

# Regenix™ Heart

## PROTOCOL

**Regenix™ Heart** is composed of various basement membrane proteins separated from heart tissues. Regenix™ Heart can be utilized for two-dimensional (2D) and three-dimensional (3D) culture of cardiomyocytes. In particular, Regenix™ Heart can provide an optimized environment for cardiac spheroids composed of pluripotent stem cells (PSC)-derived and directly reprogrammed cardiomyocytes.

### PROCEDURE

#### 3D culture of cardiac spheroid using Regenix™ Heart

- 01

Thaw Regenix™ Heart for at least 4 hours by submerging the vial in an ice bucket and storing it in a 4°C refrigerator before use. Avoid multiple freeze/thaw cycles.
- 02

Mix Regenix™ Heart by slowly pipetting; Be careful not to create air bubbles during this process.

**Note** Regenix™ Heart may form a gel at temperature above 10°C. The temperature must be lowered to between 4°C and 8°C throughout all handling processes to ensure depolymerization.
- 03

Add Regenix™ Heart to the cell pellet or spheroids and resuspend evenly by slow pipetting.

**Note** It is recommended to remove as much of the supernatant as possible before adding the Regenix™ Heart.
- 04

Dispense 30-40 µL of the mixture to each well of a 48-well plate, and then incubate at 37 °C for 40 mins.

**Note** For suspension cultures, dispense Regenix™ Heart droplets on a sheet of Parafilm for easy detachment after gelation.
- 05

Add an appropriate volume of medium very slowly.

**Note** If you need to add 300 µL per well medium to each well, add the medium slowly and carefully over 15 seconds.

**Note** For suspension cultures, solidified Regenix™ Heart droplets are removed by creating a gentle flow of medium over the parafilm to move the droplets into the 24-well plates.