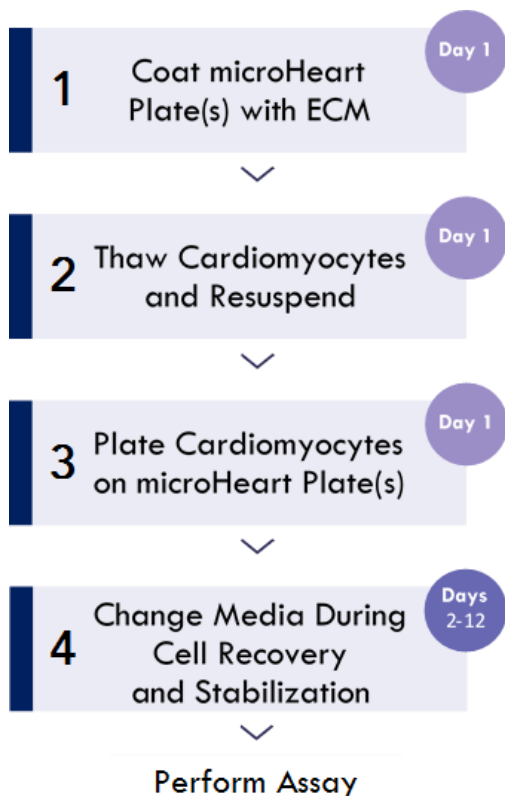


Instructions For Use: StemoniX® microHeart® Cell Ready 96-Well Plate

StemoniX® microHeart® Cell Ready 96-Well Plates are micro-engineered to enable passive self-alignment of cardiomyocytes into physiologically relevant structures that emulate correct cardiac muscle fiber organization. Alignment with microHeart plates accelerates features of human induced pluripotent stem cell (hiPSC)-derived cardiomyocyte maturity, such as improved sarcomeric organization, correct targeting of distal cardiac intercalated disc components, enhanced gene expression profiles, and faster beating rate.

Instructions Overview



Items Included With Shipment

- microHeart Cell Ready 96-Well Plate(s) (StemoniX #MHCRX-AA-0096)

Storage

- Store at room temperature.
- Once removed from the protective sterile pouch, use plates within 30 days for optimal results.

Recommended Reagents

- A coating of Extra-Cellular Matrix (ECM) (refer to cell provider's recommendation)
- Cardiomyocyte Plating Medium (refer to cell provider's instructions)
- Cardiomyocyte Maintenance Medium (refer to cell provider's instructions)
- 0.4% (w/v) Trypan Blue Solution (Corning Catalog #25-900-CI)

StemoniX microHeart Cell Ready 96-Well Plates are intended for research use only. Recipients are responsible for the safe storage, handling, and use of StemoniX microHeart Cell Ready 96-Well Plates. StemoniX is not liable for any damages or injuries arising from the receipt and/or use of this product.

Preparation and Use of StemoniX microHeart Cell Ready 96-Well Plates

Read all instructions before proceeding. Refer to the Flow Chart on Page 1 for an overview of the 4 step plate preparation process.

Step 1: Coat microHeart Plate(s) with Extra-Cellular Matrix (ECM)

- 1.1 It is highly recommended to coat microHeart plates with an ECM before plating cells. Refer to the cell provider's instructions for appropriate ECM, and follow vendor instructions for coating.

Step 2: Thaw Cardiomyocytes and Resuspend in Plating Medium

- 2.1 Thaw cardiomyocytes and resuspend in plating medium, as recommended by the cell provider. Count viable cells using the trypan blue exclusion method. Dilute the live cell suspension with plating medium to achieve the plating cell density recommended by the cell provider as a reference. Functional assays (e.g., Calcium Flux) tend to require a slightly higher cell density than imaging.

NOTE: Due to the increased surface area of the wells in StemoniX microHeart plates, an initial optimization of seeding density is highly recommended. For reference, the surface area of a well in a microHeart Cell Ready 96-Well Plate is approximately 0.16 cm².

Step 3: Plate Cardiomyocytes on microHeart Plate(s)

- 3.1 Transfer 120 µl of the adjusted cardiomyocyte suspension (step 2.1) to each well of the microHeart Cell Ready 96-Well Plate(s). Carefully shake the plate(s) in perpendicular directions for 8 to 10 seconds each, to ensure an even cell distribution.
- 3.2 Incubate the microHeart plate(s) in a cell culture incubator set to 37°C, 5% CO₂, and 95 to 98% humidity. Allow the cells to attach overnight to the surface of the microHeart plate(s).

Step 4: Change Media in the microHeart Plate(s) During Cell Recovery and Stabilization

- 4.1 On the day following cell plating, observe cell density and morphology in each plate.
NOTE: On the day following plating, cardiomyocytes may show a stressed appearance and may not display beating activity. Beating should resume within 2 to 3 medium changes.
- 4.2 Change medium according to cell provider recommendations. Then, refresh media every 2 to 3 days with cardiomyocyte maintenance medium.
NOTE: If using an automated liquid handler, set it to aspirate 80 µL of cardiomyocyte plating or maintenance medium, leaving 40 µL behind. Add 80 µL of fresh cardiomyocyte maintenance medium. If using a multichannel pipette, perform complete medium changes with 120 µL of cardiomyocyte maintenance medium. Weekend feeding can be performed with 150 µL of the cardiomyocyte media.
- 4.3 Monitor cell appearance closely to assess functionality. Refer to cell provider instructions for specific cardiomyocyte recovery and stabilization times before use. StemoniX recommends performing downstream assays no earlier than 12 days after plating.