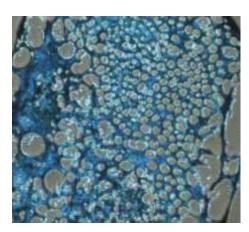


## Alcian Blue Staining Kit Specifications and Instruction Quick Steps





Lifeline's adipose-derived mesenchymal stem cells differentiated to chondrocytes and stained with Alcian Blue (100X).

#### **Alcian Blue Staining Kit Components**

Alcian Blue stains sulfated proteoglycans deposits that are indicative of functional chondrocytes and it is a useful tool when used with Lifeline's mesenchymal stem cells and Lifeline's ChondroLife™ Complete Chondrogenesis Medium. This kit contains fixative solution, sucrose stabilizer solution, Alcian Blue stain solution, and acetic acid wash solution for staining sulfated proteoglycans deposited by chondrocytes.

#### **Safety Statement**

These products are for <u>Research Use Only</u>. These products are not approved for human or veterinary use, or for use in *in vitro* diagnostics or clinical procedures. Please use and dispose of paraformaldehyde, and reagents in accordance with local and federal regulations.

#### The Lifeline Guarantee

Lifeline's rigorous quality control ensures sterility and performance to standardized testing criteria. Upon request, Lifeline will provide lot-specific QC test results, material safety data sheets and certificates of analysis. See complete guarantee/warranty statement at lifelinecelltech.com or contact your Lifeline representative for more information.

Product	Part No.	Volume	Storage	
Alcian Blue Staining Kit	LL-0051			
20% Sucrose Stabilizer	CM-0051	10 mL Room temp.		
1% Alcian Blue Stain Solution	CM-0052	100 mL	) mL Room temp.	
3% Acetic Acid Wash	CM-0053	100 mL Room temp.		
4% Paraformaldehyde Fixative Solution	CM-0055	100 mL	-20°C	

#### Additional Materials Recommended but Not Provided by Lifeline Cell Technology

- Embedding compound (such as OCT™ Compound)
- Cryomolds
- Microtome

- Absolute ethanol
- Dry ice/isopropanol bath
- Glass slides

(OTC is a trademark of Tissue-Tek®, Andwin Scientific)

To place an order, please visit lifelinecelltech.com or call customer service at 877.845.7787.

# Quick Steps for Fixation and Staining of Sulfated Proteoglycan Deposits in Chondrogenic Microbeads (Cultured in a 48-Well Plate Format)

- 1. Always wash hands before and after working with cell cultures.
- 2. Always wear eye protection and gloves when working with cell cultures.
- 3. When working with cells or medium, always use a certified biological safety cabinet.
- 4. Use 4% Paraformaldehyde Fixative Solution in a chemical fume hood.
- 5. Remove medium completely from well(s).
- 6. Add 0.5 mL 4% Paraformaldehyde Fixative Solution (CM-0055) per well.
- 7. Fix Microbeads for 3 hours at Room Temperature. Shield from light.
- 8. Remove fixative and add 0.5 mL of 20% Sucrose Stabilizer (CM-0051) per well.
- 9. Incubate overnight at room temperature. Shield from light.
- 10. Transfer Chondrogenic Microbeads to a plastic cryomold to embed them in a suitable material (such as OCT™ Compound, or equivalent) for slicing on a microtome or cryostat.
- 11. Remove residual sucrose solution.
- 12. Gently layer the embedding material over the Chondrogenic Microbeads so as to maintain separation of the individual Microbeads and maintain their location towards the bottom of the cryomold.
- 13. Snap freeze the cryomold in a dry ice/isopropanol bath. Store at -80°C until ready to cut.
- 14. Cut 5  $\mu$ m sections from block and mount on glass slide.
- 15. Place slide in 3% Acetic Acid Wash (CM-0053) for 3 minutes.
- 16. Place in 1% Alcian Blue Stain Solution (CM-0052) for 30 minutes.
- 17. Wash in running tap water for 1 minute.
- 18. Dip briefly in absolute ethanol to dehydrate and air dry.
- 19. Staining is complete.

### For any question on cell staining, please contact technical service at 877.845.7787. We are here to help.

Other Recommended Products	Part No.	Unit	Components	Storage
Human Mesenchymal Stem Cells- Wharton's Jelly (HMSC-WJ)	FC-0020	0.5 x 10 <sup>6</sup> Cells		-150°C
Human Mesenchymal Stem Cells-Adipose (HMSC-Ad)	FC-0034	10 <sup>6</sup> Cells		-150°C
StemLife™ MSC Medium Complete Kit	<u>LL-0034</u>	Kit	StemLife Basal Medium StemLife MSC LifeFactors® Kit	2-8°C when prepared
Human Mesenchymal Stem Cells-Bone Marrow (HMSC-BM)	FC-0057	10 <sup>6</sup> Cells		-150°C
StemLife MSC-BM Medium Complete Kit	<u>LL-0062</u>	Kit	StemLife Basal Medium StemLife MSC-BM LifeFactors Kit	2-8°C when prepared
Human Pre-Adipocytes, Adult (HMSC-Pre-Adipocyte)	FC-0062	10 <sup>6</sup> Cells		-150°C
FibroLife® S2 Medium Complete Kit	<u>LL-0011</u>	Kit	FibroLife Basal Medium FibroLife S2 LifeFactors Kit	2-8°C when prepared
ChondroLife™ Complete Chondrogenesis Medium	<u>LM-0022</u>	100 mL		-20°C
TrypKit™	LL-0013	Kit	PBS 0.05% Trypsin/0.02% EDTA Trypsin Neutralizing Solution	RT -20°C -20°C
Optional Supplements	Part No.	Volume	<b>Concentrations of Supplement</b>	Storage
Phenol Red Supplement	LS-1009	1 mL	33 mM	RT
Antimicrobial Supplement: Gentamicin and Amphotericin B (Provided with purchase of LL-0011, LL-0034, or LL-0062)	<u>LS-1104</u>	0.5 mL	Gentamicin 30 mg/mL Amphotericin B 15 μg/mL	-20°C

### Call Lifeline Technical Service and Sales at 877.845.7787

or visit lifelinecelltech.com for more information

Lifeline Cell Technology – 8415 Progress Drive, Suite T – Frederick, MD 21701

©2019 Lifeline Cell Technology. All Rights Reserved