



## Media for Stem Cell Biology

Unlock Extraordinary Potential with Media from Sigma-Aldrich

The life science business of Merck KGaA, Darmstadt, Germany operates as MilliporeSigma in the U.S. and Canada.



# **Optimize Your Stem Cell Expansion**

Proven consistent results and optimized formulations have made the Sigma<sup>®</sup> Stemline<sup>®</sup> Media a must-have for researchers rising to the challenges of adult stem cell expansion and maturation. Along with our broad selection of reagents, supplements, antibodies and cytokines, Stemline<sup>®</sup> Media ensures optimal expansion of robust cells.

#### **The Stemline® Media family includes:**

- Stemline<sup>®</sup> Hematopoietic Stem Cell Expansion Medium I and II
- Stemline<sup>®</sup> Neural Stem Cell Expansion Medium
- Stemline<sup>®</sup> T cell Expansion Medium
- Stemline<sup>®</sup> Mesenchymal Stem Cell Expansion Medium
- Stemline<sup>®</sup> Keratinocyte Medium II

To request more information, contact your local Sigma-Aldrich representative or visit **SigmaAldrich.com/stemcells** To inquire about volume discounts, contact your local Sigma-Aldrich representative today.



## Stemline<sup>®</sup> Hematopoietic Stem Cell Expansion Media

Developed to promote the optimal expansion of human hematopoietic stem cells (HSC) from bone marrow, mobilized peripheral blood and cord blood, Stemline<sup>®</sup> Hematopoietic Stem Cell Expansion Medium demonstrates higher total nucleated cell (TNC) fold increases than other commercially available serum-free media formulations.

The second generation of the Sigma<sup>®</sup> hematopoietic stem cell expansion media family, Stemline<sup>®</sup> II, has been developed to optimize the balance of differentiated and undifferentiated cells while maximizing their expansion. Compatible with hematopoietic stem cells from bone marrow, cord blood and mobilized peripheral blood, Stemline<sup>®</sup> II has been shown to lead to significant increases in cell expansion from all 3 sources. Through flow cytometric analysis of clinical-scale expansions, Stemline<sup>®</sup> II has also demonstrated higher capacity than other commercially available media for the expansion of CD34+/CD38+ late progenitors required for short-term engraftment. Human cord blood cells expanded in Stemline<sup>®</sup> Media demonstrate impressive self-renewal when transplanted into immunodeficient NOC/SCID mice, illustrating the Stemline<sup>®</sup> utility in a true functional trial.

Stemline<sup>®</sup> II Hematopoietic Stem Cell Expansion Medium is free of serum and all other animal-derived components with the exception of human serum albumin. This exclusion increases performance consistency and eliminates safety risks associated with potential adventitious agents.

Produced in a GMP state-of-the-art facility with a Device Master File (DMF) on file, Stemline<sup>®</sup> Hematopoietic Stem Cell Expansion Medium is an excellent choice for your HSC applications.



#### Fold Increase of Total Nucleated Cells from CD34+ Bone Marrow



Product Description

Stemline® Hematopoietic Stem Cell Expansion Medium

Stemline® II Hematopoietic Stem Cell Expansion Medium

#### **Features and Benefits**

- Serum-free formulation
- Enhanced expansion from cord blood CD34+ cells
- Expands cells from all appropriate hematopoietic lineages in a colony-forming unit
- Tested extensively in 7-day and 14-day growth assays

#### Figure 1.

Stemline<sup>®</sup> demonstrates superior expansion of bone marrow hematopoietic stem cells (HSC). To test the ability of Stemline<sup>®</sup> II Hematopoietic Stem Cell Expansion Medium to expand CD34+ HSCs, researchers at Sigma-Aldrich and the University of Kentucky designed a bench-scale expansion assay. Cells were seeded into the wells of 24-well tissue culture plates. One milliliter of medium was added to each well with the appropriate cytokines to stimulate growth (100 ng/mL each of TPO, SCF, and G-CSF). Each condition was performed in triplicate and seeded with 10,000 cells per mL in each well. Cells were counted on Day 14, and the fold increase was determined by cells final/cells initial. HSCs from bone marrow cultured in Stemline<sup>®</sup> and Stemline<sup>®</sup> II demonstrated superior expansion to those grown in other serum-free HSC media.

Size	Cat. No.
500 mL	S0189
500 mL	S0192

## Stemline<sup>®</sup> Neural Stem Cell Expansion Medium

Developed to promote optimal expansion of human neural stem cells (NSC), Stemline<sup>®</sup> Neural Stem Cell Expansion Medium demonstrates rigorous expansion of human neural stem cells in both neurosphere and monolayer cultures.

Stemline<sup>®</sup> Neural Stem Cell Expansion Medium is free of serum and all other animal components; this exclusion increases performance consistency and eliminates safety risks associated with potential adventitious agents.

Produced in a GMP state-of-the-art facility with a Device Master File (DMF) on file, Stemline<sup>®</sup> Neural Stem Cell Expansion Medium is an excellent choice for your NSC applications.

#### **Growth of Neurospheres**



#### Figure 2.

Stemline<sup>®</sup> demonstrates superior expansion of neural stem cell (NSC) neurospheres. To test the ability of Stemline<sup>®</sup> Neural Stem Cell Expansion Medium to expand human NSC neurospheres, researchers at Sigma-Aldrich and the University of Wisconsin designed a bench-scale expansion assay. Cells were prepared using the method of Svendsen et al. Spheres were grown in standard DME/F-12 medium supplemented with 20 ng/mL EGF and 1% N-2 supplement prior to splitting. Half of the spheres remained in the N-2 supplemented medium and half were placed in Stemline<sup>®</sup> Neural Stem Cell Expansion Medium (also supplemented with 20 ng/mL EGF). After several passages, overall proliferation was measured via BrdU incorporation. NSC neurospheres cultured in Stemline<sup>®</sup> demonstrated superior expansion to those grown in other serum-free NSC media.

Growth of Neurospheres Expansion of Monolayer Neural Stem Cells in 24-well Culture Plate



#### Figure 3.

Stemline<sup>®</sup> demonstrates superior expansion of monolayer neural stem cells (NSC). To test the ability of Stemline<sup>®</sup> Neural Stem Cell Expansion Medium to expand human monolayer NSCs, researchers at Sigma-Aldrich and the University of Wisconsin designed a benchscale expansion assay. Cells were grown in monolayer format by seeding the cells at 20,000 cells/cm<sup>2</sup> on poly-L-lysine coated 24-well tissue culture plates. Cells were incubated for 5 days in medium supplemented with EGF (Cat. No. E9644) and LIF (Cat. No. L5283). After several passages, overall proliferation was measured. Monolayer NSC cultured in Stemline<sup>®</sup> demonstrated superior expansion to those grown in other serum-free NSC media.



Growth of Neurospheres Neurogenesis



Figure 4.

Ordering Information Product Description Stemline<sup>®</sup> Neural Stem Cell Expansion Medium

#### **Features and Benefits**

- Serum-free formulation
- For use with neurosphere and monolayer cultures
- Cells retain differentiation capacity
- Superior expansion rates when compared to alternatives

Size	Cat. No.
500 mL	S3194

## Stemline<sup>®</sup> T Cell Expansion Medium

Developed to promote the optimal expansion of adult human T cells, Stemline<sup>®</sup> T Cell Expansion Medium demonstrates significantly greater expansion (55%) when compared to alternative media, and viability greater than 95%. Additionally, flow cytometry confirms that with Stemline<sup>®</sup>, a proper CD4/CD8 ratio is maintained. In an *ex vivo* functional assay (51Chromium Release Assay), T cells expanded in Stemline<sup>®</sup> Medium proved to be highly functional and possessed cytolytic potential greater than T cells expanded in serum-containing alternative medium (RPMI with 10% fetal bovine serum). In an *in vivo* functional assay (GvHD Induction), human T lymphocytes expanded in Stemline<sup>®</sup> Medium were injected into NOD/SCID $\beta$ 2M mice (n=12). Engraftment, perivascular infiltration, and lethal GvHD were observed by Day 15 in 100% of mice, demonstrating excellent *in vivo* expansion and functionality.

Stemline<sup>®</sup> T Cell Expansion Medium is free of serum and all other animal-derived components with the exception of human serum albumin, cholesterol and transferrin. This exclusion increases performance consistency and eliminates safety risks associated with potential adventitious agents.

Produced in a GMP state-of-the-art facility with a Device Master File (DMF) on file, Stemline<sup>®</sup> T Cell Expansion Medium is an excellent choice for your T cell applications.





#### Figure 5.

Stemline<sup>®</sup> demonstrates superior expansion of T cells. When compared with three alternative commercial media and two RPMI formulations, Stemline<sup>®</sup> demonstrated >40% more total viable cells.

Ordering Information
Product Description
Stemline® T Cell Expansion Medium

#### **Features and Benefits**

- Serum-free formulation
- Excellent expansion of T cells of human origin
- Supports high cell densities that exhibit rigorous and consistent growth kinetics
- Maintains the proper CD4/CD8 ratio in flow cytometric analysis
- Maintains functionality, both *ex vivo* and *in vivo*

# DC progenitor

#### In-Vitro Interaction of Dendritic Cells, T Cells and Tumor Cells

#### Figure 6.

As dendritic cells (DC) begin to process antigens, they mature and exhibit a more star-shaped appearance. Mature dendritic cells process antigen and present it to cytotoxic T cells (CTL). Activated cytotoxic T cells now recognize the tumor and destroy it.

Size	Cat. No.
500 mL	S1694

## **Stemline**<sup>®</sup> **Mesenchymal Stem Cell Expansion Medium**

Developed to promote optimal expansion of human mesenchymal stem cells (MSC) from bone marrow, Stemline<sup>®</sup> Mesenchymal Stem Cell Expansion Medium demonstrates greater total nucleated cell (TNC) fold increases than other commercially available formulations. Additionally, functional trials clearly demonstrate the Stemline® capacity to promote differentiation into adipocytes, chondrocytes and osteocytes.

Produced in a GMP state-of-the-art facility with a Device Master File (DMF) on file, Stemline<sup>®</sup> Mesenchymal Stem Cell Expansion Medium is an excellent choice for your MSC applications.

Stemline® Mesenchymal Stem Cell Expansion Medium requires supplementation with antibiotics, cytokines, L-glutamine and fetal bovine serum, as appropriate to individual research protocols. Known to be extremely sensitive during initial isolation and growth ex vivo, MSC proliferation depends highly on the composition of fetal bovine serum (FBS) used to supplement the medium. Pre-screening with FBS is recommended, as the specific FBS components that affect MSC growth have not been fully identified.



Stemline® Demonstrates Superior Expansion of Mesenchymal Stem Cells (MSC)



Figure 7.

To test the ability of Stemline® Mesenchymal Stem Cell Expansion Medium to promote expansion of MSCs, researchers at Sigma-Aldrich® designed a bench-scale assay. Triplicate 2 mL cultures at 5,000 MSCs/cm<sup>2</sup> were grown in a 6-well microplate culture system in Stemline® Medium or other media containing FBS. Each well was treated with trypsin/EDTA, triturated, and harvested after a 14-day expansion. MSCs were counted using a hemacytometer and average viable cell count determined for each condition. MSCs cultured in Stemline® demonstrated superior expansion to those grown in other MSC media, retained their differentiation potential and were easily passaged routinely.

Ordering Information	
Product Description	
Stemline® Mesenchym	al Stem Cell Expansion Medium

#### **Features and Benefits**

- Maximum expansion of CD34+ progenitors
- Supports robust, highdensity cell populations
- Superior expansion
- Cells retain their differentiation potential at 14 days in culture

1	

Competitor B

Size	Cat. No.
1 L	S1569

## Stemline® Keratinocyte Medium II

Developed to promote optimal expansion of human epidermal keratinocytes from adult and neonatal sources, Stemline<sup>®</sup> Keratinocyte Medium II performs most effectively when supplemented with Stemline<sup>®</sup> Keratinocyte Growth Supplement (**Cat. No. S9945**).

Stemline<sup>®</sup> Keratinocyte Medium II is free of serum and all other animal components; this exclusion increases performance consistency and eliminates safety risks associated with potential adventitious agents.

Produced in a GMP state-of-the-art facility, Stemline<sup>®</sup> Keratinocyte Stem Cell Expansion Medium is an excellent choice for your keratinocyte applications.



**Migration of Keratinocytes** 



Ordering Information		
Product Description	Size	Cat. No
Stemline <sup>®</sup> Keratinocyte Medium II	500 mL	S0196
Stemline <sup>®</sup> Keratinocyte Growth Supplement	1 vial	S9945

#### **Features and Benefits**

- Serum-free basal formulation
- Two supplement cocktails
- Regional expansion
   of NHEK cells

#### Figure 8.

The epidermis is composed of 4 layers of keratinocytes. The stratum basale, the deepest layer, is composed of columnshaped cells that constantly divide and force existing cells into higher layers. As the cells migrate through these layers, they flatten and eventually undergo terminal differentiation which leads to programmed cell death. The top layer, the stratum corneum, is composed of these dead keratinocytes, which are continuously rubbed off and replaced anew.

### **Stemline® Media Selection Guide**

Name	Key Attributes	Animal Components	Required L-Glutamine Supplementation	Regulatory	Size	Cat. No.
Stemline® I & II Hematopoietic Stem Cell Expansion Media	Serum-free formulation	Human serum	S0189 requires	Manufactured	500 mL	S0189
	Enhanced expansion from cord blood	albumin	L-Glutamine	cGMP; DMF on file	500 mL	50192
	CD34+ cells					
	Expands cells from all appropriate hematopoietic lineages in a colony-forming unit					
	Tested extensively in development in 7-day and 14-day growth assays					
	For more information, see page 4					
Stemline®	Serum-free formulation	None	None	Manufactured	500 mL	S3194
Neural Stem Cell Expansion Medium	For use with both neurospehere and monolayer cultures			CGMP; DMF on file		
	Cells retain differentiation capacity					
	Superior expansion rates					
	For more information, see page 6					
Stemline®	Serum-free formulation	Human serum	4 mM	Manufactured	500 mL	S1694
I Cell Expansion Medium	Excellent expansion of T cells of human origin	albumin, cholesterol,		CGMP; DMF on file		
	Supports high cell densities that exhibit rigorous and consistent growth kinetics	human transferrin				
	Maintains the proper CD4/CD8 ratio in flow cytometric analysis					
	Maintains functionality, both <i>ex vivo</i> and <i>in vivo</i>					
	For more information, see page 8					
Stemline <sup>®</sup> Mesenchymal Stem Cell Expansion Medium	Maximum expansion of CD34+ progenitors	Human	4 mM	Manufactured	1 L	S1569
	Supports robust, high-density cell populations	transferrin; requires FBS		cGMP; DMF on file		
	Superior expansion	supplementation				
	Cells retain their differentiation potential at 14 days in culture					
	For more information, see page 10					
Stemline® Keratinocyte Medium II	Serum-free basal formulation	Requires	4 mM	Manufactured	500 mL	S0196
	Two supplement cocktails	supplementation with S9945		cGMP		
	Regional expansion of NHEK cells					
	For more information, see page 12					

To request more information, contact your local Sigma-Aldrich representative or visit SigmaAldrich.com/stemcells

To inquire about volume discounts, contact your local Sigma-Aldrich representative today.

#### References

- Choong, M., *et al.*, MicroRNA expression profiling during human cord blood-derived CD34 cell erythropoiesis. *Experimental Hematology*, 35, 551–564 (2007).
- 2. Levay, K, *et al.*, Tescalcin is an essential factor in megakaryocytic differentiation associated with Ets family gene expression. *Journal of Clinical Investigations*, **117**, 2672–2683 (2007).
- 3. Lu, S., *et al.*, Generation of functional human gioblasts from human embryonic stem cells. *Nature Methods*, **4**, 501–509 (2007).
- 4. McNiece, I., *et al.*, Delivering cellular therapies: lessons learned from ex vivo culture and clinical applications of hematopoietic cells. *Seminars in Cell & Developmental Biology*, **18**, 839–45 (2007).

- Stec, M., et al., Expansion and differentiation of CD14+CD16and CD14++CD16+ human monocyte subsets from cord blood CD34+ hematopoietic progenitors. *Journal of Leukocyte Biology*, 82, 594–602 (2007).
- Wulf-Goldenberg, A., et al., Cytokine pre-treatment of CD34+ cord blood stem cells in vitro reduces long-term cell engraftment in NOD/ SCID mice. European Journal of Cell Biology, 87, 69–80 (2007).
- Nervi, B., *et al.*, Factors affecting human T cell engraftment, trafficking, and associated xenogenic graft-vs-host disease in NOD/ SCID β2mnull mice. *Experimental Hematology*, **35**, 1823–1838.

MilliporeSigma 400 Summit Drive Burlington, MA 01803



Copyright © 2017 EMD Millipore Corporation. All Rights Reserved. MilliporeSigma, Milli-Q, Millipore and the Vibrant M are trademarks of Merck KGaA, Darmstadt, Germany. Sigma-Aldrich, Supelco, SAFC and BioReliance are trademarks of Sigma-Aldrich Co. LLC. or its affiliates. Stemline is a registered trademark of Sigma-Aldrich Co. LLC. or its affiliates. All other trademarks are the property of their respective owners. milliporesigma.com

NFS Ver. 2.0 2016 - 00007 09/2017