

Attachment Factors

Collagen A

Application
For dissociation of tissue and cell monolayer

Storage
Solution at -20° C (frozen), powder at 2 – 8° C

Shelf life
Solution 24 months, powder 36 months. The shelf life commences with date of production. Our trypsin is tested negative for mycoplasma.

- Acid-soluble collagen from bovine placenta**
- Add an equal volume of sterile PBS to the collagen
 - Add 1 ml per 10 cm² of culture flask and incubate at 35 - 37° C for 30 min
 - Remove solution and wash 1x with PBS; use culture flasks immediately

In monolayer culture, normal human and murine liver cells were successfully grown for a period of up to one week, provided that the culture flasks were coated with collagen. Cell growth rates can often be improved by surface coating with attachment factors such as fibronectin, collagen, gelatine or poly-lysine. With a collagen coating, survival time of e.g. hepatocytes can be extended from one week for up to four weeks.
Storage: 2 – 8° C

| | | |
|------------|----------------|-----------|
| Collagen A | 1 x (6 x 5 ml) | P06-20030 |
|------------|----------------|-----------|

Collagen R (type I)

0.2 % sterile solution
Type 1 rat tail collagen; 2 mg/ml in 0.1 % acetic acid. Excellent substrate for the culture of hepatocytes, fibroblasts and epithelial cells.

0.4 % sterile solution
Type 1 rat tail collagen; 4 mg/ml in 0.1 % acetic acid. Excellent substrate for the culture of hepatocytes, fibroblasts and epithelial cells.

| | | |
|-----------------------------------|-----------------|------------------------|
| Collagen R 0.2 % sterile solution | 20 ml 100 ml | P06-20166 P06-20100 |
| Collagen R 0.4 % sterile solution | 20 ml | P06-20020 |

Gelatine Solution

Description
The gelatine solution is used for coating cell culture dishes. It is applied in adherent cell cultures working with e.g. endothelial cells or ES-cells.

| | | |
|---|--------|-----------|
| Gelatine solution 0.1 % in PBS ⁽¹⁾ | 500 ml | P06-20410 |
| Gelatine solution 2 % in PBS ⁽²⁾ | 100 ml | P06-25200 |

(1) usually on stock, (2) minimum order 10 l, (3) available upon request



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Laminin Mouse

Description
This highly purified preparation of mouse Laminin I increases cell adhesion, migration, growth, and differentiation. It is composed of 111 chains with a total MW of 800 kD and is used for the coating of culture dishes.

Source:
Murine Engelbreth-Holm-Swarm (EHS) tumor

Storage Buffer:
Dulbecco's Modified Eagle's Medium with 10 µg/ml gentamycin sulfate

Storage:
Store at -20° C or at -80° C in a manual defrost freezer

Purity:
Purity > 90 % by SDS-PAGE

Specifications

Functional assays

- Supports the formation of neuronal filaments of NG108-15 cells in a neurite outgrowth assay

Sterility testing

- No bacterial or fungal growth detected after incubation at 37° C for 14 days following USP XXIV, Chapter 71 sterility testing
- No mycoplasma contamination detected by PCR
- Endotoxin concentrations < 20 EU/ml by LAL assay

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|--------------------|------|-----------|
| Laminin from mouse | 1 mg | P06-20501 |
|--------------------|------|-----------|

Fibronectin

Description
Fibronectin is a large glycoprotein widely distributed in soluble form in the plasma and body fluids. Many cell types synthesize fibronectin. There is also an insoluble form of fibronectin in tissues. Plasma fibronectin is not identical to cellular fibronectin but equally effective in supporting cell attachment. Fibronectin promotes the attachment and spreading of many adherent cells on plastic, but also mediates binding to other extracellular matrix components such as e.g. collagen.

Preparation
Fibronectin is purified from human plasma; donors tested negative for anti-HIV antibodies and HBs antigen.

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|----------------------|---------|----------|
| Fibronectin | 5 mg | 2705005 |
| Fibronectin solution | 1 mg/ml | 2705001S |

Coating procedure
The recommended working concentration is 0.05 - 10 µg/cm² of growth surface (0.05 - 10 µg/ml) depending on cell type.

a. Thaw stock solution on ice for several hours. Place plates on ice and pre-chill pipette tips. Distribute the solution to completely cover the bottom of the wells.

b. The following table gives suggested volume required per well:

| Plate Type | Volume Laminin per Well |
|-------------------------|-------------------------|
| 6 wells (or 35 mm dish) | 1 ml |
| 24 wells | 200 µl |
| 48 wells | 50 µl |
| 96 wells | 20 µl |

c. Incubate the plates at 37° C for 1 hour. In the laminar flow hood, remove excess liquid from the wells of the tissue culture plate.

Rinse the wells once with tissue culture medium and then add your cells.

Reconstitution
Dissolve contents in sterile water. Prepare a 1 mg/ml solution by gently warming the vial to 37° C; do not agitate, this may cause precipitation.

Recommended amount for coating
For coating of cell culture vessels 1-5 µg/cm² is used.

Storage
Lyophilisate can be stored at 2-8° C; solution stored at -20° C in aliquots.

