Cardiomyocyte Selective Medium (CSM)

Catalog #5911

Product Description

Cardiomyocyte Selective Medium (CSM) is a complete medium designed for the purification of cardiomyocyte cells derived from human pluripotent stem cells (hPSC). Current protocols allow for the differentiation of hPSC to cardiomyocytes at a high efficiency, however, the derived cells are a mixed population containing non-cardiomyocyte cells. The non-cardiomyocyte cells may interfere with downstream analyses. Density-gradient centrifugation, genetic modification, and cell sorting based on cell surface markers or mitochondria dyes have been applied to enrich for the hPSC-derived cardiomyocytes. These methods, however, either involve complicated procedures or require special equipment, and are not ideal for therapeutic purposes. Based on the unique metabolic properties of fetal and hPSC-derived cardiomyocytes, which can use lactate as a major energy source, CSM is developed to enrich for the hPSC-derived cardiomyocytes using an efficient, easy, and non-invasive approach.

CSM is a serum-free medium specially designed for the selective growth of cardiomyocytes. It significantly improves the purity of hPSC-derived cardiomyocyte cell populations in 4 - 6 days, requiring only daily medium change. While culturing in the CSM, the non-cardiomyocyte cells were gradually eliminated, whereas the cardiomyocytes remain contracting in the culture.

The CSM can also be used for cardiomyocytes derived from mouse pluripotent stem cell and/or the primary cells isolated from the hearts of rat neonatal pups.

Components

CSM consists of 500 mL basal medium and 10 mL of 50X Cardiomyocyte Selective Growth Supplement (CSGS, Cat. #5962).

Product Use

CSM is for research use only. It is not approved for human or animal use, or for application in *in vitro* diagnostic procedures.

Storage

Store the basal medium and complete CSM at 4°C, and the CSGS at -20°C. Protect from light.

Shipping

Basal medium is shipped at room temperature and all other components are shipped on dry ice.

Cardiomyocyte Selective Medium Preparation

1. Warm the basal medium and 50X CSGS to room temperature. Make sure the 50X CSGS is completely dissolved before adding it to the basal medium.

2. Decontaminate the external surfaces of the medium bottle and medium supplement tube with 70% ethanol and transfer them to a sterile field.

3. Add the 50X CSGS into the basal medium using sterile techniques and mix well. The complete CSM is now ready for use.

NOTE: Store the complete CSM in the dark at 4°C, as it is light labile. We recommend warming the medium to room temperature prior to use.

Instructions for use

Before applying CSM to your differentiated cardiomyocytes for selective culturing, we recommend letting the cells grow to 70-80% confluency. If the cell density is too high, we suggest splitting the cells; if too low, allow the cells to grow until they reach 70-80% confluency.

Day 1: Warm the complete CSM to room temperature. Remove the culture medium from cells and wash them once with the complete CSM. Apply the CSM to the cells (2 ml of medium per well of a 6-well plate).

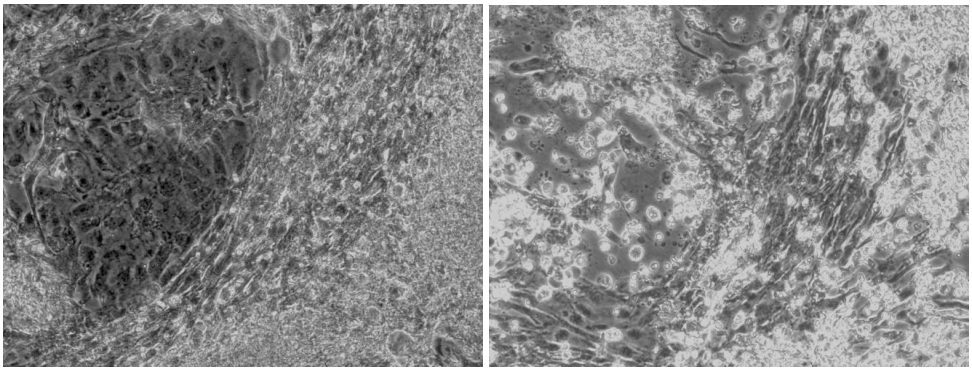
Day 2 to Day 6: Change the medium daily. Cell death is expected, however, cardiomyocytes will remain contracting in the CSM.

Day 7: After treatment with CSM for 6 days, most non-cardiomyocyte cells should be eliminated from the culture. Remove CSM from the cells and add cardiomyocyte culture medium, e.g. Cardiac Myocyte Medium (Cat. #6201), or other medium of your choice. Let the cells recover and adapt to the culture medium.

NOTE: We recommend applying the CSM for 4 - 6 days to enrich for the cardiomyocytes. If a high number of non-cardiomyocyte cells still remain in the culture after 6 days, you may continue treatment for 2 additional days. DO NOT culture the cells in the CSM beyond 10 days, as this may lead to cell death of the cardiomyocytes.

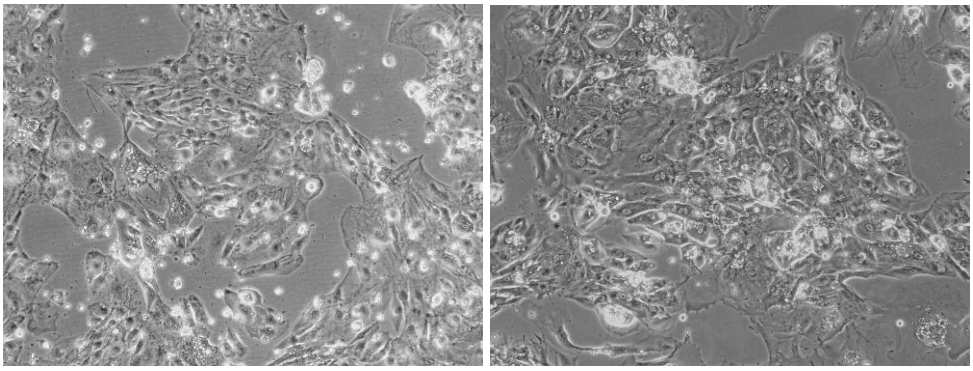
Caution: If handled improperly, some components of the medium may present a health hazard. Take appropriate precautions when handling it, including the wearing of protective clothing and eyewear. Dispose of properly.

Figure 1. CSM applied to cardiomyocytes derived from hPSC



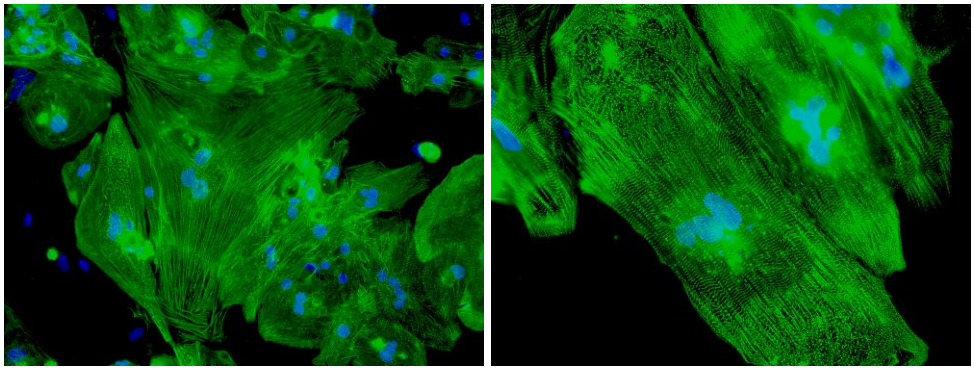
The CSM was applied to the hPSC-derived cardiomyocytes for 6 days. The majority of non-cardiomyocyte cells died during the treatment, while contracting cardiomyocytes survived. Left: Day 0 of CSM application; Right: The contracting cardiomyocytes after culturing in CSM for 6 days.

Figure 2. Cardiomyocytes were cultured in CSM for 6 days and then maintained in Cardiac Myocyte Medium.



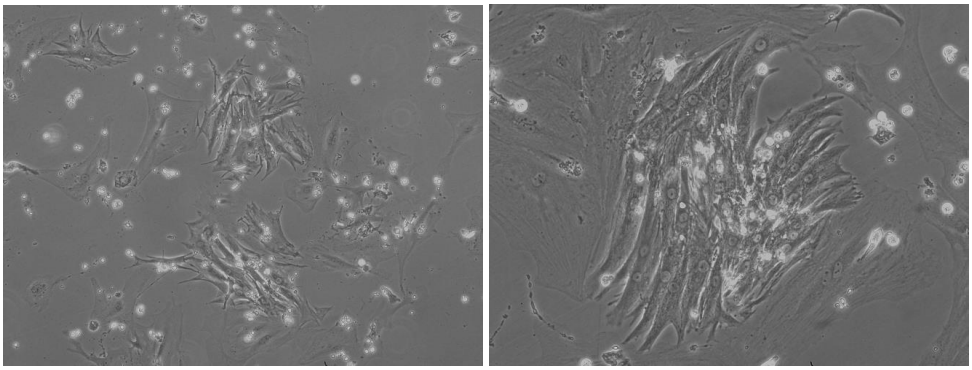
After culturing the cells for 6 days in CSM, most non-cardiomyocyte cells were eliminated leaving contracting hPSC-derived cardiomyocytes in the culture (left). Cardiomyocytes were then maintained in Cardiac Myocyte Medium (CMM, Cat. #6201).

Figure 3. Cardiomyocytes cultured in CSM and then CMM express α-actinin.



The hPSC-derived cardiomyocytes were passaged and cultured in CSM for 6 days and then recovered in CMM. After replating the cells to coverslips, the cells were fixed and immunostained for sarcomeric α-actinin (green). Nuclei were stained with DAPI (blue). Left: 200x; right: 400x

Figure 4. Rat primary cardiomyocytes purified using CSM



Rat primary cardiomyocytes were isolated from the hearts of postnatal day 8 pups and then cultured in CSM for 5 days. Most fibroblasts were eliminated from the culture, while the small contracting cell clusters or single cardiomyocytes remained. Left: 100x; right: 200x.