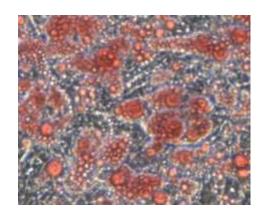


# Oil Red O Staining Kit Specifications and Instruction Quick Steps





Lifeline's adipose-derived mesenchymal stem cells differentiated to adipocytes and stained with Oil Red O (200X).

### **Oil Red O Staining Kit Components**

Oil Red O stains lipid vesicles which are created by functional adipocytes and is a useful tool when used with Lifeline's human mesenchymal stem cells and Lifeline's AdipoLife<sup>TM</sup> DfKt<sup>TM</sup>-1 or DfKt-2 Adipogenesis Media. The Oil Red O kit contains fixative solution, dehydration solution, Oil Red O stain solution, and stain differential solution for staining lipid vesicles in adipocytes.

# **Safety Statement**

These products are for Research Use Only and are not approved for human or veterinary use, or for use in *in vitro* diagnostics or clinical procedures. Paraformaldehyde releases formaldehyde gas, which is a known carcinogen. It is best to work with paraformaldehyde in a chemical fume hood. Please 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			
Oil Red O Lipid Staining Kit	LL-0052					
0.5% Oil Red O Stain Solution	CM-0054	100 mL	RT			
4% Paraformaldehyde Fixative Solution	CM-0055	100 mL	-20°C			
100% 1,2-Propanediol Dehydration Solution	CM-0056	100 mL	RT			
85% 1,2-Propanediol Stain Differential Solution	CM-0057	100 mL	RT			
Other Recommended Products	Part No.	Volume	Storage			
Phosphate Buffered Saline	<u>CM-0001</u>	500 mL	RT			
Additional Materials Recommended but Not Provided by Lifeline Cell Technology						
Deionized water (dH <sub>2</sub> O)	ter (dH <sub>2</sub> O) • Coarse filter paper					

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

# Quick Steps for Fixation and Staining of Adipocytes for Lipid Vesicles (Cultured in a 6-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.
- 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. Adipocytes may be stained with Oil Red O at either 37°C or 60°C.
- 6. Pre-heat an oven to 60°C or a water bath to 37°C.
- 7. For each well to be stained, pre-warm 2.5 mL of Oil Red O Solution (37°C or 60°C) in a 50 mL conical tube.
- 8. Once Oil Red O Solution has come to temperature, filter the solution through a coarse Whatman® filter paper, or equivalent, into a fresh tube to remove small particulates.
- 9. Return Oil Red O Solution to warming at 37°C or 60°C.
- 10. Partially aspirate AdipoLife™ Adipogenesis Medium from each well. Never allow monolayer to be exposed to air at any point during staining. If the cells are allowed to dry the lipid vesicles will burst.
- 11. Gently, from the side of the well, add 2 mL of Lifeline® PBS (CM-0001).
- 12. Repeat steps 10 and 11 two more times.
- 13. Partially aspirate PBS from the wells.
- 14. Gently add 2 mL of 4% Paraformaldehyde Fixative Solution (CM-0055) to each well of a 6-well plate.
- 15. Partially remove 4% Paraformaldehyde from each well, and replace with another 2 mL of 4% Paraformaldehyde.
- 16. Fix for at least 20 minutes at room temperature.
- 17. Partially remove Paraformaldehyde solution.
- 18. Rinse each well twice with 2 mL of deionized water.
- 19. Gently add 1 mL of the 100% 1,2-Propanediol Dehydration Solution (CM-0056) to each well.
- 20. Incubate for 5 minutes at room temperature. Gently tilt the plate 2 to 3 times to mix.
- 21. Partially remove Dehydration Solution.
- 22. Gently add another 1 mL of 100% 1,2-Propanediol Dehydration Solution to each well.
- 23. Incubate for 5 minutes at room temperature. Gently tilt the plate 2 to 3 times to mix.
- 24. Partially remove Dehydration Solution.
- 25. Gently add 2 mL Oil Red O Stain Solution (CM-0054) to each well.
- 26. Incubate at 37°C for 30 minutes or at 60°C for 8 minutes. Tilt the plate 2 to 3 times during the incubation.
- 27. Partially remove Oil Red O Solution.
- 28. Gently add 2 mL of 85% 1,2-Propanediol Stain Differential Solution (CM-0057) for 1 minute to differentiate stain. **Do not tilt the plate, or agitate the Stain Differential Solution in the well(s)!**
- 29. Partially remove the Stain Differential Solution.
- 30. Rinse wells carefully twice with 2 mL deionized water per well.
- 31. Leave the final deionized water rinse in the well.
- 32. Fixation and staining is complete.

(Whatman is a registered trademark of GE Healthcare.)

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

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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-Adipocytes)	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
AdipoLife™ DfKt™-1 Adipogenesis Medium	<u>LL-0050</u>	Kit	AdipoLife Basal Medium DifFactor™ 1 DifFactor 2	2-8°C when prepared
AdipoLife DfKt-2 Adipogenesis Medium	<u>LL-0059</u>	Kit	AdipoLife Basal Medium DifFactor 3	2-8°C when prepared
TrypKit™	<u>LL-0013</u>	Kit	PBS 0.05% Trypsin/0.02% EDTA Trypsin Neutralizing Solution	RT -20°C -20°C
Optional Supplements	Part No.	Volume	Concentrations of Supplement	Storage
Phenol Red Supplement	<u>LS-1009</u>	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's Customer Service at 877.845.7787

or visit lifelinecelltech.com for more information

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

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