# Cascade Biologics™

invitrogen cell culture



# Medium 131, MVGS, and AF

# Medium 131

M-131-500 500 ml

## **Product Description**

Medium 131 is a sterile, liquid tissue culture medium intended for use as one component in a complete culture environment for the growth of normal human microvascular endothelial cells. Medium 131 is a basal medium containing essential and non-essential amino acids, vitamins, other organic compounds, trace minerals, and inorganic salts. This medium does not contain antibiotics, antimycotics, hormones, growth factors, or proteins. This medium is bicarbonate buffered and is designed for use in an incubator with an atmosphere of 5% CO<sub>2</sub>/95% air. To support the plating and proliferation of human microvascular endothelial cells, Medium 131 must be supplemented with Microvascular Growth Supplement (MVGS, cat. no. S 005-25). In addition, we recommend coating tissue culture surfaces with Attachment Factor (AF, supplied with Medium 131) before use.

#### **Intended Use**

Medium 131 is intended for use in the routine culture of normal human microvascular endothelial cells. When supplemented with MVGS, Medium 131 will support the plating and proliferation of microvascular endothelial cells at densities between 5 x 10<sup>3</sup> cells/cm<sup>2</sup> and 6 x 10<sup>4</sup> cells/cm<sup>2</sup>. Additional applications for use may include primary isolation of human microvascular endothelial cells. *This product is for research use only. Not for use in animals, humans, or diagnostic procedures.* 

Caution: If handled improperly, some components of this product may present a health hazard. Take appropriate precautions when handling this product, including the wearing of protective clothing and eyewear. Dispose of properly.

#### Storage and Stability

Medium 131 is stored at 4° C in our facility and is shipped at ambient temperature. Upon receipt, Medium 131 should be stored at 4° C and should <u>not</u> be frozen. **Protect from light.** Several components of this tissue culture medium are light-labile, and we recommend that the medium not be exposed to light for lengthy periods of time. If the medium is warmed prior to use, do not exceed 37° C. When stored in the dark at 4° C, the product is stable until the expiration date shown on the label.

## **Preparation of Supplemented Medium 131**

- Thaw one bottle of MVGS. Take one bottle of Medium 131 from cold storage. Make sure that the caps of the vessels are tight.
- 2. Gently swirl the bottle of supplement. Avoid splashing the supplement into the cap of the bottle or causing the supplement to foam.
- Wipe the outside of the containers with a disinfecting solution such as 70% ethanol or isopropanol.
- Using sterile technique in a laminar flow culture hood, transfer the entire contents of the bottle of supplement to the bottle of Medium 131.
- 5. Tightly cap the bottle of supplemented medium and swirl the contents to ensure a homogeneous solution. Avoid causing the medium to foam.

# Storage and Stability of Supplemented Medium 131

Once Medium 131 has been supplemented with MVGS, the supplemented medium should be stored in the dark at 4° C and should <u>not</u> be frozen. When stored in the dark at 4° C, the supplemented medium is stable for 1 month.

#### References

The Medium 131 formulation is based on medium MCDB 131. Knedler, A. and Ham, R.G.; In Vitro Cell. Dev. Biol. 23: 481-491; 1987

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# MVGS Microvascular Growth Supplement

Cat. no. S-005-25 25 ml

## **Product Description**

Microvascular Growth Supplement (MVGS) is a sterile, concentrated (20X) solution intended for use as one component in a complete culture environment for the growth of human microvascular endothelial cells. Each 25 ml bottle of MVGS contains all of the growth factors, hormones, and tissue extracts necessary for the culture of these cells, and is the correct amount of supplement for a 500 ml bottle of basal Medium 131. MVGS contains fetal bovine serum (5% v/v final concentration), hydrocortisone, recombinant human fibroblast growth factor, heparin, recombinant human epidermal growth factor, and dibutyryl cyclic AMP.

#### Intended Use

MVGS is intended for use in conjunction with Medium 131 and Attachment Factor for the routine culture of normal human microvascular endothelial cells. Additional applications for use may include primary isolation of human microvascular endothelial cells. *This product is for research use only. Not for use in animals, humans, or diagnostic procedures.* 

Caution: If handled improperly, some components of this product may present a health hazard. Take appropriate precautions when handling this product, including the wearing of protective clothing and eyewear. Dispose of properly.

## Storage and Stability of MVGS

MVGS is stored at -20° C at our facility and is shipped on dry ice. Upon receipt, the product should be stored at -20° C in a freezer that is <u>not</u> self-defrosting. When stored at -20° C, the product is stable until the expiration date shown on the label.

After long-term storage at -20° C, MVGS may contain a small amount of precipitate. This precipitate is formed from cold-insoluble material in the serum component of the MVGS and will not affect the performance of the product.

### **Thawing**

To thaw, place the product in a 37° C water bath or overnight at 4° C. If thawed in a water bath, do not leave the product at 37° C after the product has thawed. For instructions on adding MVGS to Medium 131, refer to the instructions that accompany the basal medium.

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# AF Attachment Factor

Cat. no. S-006-100 100 ml

## **Product Description**

Attachment Factor (AF) is a sterile solution (1X) containing gelatin at 0.1%. AF is intended for use as one component in a complete culture environment for the growth of endothelial cells.

#### Intended Use

AF is intended for use in conjunction with Medium 131 and MVGS or Medium 200 and LSGS for the routine culture of endothelial cells. Additional applications for use may include primary isolation of endothelial cells. *This product is for research use only. Not for use in animals, humans, or diagnostic procedures.* 

## Storage and Stability

AF is stored at 4° C at our facility. When stored at 4° C, AF is stable until the expiration date shown on the label.

# Instructions for coating of culture surfaces

- Using sterile technique in a laminar flow culture hood, add 2–3 ml of the AF solution to one T25 flask or 3–4 ml to one T75 flask. Rock the flask to cover culture surface completely with AF.
- 2. Incubate flask at 37° C for 30 minutes or at room temperature for 2 hours.
- Using sterile technique in a laminar flow culture hood, completely remove the AF solution from the culture vessel by aspiration prior to the addition of cell suspension or medium. It is not necessary to wash the culture surface before adding cells or medium.
- Coated flasks may be used immediately or capped tightly and stored at room temperature for up to 24 hours.

#### Limited Use Label License No. 5: Invitrogen Technology

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