



EndoGRO™ Media Products for Human Endothelial Cell Culture

Product Manual for following Cat. Nos.

SCME001

SCME002

SCME003

SCME004

FOR RESEARCH USE ONLY
Not for use in diagnostic procedures.

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Introduction

EndoGRO Media Formulations provide an optimal cell culture environment for many types of endothelial cells, including HUVEC, aortic endothelial cells, and other human large vessel and microvascular endothelial cells. These media have been shown to grow endothelial 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. EndoGRO Media contain no antimicrobials and no phenol red, components that can cause cell stress and masking effects that may influence experimental results.

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

Media Format	Catalog Code:	Application	Notes
EndoGRO™-LS	SCME001	Low serum culture of Human Endothelial Cells, HUVEC, aortic endothelial cells and other large vessel endothelial cells	Low serum formulation. Does not contain VEGF. Contains EndoGRO-LS Growth Supplement.
EndoGRO™-VEGF	SCME002	Low serum formulation for rapid proliferation of Human Endothelial Cells, HUVEC, aortic endothelial cells and other large vessel endothelial cells	Low serum formulation. Contains VEGF. Rapid proliferation rates. Not appropriate for stimulation assays where VEGF is primary stimulator
EndoGRO™-MV-VEGF	SCME003	Low serum formulation for rapid proliferation of human microvascular endothelial cells	5% serum formulation. Contains VEGF. Not appropriate for stimulation assays where VEGF is primary stimulator
EndoGRO™-MV	SCME004	Low serum formulation for culturing human microvascular endothelial cells	5% serum formulation. Does not contain VEGF. Contains Endothelial Cell Growth Supplement.

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Storage

Each media kit consists of two components:

a) one bottle of basal medium

Basal Medium should be stored at 4 to 8°C. The special UV protective packaging protects EndoGRO™ medium from light damage, however users should take care to protect basal medium from extended exposure to light.

b) one supplement kit containing various growth factors and components.

Supplement Kits should be stored at -20°C for up to 6 months. 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 EndoGRO Media is extensively tested using normal human endothelial cells for the following parameters:

Sterility testing:	Negative for bacteria and fungal growth
pH:	7.8 +/- 0.3
Cell testing:	Rate of proliferation and morphology
Osmolality:	270 +/- 10mOsm
Endotoxin level:	<0.5 EU/mL

Product Specifications: EndoGRO™-LS Complete Media Kit

Description	EndoGRO-LS Complete Media Kit (Catalog # SCME001)			
Kit Components	EndoGRO Basal Medium (Part# SCME-BM) EndoGRO-LS Supplement Kit (Part# SCME-001S)			
Application	EndoGRO-LS Medium (containing EndoGRO-LS Supplement) is a new low serum medium optimized for the culture of human endothelial cells, including Human Umbilical Vein Endothelial Cells (HUVEC), aortic endothelial cells and other human large-vessel endothelial cells. EndoGRO-LS Medium supports the growth of these cells in a low serum environment without human VEGF. EndoGRO-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).			
Cells supported by EndoGRO-LS Medium	<ul style="list-style-type: none"> • Human endothelial cells • HUVEC • Aortic endothelial cells • Other large vessel endothelial cells 			
	Part #	Volume	Final Concentration in Supplemented Medium	Storage
EndoGRO™-LS Complete Media Kit	SCME001	Kit		See individual components
EndoGRO™ Basal Medium	SCME-BM	475 mL		2-8°C
EndoGRO™-LS Supplement Kit, containing:	SCME001-S			-20°C
EndoGRO-LS Supplement		1.0 mL	0.2%	-20°C
rh EGF		0.5 mL	5 ng/mL	-20°C
Ascorbic Acid		0.5 mL	50 µg/mL	-20°C
L-Glutamine		25 mL	10 mM	-20°C
Hydrocortisone Hemisuccinate		0.5 mL	1.0 µg/mL	-20°C
Heparin Sulfate		0.5 mL	0.75 U /mL	-20°C
FBS		10 mL	2%	-20°C

Product Specifications: EndoGRO™-VEGF Complete Media Kit

Description	EndoGRO-VEGF Complete Media Kit (Catalog # SCME002)			
Kit Components	EndoGRO Basal Medium (Part# SCME-BM) EndoGRO-VEGF Supplement Kit (Part# SCME-002S)			
Application	EndoGRO-VEGF (containing Vascular Endothelial Growth Factor) is a new low serum medium optimized for the rapid proliferation of human endothelial cells including Human Umbilical Vein Endothelial Cells (HUVEC), aortic endothelial cells and other human large-vessel endothelial cells. EndoGRO-VEGF supports the growth of these cells in a low serum environment. EndoGRO-VEGF 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). EndoGRO-VEGF contains rh VEGF, which may interfere with stimulation assays especially where VEGF is the primary stimulator. For these studies, we recommend the use of EndoGRO-LS Medium.			
Cells supported by EndoGRO-VEGF Medium	<ul style="list-style-type: none"> • Human endothelial cells • HUVEC • Aortic endothelial cells • Other large vessel endothelial cells 			
	Part #	Volume	Final Concentration in Supplemented Medium	Storage
EndoGRO™-VEGF Complete Media Kit	SCME002	Kit		See individual components
EndoGRO™ Basal Medium	SCME-BM	475 mL		2-8°C
EndoGRO™-VEGF Supplement Kit, containing:	SCME002-S			-20°C
rh VEGF		0.5 mL	5 ng/mL	-20°C
rh EGF		0.5 mL	5 ng/mL	-20°C
rh FGF basic		0.5 mL	5 ng/mL	-20°C
rh IGF-1		0.5 mL	15 ng/mL	-20°C
Ascorbic Acid		0.5 mL	50 µg/mL	-20°C
Hydrocortisone Hemisuccinate		0.5 mL	1.0 µg/mL	-20°C
Heparin Sulfate		0.5 mL	0.75 U/mL	-20°C
L-Glutamine		25 mL	10 mM	-20°C
FBS		10 mL	2%	-20°C

Product Specifications: EndoGRO™-MV-VEGF Complete Media Kit

Description	EndoGRO-MV-VEGF Complete Media Kit (Catalog # SCME003)			
Kit Components	EndoGRO Basal Medium (Part# SCME-BM) EndoGRO-MV-VEGF Supplement Kit (Part# SCME-003S)			
Application	EndoGRO-MV-VEGF (containing Vascular Endothelial Growth Factor) is a new medium optimized for the culture of human microvascular endothelial cells. EndoGRO-MV-VEGF supports the rapid proliferation of these cells in a 5% serum environment. EndoGRO-MV-VEGF 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). EndoGRO-MV-VEGF contains rh VEGF which may affect stimulation assays. We recommend using EndoGRO-MV for these studies.			
Cells supported by EndoGRO-MV-VEGF Medium	Human Microvascular Endothelial Cells			
	Part #	Volume	Final Concentration in Supplemented Medium	Storage
EndoGRO™-MV-VEGF Complete Media Kit	SCME003	Kit		See individual components
EndoGRO™ Basal Medium	SCME-BM	475 mL		2-8°C
EndoGRO™-MV-VEGF Supplement Kit, containing:	SCME003-S			-20°C
rh VEGF		0.5 mL	5 ng/mL	-20°C
rh EGF		0.5 mL	5 ng/mL	-20°C
rh FGF		0.5 mL	5 ng/mL	-20°C
rh IGF-1		0.5 mL	15 ng/mL	-20°C
L-Glutamine		25 mL	10 mM	-20°C
Hydrocortisone Hemisuccinate		0.5 mL	1.0 µg/mL	-20°C
Heparin Sulfate		0.5 mL	0.75 U/mL	-20°C
Ascorbic Acid		0.5 mL	50 µg/mL	-20°C
FBS		25 mL	5%	-20°C

Product Specifications: EndoGRO™-MV Complete Media Kit

Description	EndoGRO-MV Complete Media Kit (Catalog # SCME004)			
Kit Components	EndoGRO Basal Medium (Part# SCME-BM) EndoGRO-MV Supplement Kit (Part# SCME-004S)			
Application	EndoGRO-MV (containing EndoGRO-LS Supplement) is a new medium optimized for the culture of human microvascular endothelial cells in a 5% serum environment. EndoGRO-MV 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).			
Cells supported by EndoGRO-MV Medium	Human Microvascular Endothelial Cells			
	Part #	Volume	Final Concentration in Supplemented Medium	Storage
EndoGRO™-MV Complete Media Kit	SCME004	Kit		See individual components
EndoGRO™ Basal Medium	SCME-BM	475 mL		2-8°C
EndoGRO™-MV Supplement Kit, containing:	SCME004-S			-20°C
EndoGRO-LS Supplement		1.0 mL	0.2%	-20°C
rh EGF		0.5 mL	5 ng/mL	-20°C
L-GLutamine		25 mL	10 mM	-20°C
Hydrocortisone Hemisuccinate		0.5 mL	1.0 µg/mL	-20°C
Heparin Sulfate		0.5 mL	0.75 U/mL	-20°C
Ascorbic Acid		0.5 mL	50 µg/mL	-20°C
FBS		25 mL	5%	-20°C

Media Preparation

All EndoGRO Media are provided as kits containing a basal media (475 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. EndoGRO 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 475 mL bottle of EndoGRO 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

EndoGRO 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 EndoGRO 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	EndoGRO 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 EndoGRO 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 8.

Related Products

EndoGRO™ Normal HUVEC (Catalog Code SCCE001)

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