



FibroGRO™ Media Products for culture of Human Fibroblasts

Product Manual for Cat. Nos.
SCMF001
SCMF002

FOR RESEARCH USE ONLY
Not for use in diagnostic procedures.

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Introduction

FibroGRO™ Media formulations provide an optimal cell culture environment for many types of human fibroblasts and mouse embryonic fibroblasts. These low serum/serum-free media formulations have been shown to grow these cells at rates that meet or exceed commercially available serum containing media, while maintaining excellent cell morphology. The media are packaged in a specially designed UV protective shrinkwrap for added stability, which includes a temperature gauge for added convenience. FibroGRO Media contain no antimicrobials and no phenol red, components that can cause cell stress and masking effects that may influence experimental results.

Two different media formulations are available for various applications and cell culture requirements:

Media Format	Catalog Code:	Application	Notes
FibroGRO™ Complete Kit	<i>SCMF001</i>	Serum-free culture of human fibroblasts, stromal cells, mesenchymal cells and also mouse embryonic fibroblasts.	Serum-free formulation
FibroGRO™-LS Complete Media	<i>SCMF002</i>	Low serum formulation for culture of human fibroblasts, stromal cells, mesenchymal cells and also mouse embryonic fibroblasts.	2% serum containing medium

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Storage

Each media kit consists of two components: a) a bottle of basal medium and b) a supplement kit containing various growth factors and components.

Basal Medium should be stored at 2 to 8°C. The special UV protective packaging helps protect FibroGRO medium from light damage, however users should take care to protect basal medium from extended exposure to light.

Supplement Kits should be stored at -20°C. Do not use product beyond expiration date.

All components are guaranteed stable until the expiration date stated on the individual labels.

Quality Control

These products are manufactured with the highest quality of raw materials, with exacting standards and production procedures to ensure lot-to-lot consistency. Every lot of FibroGRO Media is extensively tested using normal human fibroblasts for the following parameters:

Sterility testing:	Negative for bacteria and fungal growth
pH:	7.4 +/- 0.2
Cell testing:	Rate of proliferation and morphology
Osmolality:	270 +/- 10mOsm
Endotoxin:	<0.5 EU/mL

Product Specifications: FibroGRO™-LS Complete Media Kit

Description	FibroGRO-LS Complete Media Kit (Catalog # SCMF002)			
Kit Components	FibroGRO Basal Medium (Part# SCMF-BM) FibroGRO-LS Supplement Kit (Part# SCMF002-S)			
Application	FibroGRO-LS Medium is optimized for low-serum (2%) culture of human fibroblasts, and supports the growth of these cells in a 2% serum environment at rates equal to or greater than comparable media supplemented with much higher concentrations of FBS. FibroGRO-LS Medium contains no antimicrobials and no phenol red; components that can cause cell stress and “masking effects” that may influence experimental results (these are not needed, or recommended, to achieve optimal cell performance). FibroGRO provides a perfect cell system to establish low serum human feeder layers for stem cell culture, or as a model to study wound healing, toxicology or basic cell biology			
Cells supported by FibroGRO-LS Medium	<ul style="list-style-type: none"> • Human Fibroblasts • Mesangial Cells • Mesenchymal Cells • Mouse embryonic fibroblasts 			
	Part #	Volume	Final Concentration in Supplemented Medium	Storage
FibroGRO™-LS Complete Media Kit	SCMF002	Kit		See individual components
FibroGRO™ Basal Medium	SCMF-BM	480 mL		2-8°C
FibroGRO™-LS Supplement Kit, containing:	SCMF002-S			-20°C
rhFGF-b		1.0 mL	5 ng/mL	-20°C
Ascorbic Acid		0.5 mL	50 ug/mL	-20°C
L-Glutamine		18.75 mL	7.5 mM	-20°C
Hydrocortisone Hemisuccinate		0.5 mL	1.0 µg/mL	-20°C
rh Insulin		0.5 mL	5 µg/mL	-20°C
FBS		10 mL	2%	-20°C

Product Specifications: FibroGRO™ Complete Media Kit

Description	FibroGRO Complete Media Kit (Catalog # SCMF001)			
Kit Components	FibroGRO Basal Medium (Part# SCMF-BM) FibroGRO Supplement Kit (Part# SCMF001-S)			
Application	FibroGRO Complete Medium is a defined medium optimized for serum-free culture of human fibroblasts, and supports the growth of these cells in a serum-free environment at rates equal to or greater than comparable media supplemented with much higher concentrations of FBS. FibroGRO Medium contains no antimicrobials and no phenol red; components that can cause cell stress and “masking effects” that may influence experimental results (these are not needed, or recommended, to achieve optimal cell performance). FibroGRO medium provides a perfect cell system to establish serum-free human feeder layers for stem cell culture, or as a model to study wound healing, toxicology or basic cell biology			
Cells supported by FibroGRO-VEGF Medium	<ul style="list-style-type: none"> • Human Fibroblasts • Mesangial Cells • Mesenchymal Cells • Mouse embryonic fibroblasts 			
	Part #	Volume	Final Concentration in Supplemented Medium	Storage
FibroGRO™ Complete Media Kit	SCMF001	Kit		See individual components
FibroGRO™ Basal Medium	SCMF-BM	480 mL		2-8°C
FibroGRO™ Supplement Kit, containing:	SCMF001-S			-20°C
rh EGF/TGFβ1		0.5 mL	5 ng/mL/ 30 pg/mL	-20°C
rh FGF basic		0.5 mL	5 ng/mL	-20°C
HLL Supplement: HSA, linoleic acid and lecithin		1.25 mL	HSA: 500 µg/mL Linoleic Acid: 0.6 µM Lecithin: 0.6 µg/mL	-20°C
Ascorbic Acid		0.5 mL	50 µg/mL	-20°C
L-Glutamine		18.75 mL	7.5 mM	-20°C
Hydrocortisone Hemisuccinate		0.5 mL	1.0 µg/mL	-20°C
rh Insulin		0.5 mL	5 µg/mL	-20°C

Media Preparation

All FibroGRO™ Media are provided as kits containing a basal media (480 mL) and a supplement kit containing supplements and growth factors, unique to the specific media/application. This allows you to prepare fresh medium each time, providing optimal cell culture conditions. To support proliferation, you must add the necessary supplements in the appropriate concentrations to the basal medium. FibroGRO Media do not contain phenol red or antibiotics. These components are not necessary for cell proliferation, but may be added if desired.

Pre-warming the Medium

Medium will take from 10 to 30 minutes to warm to 37°C depending on the volume. Media temperature may be checked by referencing the thermometer attached to the side of the media bottle. Do not leave medium in water bath for extended periods. If only using a small volume of medium (less than 50 mL), warm only the volume needed in a sterile conical tube. Repeated warming of the entire bottle over extended periods may cause degradation of the medium and reduced shelf life.

Adding Supplements

Supplement Kits contain sufficient reagents to supplement one 480 mL bottle of FibroGRO Basal Medium. Supplements should be thawed immediately prior to supplementation; Mix supplemented medium by gently pipetting up and down with a large volume pipette (25 or 50 mL) or gently invert the tightly closed 500 mL bottle. Do not shake or froth the medium. The supplemented medium may be stored at 2 to 8°C for up to two weeks. All procedures should be done using sterile technique (see section on basic sterile technique below).

Please note that L-Glutamine is best warmed to 37°C in a water bath, and shaken to dissolve the precipitate prior to use.

Recommended Feeding Schedule

The following guidelines are for a T-25 flask. Adjust volumes according to culture surface area.

Cultures under 20% confluent (to be re-fed in 2 days)	Re-feed with 5 mL of warmed medium
Cultures under 20% confluent (to be re-fed in 3 days)	Re-feed with 7 mL of warmed medium
Cultures 20-30% confluent (to be re-fed or passaged in 2 days)	Re-feed with 8 mL of warmed medium
Cultures over 30% confluent (to be passaged in 2 days)	Re-feed with 7-10 mL of warmed medium

Basic Sterile Technique

FibroGRO Media should only be used in a sterile environment, a Class II biological safety cabinet with front access and filtered laminar airflow, or an equivalent device. Always wear gloves and eye protection when working with these materials. Wipe or spray medium bottle and Supplement Kits with 70% ethanol or isopropanol before opening, especially around the area of the cap. Make sure these surfaces have dried before opening the bottle or vials. Transfer of solutions should be done with disposable sterile pipettes. **Do not mouth pipette!** Withdraw the volume needed into the pipette, being careful not to touch the sterile tip to the rim of the container or any other surface. Close the container and open the container into which the transfer is being made, again being careful not to touch any surfaces with the sterile tip. Transfer the material and close the container. Wash your hands before and after working with cell cultures. Do not block airflow in a laminar flow hood as this may compromise sterility. Ensure that biological cabinets are certified routinely and the HEPA filters are replaced regularly.

A note on Optional Supplements

Phenol Red:

Phenol red is a pH indicator that is not required in cell culture and may adversely influence the behavior of some cell types, since it has estrogenic properties. Medium with phenol red will appear more yellow than red in acidic conditions and will appear more purple than red in basic conditions. This supplement is not included in FibroGRO media kits, but may be added if desired.

Penicillin-Streptomycin-Amphotericin (PSA):

PSA is used to minimize contamination. These antimicrobials react with cells and may inhibit optimal growth. If proper sterile technique is used, antimicrobials should not be necessary.

Troubleshooting

Problem	Cause	Solution
No growth of cells	FibroGRO™ Basal Medium does not support cell growth	Be sure to add all supplements to Basal Medium. Ensure all supplements are within expiration date.
Grainy morphology	A particular supplement has been left out of medium	Be sure to add all supplements to FibroGRO Basal Medium; remake if necessary
Growth of cell culture slows down when 30% or more confluent	Culture is not being fed frequently enough, or not being fed with an appropriate volume of medium	Adhere to recommended feeding guidelines on page 6.

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