

Geltrex™ Flex LDEV-Free Reduced Growth Factor Basement Membrane Matrix

Catalog Numbers A4000046701, A4000046702, A4000046703

Pub. No. MAN1000812 Rev. A



WARNING! Read the Safety Data Sheets (SDSs) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves. Safety Data Sheets (SDSs) are available from [thermofisher.com/support](https://www.thermofisher.com/support).

Product description

Gibco™ Geltrex™ Flex LDEV-Free Reduced Growth Factor Basement Membrane Matrix (Geltrex™ Flex Matrix) is a soluble form of basement membrane that is purified from Engelbreth-Holm-Swarm (EHS) tumor cells. The solution which gels at 37°C, can be used for promotion and maintenance of a differentiated phenotype in various cell cultures, including primary epithelial cells, primary neurons, and smooth muscle cells. Geltrex™ Flex Matrix has been used in three-dimensional (3D) and two-dimensional (2D) cell culture, including angiogenesis assays, neurite outgrowth assays, and bile canaliculi formation assays in polarized hepatocytes.

In addition to its role in the physical support and compartmentalization of tissues, basement membrane influences several cellular functions, such as proliferation, adhesion, migration, differentiation, and polarization. As a result, basement membrane is implicated in diverse biological processes, including development, tissue maintenance, regeneration, wound repair, and pathological processes such as tumor growth and metastasis.

The major components of Geltrex™ Flex Matrix include laminin, collagen IV, entactin, and heparan sulfate proteoglycan. The solution is formulated without phenol red to minimize potential for estrogen-like effects.

For the culture of hESC or iPSC, Geltrex™ Flex LDEV-Free, hESC Qualified, Reduced Growth Factor Basement Membrane Matrix is recommended. See *Geltrex™ Flex LDEV-Free, hESC Qualified, Reduced Growth Factor Basement Membrane Matrix User Guide* (Pub. No. [MAN1000814](#)).

For the culture of cancer and other organoids, Geltrex™ Flex LDEV-Free, Organoid Qualified, Reduced Growth Factor Basement Membrane Matrix is recommended. See *Geltrex™ Flex LDEV-Free, Organoid Qualified, Reduced Growth Factor Basement Membrane Matrix User Guide* (Pub. No. [MAN1000813](#)).

Contents and storage

Component	Cat. No.	Amount	Storage	Shelf Life ^[1]
Geltrex™ Flex LDEV-Free Reduced Growth Factor Basement Membrane Matrix	A4000046701	1 mL	-80°C to -20°C	24 months
	A4000046702	5 mL		
	A4000046703	10 mL		

^[1] Shelf life duration is determined from the date of manufacture.

Recommended methods for specific applications

Geltrex™ Flex Matrix can be used for a variety of applications that require different basement membrane thicknesses and concentrations. Geltrex™ Flex Matrix comes at lot specific protein concentrations between 9–13 mg/mL.

- For angiogenesis studies, a protein concentration >9 mg/mL Geltrex™ Flex Matrix is recommended. Geltrex™ Flex Matrix below 4 mg/mL does not form a firm gel.
- For applications such as endothelial cell differentiation into capillary-like structures (Tube Assay), see the *Endothelial Tube Formation Assay (In Vitro Angiogenesis) Product Information Sheet* (Pub. No. MAN0001687) for further guidance on whether a thin or thick gel method is recommended.
- For 96-well plates it is recommended to use 50 µL/cm² (thin gel protocol) of Geltrex™ Flex Matrix and for 24- 48-well plates it is recommended to use 100 µL/cm² (thick gel protocol).

- For applications such as the differentiation of rat aorta tissue into capillary-like structures (Ring Assay), or cell invasion assays, a thick gel is needed. See “Coat the growth surface thick gel method” on page 2.

Procedural guidelines

- Perform all procedures in an aseptic environment, using aseptic techniques, to prevent contamination.
- Source—Murine Engelbreth-Holm-Swarm (EHS) tumor (protein concentration ranges from 9–13 mg/mL). See the Certificate of Analysis for lot-specific information.
- When working with small volumes of Geltrex™ Flex Matrix, dispense the required working volumes, then aliquot and store the remaining solution at -80°C to -20°C in a frost-free freezer.
- Avoid multiple freeze/thaw cycles.
- Geltrex™ Flex Matrix gels in 5–10 minutes when kept above 15°C. Keep Geltrex™ Flex Matrix on ice at all times during handling to prevent premature gelling.
- It is recommended to use Geltrex™ Flex Matrix within a week (7 days) from the time of thawing at 2°C to 8°C.

Before you begin

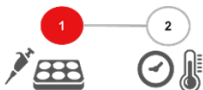


1. Thaw Geltrex™ Flex Matrix in a refrigerator overnight at 2°C to 8°C.
Consider using an aluminum or other cooling block to reduce temperature fluctuations.
2. Mix Geltrex™ Flex Matrix by slowly pipetting up and down. Be careful not to introduce air bubbles.

Coating method

Coat the growth surface: thin gel method

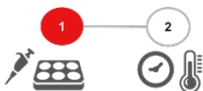
Note: Use this method for cell attachment and proliferation.



1. Pipet 50 µL of Geltrex™ Flex Matrix per cm² onto the growth surface.
2. Place coated plates at 37°C for 30 to 60 minutes.
The coated plates are ready for use.

Coat the growth surface thick gel method

Note: Use this method for ring assay, cell invasion, angiogenesis assay and other applications that require thick gels.



1. Pipet 100–200 µL of Geltrex™ Flex Matrix solution per cm² onto the growth surface.
2. Place the coated plates at 37°C for 30 to 60 minutes.
The coated plates are ready for use.

Coat the growth surface: thin layer method (non-gelling)

Note: Use this method for cell attachment and proliferation applications.

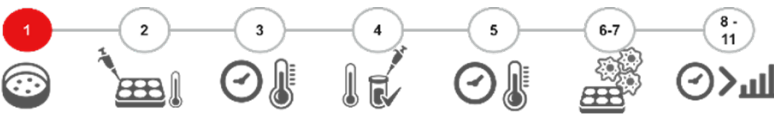


1. Dilute Geltrex™ Flex Matrix solution to the desired concentration in ice-cold serum-free medium.

Note: Empirical determination of the optimal coating concentration for your application may be required. A protein concentration of 0.1 mg/mL is recommended starting concentration for the propagation of primary cells.

2. Add enough of the solution to cover the entire growth surface.
3. Place the coated plates at 37°C until dry (may take up to 60 minutes).
The coated plates are ready for use.

Coat the growth surface: 3D sandwich culture method



Note: The following procedure is designed for coating an entire 48-well plate. A total of 15 mL of Geltrex™ Flex Matrix is required. To coat fewer or more wells, adjust the volumes accordingly.

1. Culture cells as recommended by the cell supplier to establish a stable population at 37°C in a CO₂ incubator. Growth media, growth factors, serum requirements, and incubation period can vary by cell type.
2. Work on ice and add 250 µL of Geltrex™ Flex Matrix to each well of a sterile 48-well plate.
3. Incubate the plate at 37°C for 30 to 60 minutes to promote gelling of the matrix.
4. Work on ice and add 2 mL of Geltrex™ Flex Matrix to 98 mL of growth medium (2% final concentration) in a sterile container. Label the container "Assay Medium", then swirl to mix.
5. Optional step: When working with very cold-sensitive cells, incubate Assay Medium at 37°C for 5 to 10 minutes in preparation for cell dilution.
6. Harvest cells from culture, then dilute cells in Assay Medium. Generally, cells are diluted 1×10^4 – 1×10^5 cells/mL, depending on the cell type and assay conditions. Optimization may be required.
7. Add 500 µL of cell suspension to each well of the 48-well plate containing Geltrex™ Flex Matrix. Test compounds may also be added at this time.
8. Incubate the plate at 37°C in a CO₂ incubator for 4 days.
9. Observe cell growth and 3D structure formation daily.
10. On day 4, carefully remove old medium and replace with new Assay Medium. Repeat on day 8 and day 12.
11. When 3D structures have grown to the desired size, harvest cells and analyze.

Recommendations for analysis

- Cells can be analyzed in the plate on Geltrex™ Flex Matrix. Alternatively, cells can be carefully transferred to a microscope slide, or embedded in paraffin, then sectioned.
- To fix cells, incubate in 2% formalin in PBS (1X) at room temperature for 20 minutes.
- To fix cells embedded in Geltrex™ Flex Matrix, incubate in 4% formaldehyde plus 1% glutaraldehyde in PBS for 30 minutes to prevent matrix dissolution.

Related products

Unless otherwise indicated, all materials are available through [thermofisher.com](https://www.thermofisher.com).

Item	Source
Geltrex™ Flex LDEV-Free, hESC Qualified, Reduced Growth Factor Basement Membrane Matrix (1 mL, 5 mL, 10 mL)	A4000046801 , A4000046802 , A4000046803
Geltrex™ Flex LDEV-Free, Organoid Qualified, Reduced Growth Factor Basement Membrane Matrix (1 mL, 5 mL, 10 mL)	A4000046901 , A4000046902 , A4000046903
StemFlex™ Medium	A3349401
OncoPro™ Tumoroid Culture Medium Kit	A5701201
DMEM/F-12 Medium	11320033
Human Umbilical Vein Endothelial Cells (HUVEC)	C0035C
Human Large Vessel Endothelial Cell Basal Medium (formerly Medium 200)	M-200-500
Large Vessel Endothelial Supplement (LVES)	A1460801
Primary Hepatocyte Thawing and Plating Supplements	CM3000
Primary Hepatocyte Maintenance Supplements	CM4000
Human Plateable Hepatocytes, Transporter Qualified	HMCPTS

Limited product warranty

Life Technologies Corporation and its affiliates warrant their products as set forth in the Life Technologies' General Terms and Conditions of Sale at www.thermofisher.com/us/en/home/global/terms-and-conditions.html. If you have questions, contact Life Technologies at www.thermofisher.com/support.



Life Technologies Corporation | 3175 Staley Road | Grand Island, New York 14072 USA

For descriptions of symbols on product labels or product documents, go to [thermofisher.com/symbols-definition](https://www.thermofisher.com/symbols-definition).

Revision history: Pub. No. MAN1000812 A

Revision	Date	Description
A	24 March 2025	Initial release.

The information in this guide is subject to change without notice.

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