

Product Name

Name: mSRCgel™ Extracellular Matrix
extracted from EHS mouse sarcoma
[-] Phenol Red

Cat. No.: C2011-0005, C2011-0010

Size: 5 mL, 10 mL

Product Description

The extracellular matrix (ECM) can provide structural support for cells and tissues in a dynamic three-dimensional network of macromolecules. The ECM is a molecular network which holds bioactive molecules and growth factors together. It is of vital importance that it controls the basal behaviors and characteristics of cells such as adhesion, migration, polarity, differentiation, proliferation, and apoptosis.

mSRCgel is a natural basement membrane (BM) extracted from **mouse sarcoma** cells, a type of connective tissue tumor. mSRCgel is high in ECM proteins, including laminin, collagen IV, heparan sulfate proteoglycan (perlecan), entactin, and many essential growth factors.

Application

It is suitable for research in cell growth, differentiation, morphological study, cytochemical function, and cell invasion.

Protein concentration: 8 - 13 mg/mL

Storage and Stability

The product should be kept at **-20°C**. Avoid multiple freeze-thaws.

The product is **light-sensitive** and therefore should not be left in the light.

Shelf life: Stable until expiry date on the label

Procedure

During the thawing process, store mSRCgel Extracellular Matrix at 2 - 8°C overnight. Thawed mSRCgel solidifies quickly above 15°C; when ready to use mSRCgel, keep it on ice to prevent untimely gelling.

Different thicknesses and concentrations are suited to different applications of mSRCgel. A thick gel is needed for applications such as endothelial cell formation of capillary-like structures, epithelial organoid formation, or tumor organoid formation. Some applications require a thin layer coating but not a thick gel, such as propagation of primary cells.

Thick Gel Method:

1. Thaw mSRCgel as described above.
2. Slowly pipet up and down to mix the mSRCgel solution well, and do not introduce air bubbles.
3. Pipette 200 - 300 $\mu\text{L}/\text{cm}^2$ of the mSRCgel solution onto the growth surface.
4. Place the coated plate at 37°C for 30 minutes to solidify.
5. Coated plates are ready for use.

Thin Layer Method (non-gelling):

1. Thaw mSRCgel as described above.
2. Slowly pipet up and down to mix the mSRCgel the solution, do not to introduce air bubbles.
3. Dilute mSRCgel to a desired concentration in a cold serum-free medium. A 1:100 dilution is suitable for propagating primary cells. An appropriate concentration should be tested out according to the application.
4. Cover the growth surface area with enough solution. Generally, a volume of 300 $\mu\text{L}/\text{cm}^2$ is suitable.
5. Incubate the coated plate at room temperature for one hour.
6. Aspirate the coating solution and plate cells at once. Prevent the coated surface from drying out.

Angiogenesis assay

1. Equipment, reagents and consumables

- 1.1 Equipment: Biosafety cabinet, pipette, carbon dioxide incubator, inverted microscope, centrifuge (low-speed).
- 1.2 Reagents: DMEM, ECM complete medium, trypsin solution, 1 \times PBS solution.
- 1.3 Consumables: sterile pipette tips, 96-well cell culture plate, sterile 1.5 ml microcentrifuge tubes and other consumables. (And any others according to the experimental design).

2. Experimental methods

2.1 Preparation before Experiment

- 1) Put the mSRCgel in the ice box and put it (in the refrigerator) at 4°C, so the mSRCgel can slowly melt overnight Do not allow this product to warm up above 4°C during manipulation. Keep the product on ice and dilute using ice-cold solutions or cell suspensions.
- 2) Consumables or reagents that come into contact with mSRCgel, such as sterile centrifuge tube, sterile pipette tips and DMEM, should be pre-cooled at 4°C in advance.

2.2 Plate coating procedure

- 1) Place the microcentrifuge tubes on ice. Add each component according to the following table.

Dilution (mSRCgel: DMEM)	1:0	2:1	1:1
mSRCgel (μL)	100	80	60
DMEM (μL)	0	40	60

Note: Dilution at 2:1 is appropriate in most case.

- 2) After mixing the above components well, add 50 μL /well of the diluted mSRCgel solution to the 96-well plate, (two repeated wells are recommended), mark the information and the date, and allow to solidify in a 37°C incubator for at least 1 hour.

2.3 Seeding the cells

- 1) Digest HUVEC with pancreatic enzyme. Adjust the cell density to 4×10^5 cells/mL by using ECM medium.
- 2) Take the coated 96-well plate and add the cell suspension in a volume of 50 μL per well (the final number of HUVEC is 2×10^4 cells).
- 3) Observe the results of vascular structures after 4 h and 24 h incubation in a carbon dioxide incubator at 37°C.

Quality Control

mSRCgel Extracellular Matrix is tested for the presence of bacteria, fungi, and mycoplasma. In addition, osmolality, protein concentration, endotoxin, gel stability, and biological activity are tested.

Precaution and Disclaimer

For research use only, not for clinical diagnosis, and treatment.