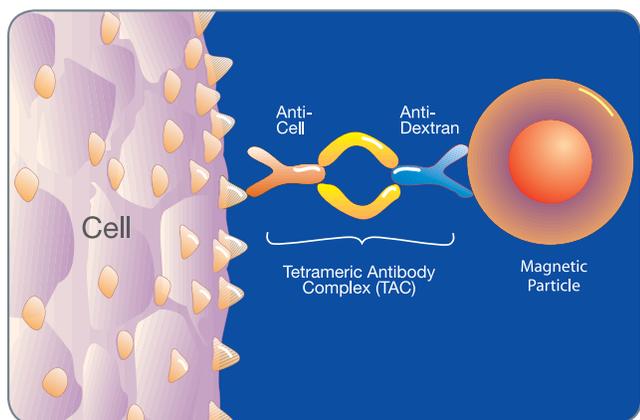
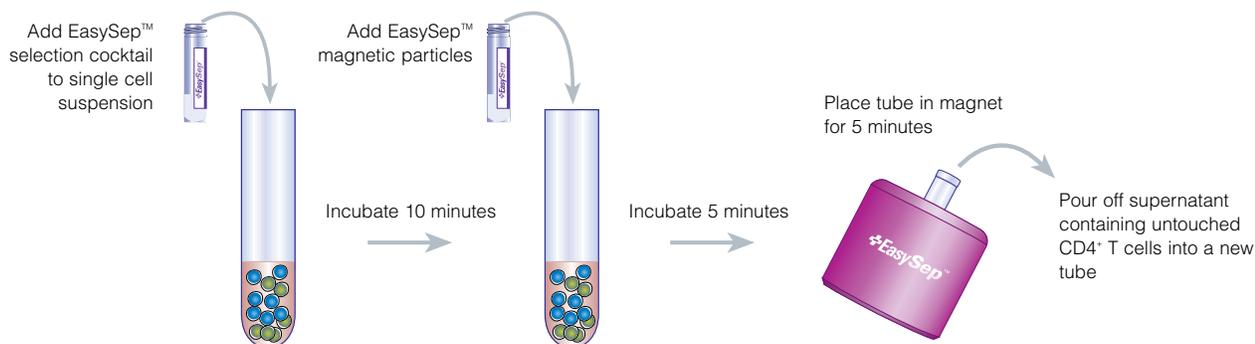
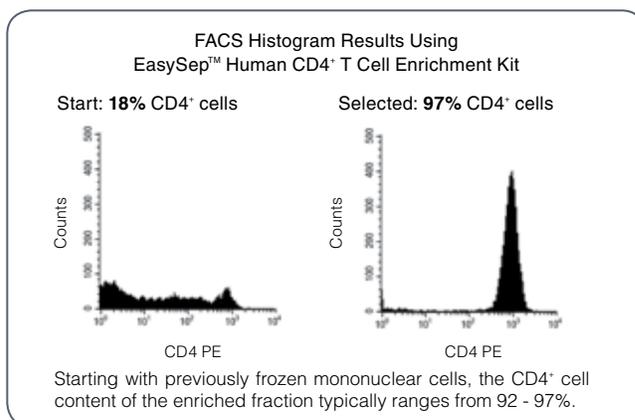


FIGURE 1. Tetrameric Antibody Complex (TAC) crosslinking a cell to a dextran-coated magnetic particle.



Fully Automated T Cell Isolation



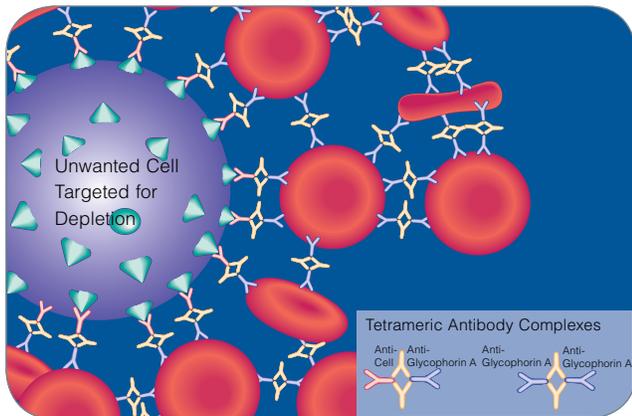
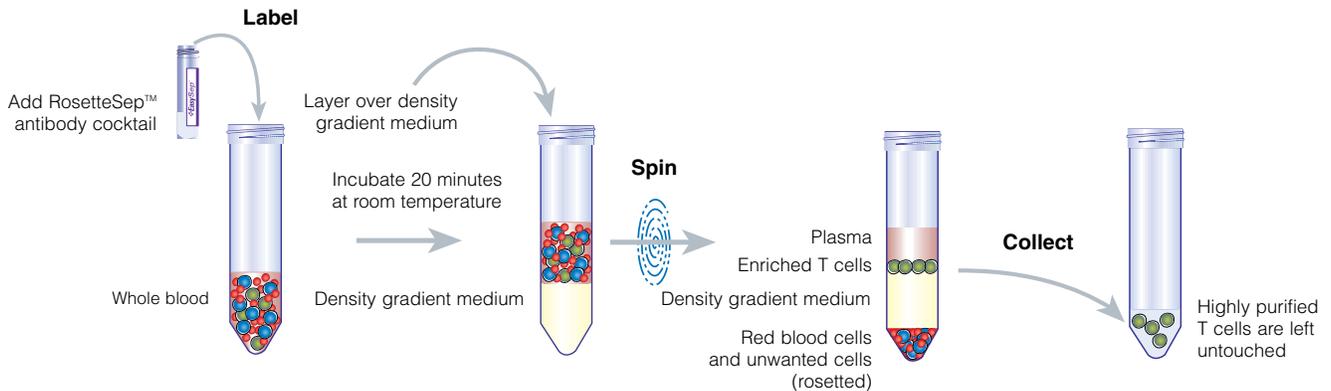
Streamline your T cell separations using RoboSep™, the fully automated cell separator. By performing all EasySep™ cell labeling and separation steps, RoboSep™ maintains the speed and simplicity of EasySep™ while offering walk-away automation, enabling the high-throughput, versatile isolation of highly purified cells. Minimize sample handling and eliminate cross-contamination while isolating T cells with just 5 minutes of “hands-on” technician time.



T Cell Isolation Directly from Whole Blood with a Simple Spin

RosetteSep™ is a fast and easy immunodensity procedure for the isolation of untouched T cells directly from whole blood. By crosslinking unwanted cells to red blood cells present in the sample, RosetteSep™ eliminates the need for a separate magnetic separation step because cells are purified during standard density gradient centrifugation. This approach significantly reduces handling time and maximizes convenience.

RosetteSep™ Human T Cell Enrichment Procedure



FAST AND EASY. Column-free T cell isolation during standard density centrifugation.

UNTOUCHED CELLS. T cells are unlabeled and ready for immediate use.

FLEXIBLE AND SAFE. Process small or large volumes and multiple samples simultaneously with minimal sample handling.



Video:
RosetteSep™ Introduction
www.stemcell.com/RosetteSepVideo

Equipment

+EasySep™



EasySep™ Magnet

The EasySep™ Magnet is designed to hold one 5 mL polystyrene tube to isolate up to 2.5×10^8 cells (or up to 5×10^8 cells when isolating rare cells [e.g. CD34⁺]) per separation. Catalog #18000



The Big Easy EasySep™ Magnet

The Big Easy EasySep™ Magnet is designed to hold one 14 mL polystyrene tube to isolate up to 10^9 cells (or up to 2×10^9 cells when isolating rare cells [e.g. CD34⁺]) per separation. Catalog #18001



Easy 50 EasySep™ Magnet

The Easy 50 EasySep™ Magnet is designed to hold one 50 mL conical tube to isolate up to 4×10^9 cells by negative selection from whole blood, leukapheresis products or mouse splenocytes (or 2×10^9 cells from PBMC) per separation. Catalog #18002



EasyPlate™ EasySep™ Magnet

The EasyPlate™ EasySep™ Magnet, which is designed to hold a standard 96-well plate, enables high-throughput cell isolation from small sample sizes. Catalog #18102



EasySep™ Multistand

Separate up to 4 samples at once using the EasySep™ Multistand. The multistand is designed to hold up to 4 EasySep™ Magnets or 4 The Big Easy EasySep™ Magnets. Catalog #18010

RoboSep™

System is Supplied With:

- RoboSep™ Carousel
- Hydraulic Fluid Bottle
- One-Year Warranty
- RoboSep™ Accessory Kit
 - 4 "The Big Easy" EasySep™ Magnets
 - 2 Boxes of RoboSep™ Filter Tip Racks (16 Filter Tip Racks in total)
 - RoboSep™ Buffer (250 mL)
 - RoboSep™ Service Rack
 - USB Keyboard, Mouse, Memory Stick
 - Magnet Shields
 - Tip Head Polishing Compound
 - User Manual
 - RoboSep™ Timer
 - Quick Start Sheet

RoboSep™ & Accessories

PRODUCT NAME	CATALOG #
RoboSep™	20000
RoboSep™ Warranty	20200
RoboSep™ Warranty with Preventative Maintenance	20202
RoboSep™ Preventative Maintenance (No Warranty)	20203
RoboSep™ Preventative Maintenance (Active Warranty)	20209
RoboSep™ Service Rack	20101
Hydraulic Fluid Bottle	20102
RoboSep™ Buffer (250 mL) ¹	20104
RoboSep™ Buffer 5X Concentrate (250 mL)	20124
RoboSep™ Filter Tip Racks (1 box of 8 racks) ¹	20125
EasySep™ RBC Lysis Buffer 10X Concentrate (100 mL) ²	20120
RoboSep™ Tip Head Polishing Compound (7 mL)	20119

1. RoboSep™ Buffer and 1 - 2 boxes of RoboSep™ Filter Tip Racks are included with every purchase of a RoboSep™ Reagent Kit.
2. 10 mL of 10X RBC Lysis Buffer is included with every purchase of an EasySep™ or RoboSep™ Whole Blood Selection Kit (see pages 8 - 9).

Technical Specifications:

Dimensions:

- Height with lid: 56 cm (21 7/8")
- Width: 71 cm (27 7/8")
- Depth: 39 cm (15 3/8")
- Weight: 26 kg (57 lb)

Power Requirements:

- 50/60 Hz, AC 100-240V
- Connections: RJ-45 10/100 Ethernet port, 2 USB ports

Conditions for Operation:

- Temperature: 10 - 30°C (50 - 86°F). RoboSep™ is not specified for use in a cold room (4°C, 39°F)
- Humidity 20 - 85% (non-condensing)

Human T Cell Isolation Kits

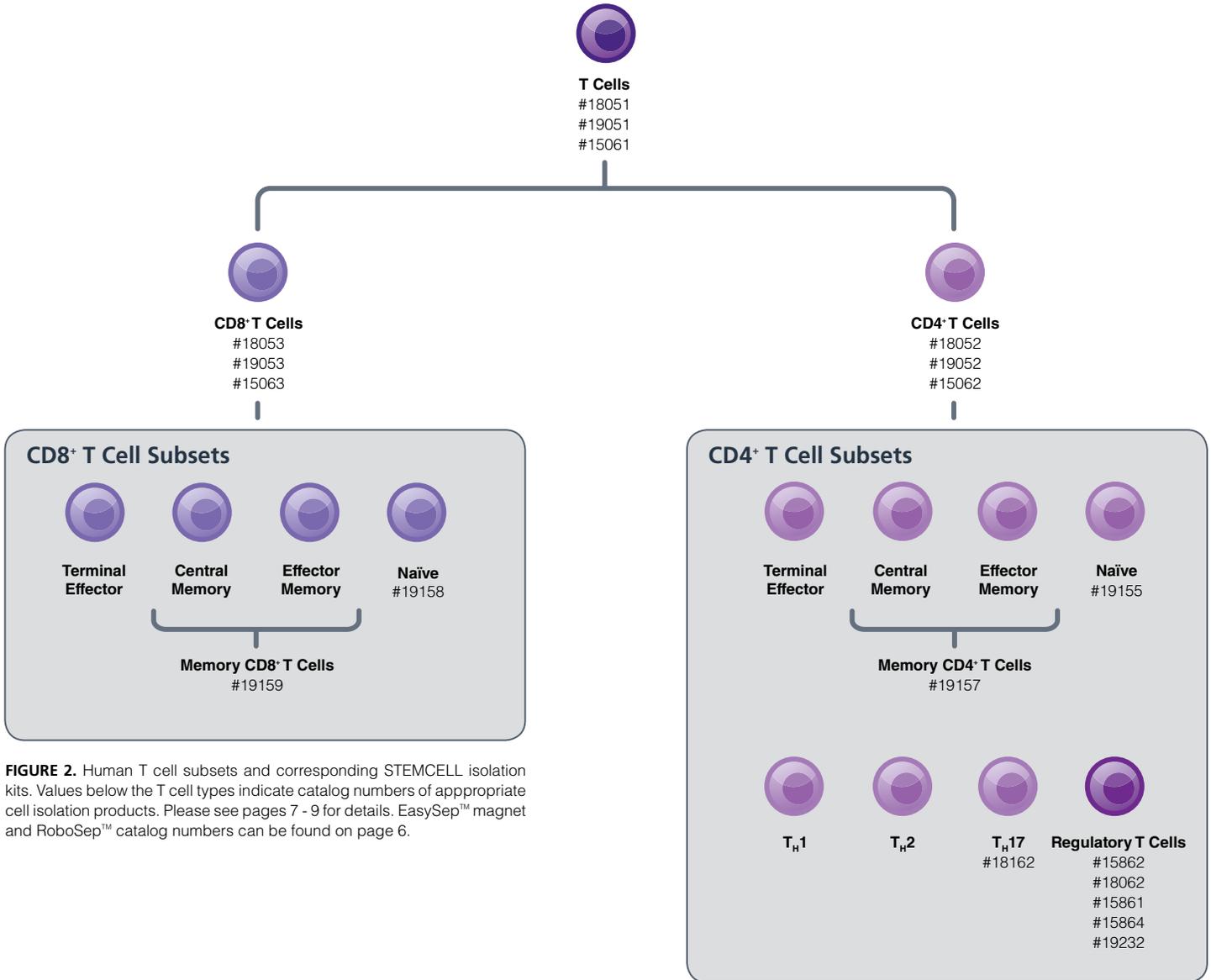


FIGURE 2. Human T cell subsets and corresponding STEMCELL isolation kits. Values below the T cell types indicate catalog numbers of appropriate cell isolation products. Please see pages 7 - 9 for details. EasySep™ magnet and RoboSep™ catalog numbers can be found on page 6.

RosetteSep™ Human T Cell Products

PHENOTYPE	TISSUE ¹	PRODUCT	TYPICAL PERFORMANCE DATA	
			% PURITY	CATALOG #
T Cells	WB, BC	RosetteSep™ Human T Cell Enrichment Cocktail	90 - 97	15021/15061
	WB, BC	RosetteSep™ HLA T Cell Enrichment Cocktail	90 - 97	15061HLA/ 15081HLA
CD4 ⁺	WB, BC	RosetteSep™ Human CD4 ⁺ T Cell Enrichment Cocktail	89 - 99	15022/15062
CD8 ⁺	WB, BC	RosetteSep™ Human CD8 ⁺ T Cell Enrichment Cocktail	81 - 95	15023/15063
CD3 ⁻ -Depleted	WB, BC	RosetteSep™ Human CD3 Depletion Cocktail	3.0 Log Depletion	15621/15661
CD4 ⁻ -Depleted	WB, BC	RosetteSep™ Human CD4 Depletion Cocktail	2.0 Log Depletion	15622/15662
CD8 ⁻ -Depleted	WB, BC	RosetteSep™ Human CD8 Depletion Cocktail	2.0 Log Depletion	15623/15663

Each cocktail contains enough reagents to label either 40 mL or 200 mL of whole blood, except the HLA cocktails, which can label 250 mL or 1000 mL of whole blood. Depletion cocktails can be added to a standard RosetteSep™ cocktail, if not already present.

1. WB - whole blood; BC - buffy coat.

Human T Cell Negative Selection

PHENOTYPE	TISSUE ¹	PRODUCT	TYPICAL PERFORMANCE DATA	CATALOG # ³
			% PURITY ²	
T Cells	PBMC	EasySep™/RoboSep™ Human T Cell Enrichment Kit	95 - 99	19051/19051RF
CD4 ⁺	PBMC	EasySep™/RoboSep™ Human CD4 ⁺ T Cell Enrichment Kit	92 - 97	19052/19052RF
Naïve CD4 ⁺	PBMC	EasySep™/RoboSep™ Human Naïve CD4 ⁺ T Cell Enrichment Kit	91 - 95	19155/19155RF
Memory CD4 ⁺	PBMC	EasySep™/RoboSep™ Human Memory CD4 ⁺ T Cell Enrichment Kit	86 - 98	19157/19157RF
CD8 ⁺	PBMC	EasySep™/RoboSep™ Human CD8 ⁺ T Cell Enrichment Kit	84 - 89	19053/19053RF
Naïve CD8 ⁺	PBMC	EasySep™/RoboSep™ Human Naïve CD8 ⁺ T Cell Enrichment Kit	85 - 92	19158/19158RF
Memory CD8 ⁺	PBMC	EasySep™/RoboSep™ Human Memory CD8 ⁺ T Cell Enrichment Kit	72 - 92	19159/19159RF
Isolate any human cell type by negative selection		EasySep™/RoboSep™ Human Custom Enrichment Kit	-	19309/19309RF

Human T Cell Positive Selection

PHENOTYPE	TISSUE ¹	PRODUCT	TYPICAL PERFORMANCE DATA	CATALOG # ³
			% PURITY ²	
CD2 ⁺	PBMC	EasySep™/RoboSep™ Human CD2 Positive Selection Kit	86 - 98	18657/18657RF
	WB	EasySep™/RoboSep™ Human Whole Blood CD2 Positive Selection Kit	96 - 100	18687/18687RF
CD3 ⁺	PBMC	EasySep™/RoboSep™ Human CD3 Positive Selection Kit	99 - 100	18051/18051RF
	WB	EasySep™/RoboSep™ Whole Blood CD3 Positive Selection Kit	98 - 100	18081/18081RF
CD4 ⁺	PBMC	EasySep™/RoboSep™ Human CD4 Positive Selection Kit	97 - 100	18052/18052RF
	WB	EasySep™/RoboSep™ Whole Blood CD4 Positive Selection Kit	97 - 100	18082/18082RF
Th17	PBMC	EasySep™/RoboSep™ Human Th17 Cell Enrichment Kit	85 - 94	18162/18162RF
CD8 ⁺	PBMC	EasySep™/RoboSep™ Human CD8 Positive Selection Kit	94 - 100	18053/18053RF
	WB	EasySep™/RoboSep™ Whole Blood CD8 Positive Selection Kit	97 - 100	18083/18083RF
CD25 ⁺	PBMC	EasySep™/RoboSep™ Human Pan-CD25 Positive Selection and Depletion Kit	90 - 96	18251/18251RF
Isolate any human cell type by positive selection		EasySep™/RoboSep™ Human Custom Selection Kit	-	18309/18309RF

Each kit contains enough reagents to select desired cells from 10⁹ total cells. Kits contain Fc receptor blocker when necessary to prevent non-specific binding.

1. PBMC - peripheral blood mononuclear cells; WB - whole blood; BC - buffy coat.

2. Combined purity data of EasySep™ and RoboSep™ kits (when available).

3. RoboSep™ Reagent Kits (RF) contain an EasySep™ Selection Kit with 1-2 boxes of RoboSep™ Filter Tip Racks and a 250 mL bottle of RoboSep™ Buffer.

Human Regulatory T Cell Isolation

PHENOTYPE	TISSUE ¹	PRODUCT	TYPICAL PERFORMANCE DATA	
			% PURITY ²	CATALOG # ³
CD4 ⁺ CD25 ⁺	PBMC	EasySep™/RoboSep™ Human CD4 ⁺ CD25 ⁺ T Cell Isolation Kit	61 - 83	18062/18062RF
	WB, BC	Complete Kit for Human CD4 ⁺ CD25 ⁺ T Cells	89 - 99	15862/15862RF
CD4 ⁺ CD127 ^{low}	PBMC	EasySep™/RoboSep™ Human CD4 ⁺ CD127 ^{low} T Cell Enrichment Kit	92 - 96	19231/19231RF
CD4 ⁺ CD127 ^{low} CD25 ⁺	WB, BC	Complete Kit for Human CD4 ⁺ CD127 ^{low} CD25 ⁺ Regulatory T Cells	75 - 98 (65 - 91 CD25 ⁺ FOXP3 ⁺)	15861/15861RF
CD4 ⁺ CD127 ^{low} CD49d ⁺ CD25 ⁺	WB, BC	Complete Kit for Human CD4 ⁺ CD127 ^{low} CD49d ⁺ CD25 ⁺ Regulatory T Cells	79 - 99 (67 - 93 CD25 ⁺ FOXP3 ⁺)	15864/15864RF
CD4 ⁺ CD127 ^{low} CD49 ⁺	PBMC	EasySep™/RoboSep™ Human CD4 ⁺ CD127 ^{low} CD49d ⁺ Regulatory T Cell Enrichment Kit	57 - 87 CD25 ⁺ FOXP3 ⁺	19232/19232RF
CD25 ^{high}	CD4 ⁺ T Cells ⁴	EasySep™ Human CD25 Positive Selection Kit	90 - 98	18231

T Cell Kits for HLA Analysis

PHENOTYPE	TISSUE ¹	PRODUCT	TYPICAL PERFORMANCE DATA	
			% PURITY ²	CATALOG # ³
T Cells	PBMC	EasySep™/RoboSep™ HLA T Cell Enrichment Kit	95 - 99	19051HLA/ 19051HLARF
	WB	EasySep™/RoboSep™ HLA Whole Blood T Cell Enrichment Kit	93 - 98	19951HLA/ 19951HLARF
CD2 ⁺	PBMC	EasySep™/RoboSep™ HLA CD2 Positive Selection Kit	86 - 98	18657HLA/ 18657HLARF
	WB	EasySep™/RoboSep™ HLA Whole Blood CD2 Positive Selection Kit	96 - 100	18687HLA/ 18687HLARF
CD3 ⁺	PBMC	EasySep™/RoboSep™ HLA CD3 Positive Selection Kit	95 - 100	18051HLA/ 18051HLARF
	WB	EasySep™/RoboSep™ HLA Whole Blood CD3 Positive Selection Kit	93 - 99	18081HLA/ 18081HLARF

Each kit for PBMC contains enough reagents to select desired cells from 10⁹ total cells, except the CD4⁺CD127^{low}CD49d⁺ Regulatory T Cell Enrichment Kit, which can process up to 2 x 10⁸ cells. Each whole blood kit contains enough reagents to label 200 mL of whole blood, except the HLA CD2 and CD3 Positive Selection Kits, which can label 60 mL of whole blood.

1. WB - whole blood; PBMC - peripheral blood mononuclear cells; BC - buffy coat.
2. Combined manual EasySep™ and RoboSep™ purity data (when available).
3. RoboSep™ Reagent Kits (RF) contain an EasySep™ Selection Kit with RoboSep™ Buffer and 1-2 boxes of RoboSep™ Filter Tip Racks.
4. RosetteSep™ enriched (catalog #15062).

Mouse T Cell Isolation Kits

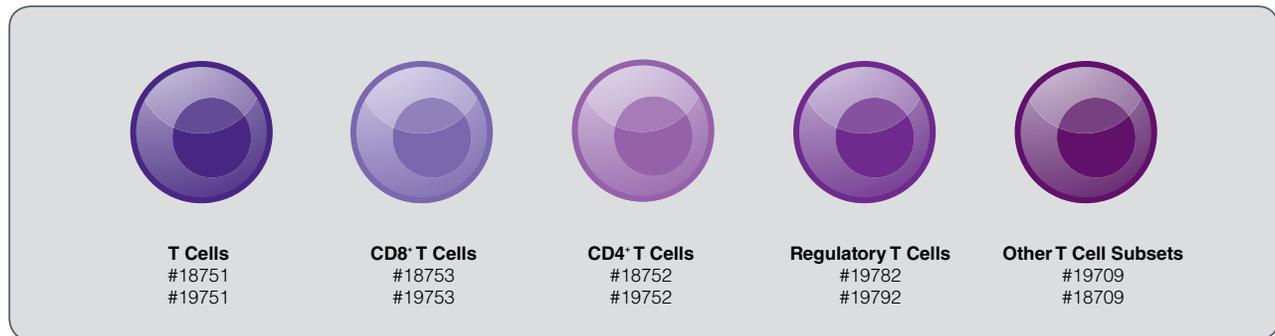


FIGURE 3. Mouse T cell subsets and corresponding STEMCELL isolation kits. Values below each cell type indicate catalog numbers of appropriate isolation kits. Please see page 6 for EasySep™ magnet and RoboSep™ catalog numbers.

Mouse T Cell Negative Selection

PHENOTYPE	TISSUE	PRODUCT	TYPICAL PERFORMANCE DATA	
			CATALOG # ²	
			% PURITY ¹	
T cells	Spleen	EasySep™/RoboSep™ Mouse T Cell Enrichment Kit	95 - 99	
CD4 ⁺	Spleen	EasySep™/RoboSep™ Mouse CD4 ⁺ T Cell Enrichment Kit	89 - 96	
CD8 ⁺	Spleen	EasySep™/RoboSep™ Mouse CD8 ⁺ T Cell Enrichment Kit	86 - 94	
Isolate any mouse cell type by negative selection		EasySep™ Mouse Custom Enrichment Kit	-	

Mouse T Cell Positive Selection

PHENOTYPE	TISSUE	PRODUCT	TYPICAL PERFORMANCE DATA	
			CATALOG # ²	
			% PURITY ¹	
CD4 ⁺	Spleen	EasySep™/RoboSep™ Mouse CD4 Positive Selection Kit	94 - 99	
CD4 ⁺ CD25 ⁺	Spleen	EasySep™ Mouse CD4 ⁺ CD25 ⁺ Regulatory T Cell Isolation Kit	85 - 97	
CD8a	Spleen	EasySep™/RoboSep™ Mouse CD8a Positive Selection Kit	90 - 98	
CD90.2 (Thy 1.2)	Spleen	EasySep™/RoboSep™ Mouse CD90.2 (Thy 1.2) Positive Selection Kit	96 - 99	
Isolate any mouse cell type by positive selection		EasySep™ Mouse Custom Selection Kit	-	

Mouse Negative Selection Kits contain enough reagents to enrich desired cells from 10⁸ total cells. Kits contain mouse Fc receptor blocker when necessary to prevent non-specific binding. Mouse Positive Selection Kits contain enough reagents to select desired cells from 2 x 10⁸ total cells, with the exception of the CD4⁺CD25⁺ Regulatory T Cell Kit, which contains enough reagents to select desired cells from 10⁸ total cells.

1. Combined purity data of EasySep™ and RoboSep™ kits (when available).
2. RoboSep™ Reagent Kits (RF) contain an EasySep™ Selection Kit with 1-2 boxes of RoboSep™ Filter Tip Racks and a 250 mL bottle of RoboSep™ Buffer.

Custom T Cell Isolation Kits

With many years of cell separation expertise, an extensive range of antibodies and a variety of cell isolation methods, STEMCELL Technologies scientists can create tailor-made products to isolate desired cell types. This is particularly useful to researchers working with rare cell types, difficult tissues, non-standard species or any other special applications.

Design Your Own Custom Kit

Choose your desired cell type or the cell surface antigens you would like to target and let STEMCELL design a kit for you.

Modify Existing Kits

Antibodies can be added or removed from any of STEMCELL's kits to isolate your desired cell type.

Use Your Own Labeled Antibody

Isolate cells using conjugated antibodies with EasySep™ PE, Biotin, FITC or APC Selection Kits.

Use Any Mouse IgG₁ Antibody

Isolate cells using any mouse IgG₁ antibody with the EasySep™ "Do-It-Yourself" Selection Kit.

Researchers Have Used Custom Cocktails to Isolate:

- Human NKT cells
- Human memory T cells (CD3⁺/CD45RO⁺)
- Human resting memory T cells
- Mouse naïve CD4⁺ T cells (CD4⁺/CD62L⁺)
- Mouse $\gamma\delta$ T cells
- Non-human primate regulatory T cells (CD4⁺/CD25⁺)

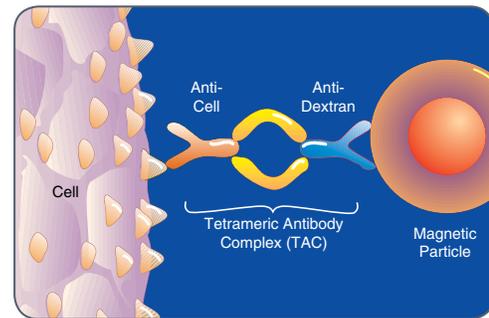


FIGURE 4. Cells are labeled with bispecific Tetrameric Antibody Complexes (TAC). Magnetically labeled cells are then separated from unlabeled cells using an EasySep™ magnet.

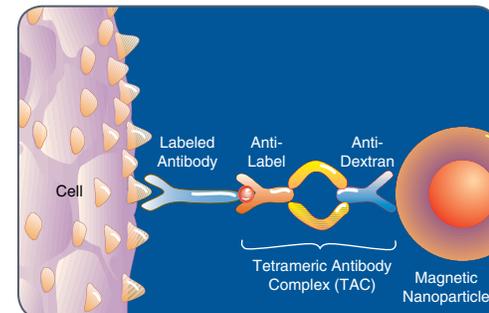


FIGURE 5. Virtually any cell type can be isolated using your own biotinylated, PE-, APC- or FITC-conjugated antibody, or any mouse IgG₁ antibody.

References

1. L. Weiss et al., Proc. Natl. Acad. Sci. U.S.A. 107, 10632 (2010)
2. A. Sáez-Cirión et al., Trends Immunol. 28, 532 (2007)
3. A. Sáez-Cirión et al., Nat. Protoc. 5, 1033 (2010)
4. L. Garidou et al., J. Virol. 83, 8905 (2009)
5. X. Guo et al., Vaccine. 29, 772 (2011)

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