aceRNA Technologies Co., Ltd.

CM purifier RNA Switch™

PROTOCOL

Product name	Cat. No.	Size***	Storage
CM purifier RNA Switch™	P-0004	9 μg	-80°C

*** 0.5 μg/μL

Additional materials required

- 6-well tissue culture plates
- 1.5 mL microcentrifuge tubes (RNase/DNase free, Sterile)
- Lipofectamine MessengerMAX™ Transfection Reagent (ThermoFisher)
- puromycin
- puro resistant mRNA (P-0007)

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Cardiomyocyte purification using CM purifier RNA Switch™

The following procedure is for the transfection of cell populations containing human cardiomyocytes with CM purifier RNA Switch™ in one well of a 6-well plate. Transfection efficiency and mRNA expression can be influenced by the cell type being transfected.

RNA Switch™ is synthetic mRNA. Therefore, transfection should be performed in a RNase-free working-environment and mRNA should be diluted and aliquoted in RNase-free Water. Store them at -80°C.

1. Plate cell population containing human cardiomyocytes in a 6-well tissue culture plate at a density of 8.0 ~ 10.0 x 10^5 cells/well and incubate the cells at 37°C and 5% CO_2 24 h prior to transfection

2. 24 h after seeding, aspirate the medium and add 2.0 mL of fresh medium to wells and incubate the cells at 37°C and 5% CO_2 1 to 2 hrs before transfection.

3. Thaw CM purifier RNA Switch™ and puro resistant mRNA (P-0007) on ice or at 4°C. Transfect the cells with 0.5 μg of CM purifier RNA Switch™ and puro resistant mRNA (P-0007) with 5 μL of Lipofectamine MessengerMAX™ Transfection Reagent (ThermoFisher) according to the manual. Incubate the cells at 37°C and 5% CO_2.

4. 4 h after transfection, discard the medium and add 2 mL of medium containing puromycin (2 μg/mL) to eliminate non-transfected cells. Incubate the cells at 37°C and 5% CO_2.

5. Continue incubating the cells, changing media every 24 h.

6. 48 h after transfection, cell morphology can be observed by microscope and harvested for c-TnT FACS analysis.