

Mouse Endothelial Cell (mEC) Complete Media and Reagents

mEC Complete Media***

200mL DMEM media (Low Glucose) – ([Gibco 11885-092, Thermo Fisher](#))

200mL Ham's F-12 media – ([Corning 10-080-CV, VWR](#))

100mL Heat Inactivated FBS – ([FB5002, Thomas Scientific](#))

10mL 1M HEPES - ([Corning 25-060-CI, VWR](#))

5mL 100X Pen/Strep/Fungizone – ([Corning 30-004-CI, VWR](#))

5mL 100X MEM Nonessential Amino Acids – ([Corning 25-025-CI, VWR](#))

5mL 10 mg/mL Heparin (Sodium Salt) Stock** – ([H3149-100KU, Millipore-Sigma](#))

5mL Endothelial Cell Growth Supplement* (ECGS, 50µg/mL final conc.) – ([Corning 356006, VWR](#))

* To add ECGS, resuspend bottle (100mg) in 20mL DMEM:F-12/FBS and add back 5 mL to media (i.e. one lyophilized bottle/2L media)

** To make Heparin Stock; resuspend to stock concentration in 1:1 DMEM:F-12 media, 0.22µm filter and store at -20°C (100KU in 50mL volume for 10mg/mL)

***Complete mEC media should be 0.22µm filtered and stored at 4°C for up to 1 month

Accutase Cell Detachment Solution ([423201, Biolegend](#))

Fibronectin ([F0895-2MG, Millipore-Sigma](#))

To coat plates, dilute fibronectin 1:1000 in 1X PBS and add enough to each dish to cover the bottom of the plate. Incubate at room temp for 20 minutes. When you are ready to plate cells, aspirate the fibronectin (don't wash) and add cells directly.

- 0.5mL for a 12-well
- 1-2mL for a 6-well
- 3mL for a T25
- 5mL for a T75