

RFP Expressing Human Aortic Smooth Muscle Cells (adult)

ORDER INFORMATION

Name of Cells: RFP Expressing Human Aortic Smooth Muscle Cells (RFP-

huASMCs)

Catalogue Number: cAP-0026RFP

Product Format: Proliferating in flasks or Frozen Vials

Cell Number: > 90% confluent in T25 flask or $> 5 \times 10^5$ cells/vial

General Information:

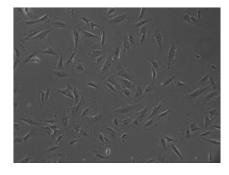
RFP-huASMCs (**cAP-0026RFP**) are Puromycin resistant selected from human adult aortic smooth muscle cells (**huASMCs**, **cAP-0026**)after transfected with RFP-lentivirues. The cells are shipped in proliferating culture with >90 confluence (the cells are provided @ passage 3-4) or in frozen vials. Smooth Muscle Cells Growth Medium (cAP-24) is recommended for the expansion of RFP-huASMCs and these cells can be propagated to extra 3-4 passages without losing their morphologic and phenotypic characteristics when cultured <u>following the detailed protocol described below</u>).

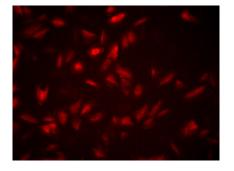
Characterization of the cells

α-Vascular SMA: > 98% positive by immunofluorescence VE-Cadherin: < 1% positive by immunofluorescence

negative for mycoplasma.

Product Use: RFP-huASMCs are for research use only.







Shipping: Proliferating culture in T25 flask or Frozen vials.

Handling of Arriving Cells

Cells in T25 Flask

When you receive the cells, leave the flask in 37°C CO2 incubator for 1 hour first, and then replace the transport medium with Smooth Muscle Cells Growth Medium. Let the cells to grow for 24 hour before subculture if the cells are not completely confluent.

Cells in frozen vials:

When you receive the frozen vials, you can keep the frozen vials in a -80°C Freezer for short term storage or in a liquid nitrogen tank for long term storage. Frozen vials should be thawed in 37°C water bath immediately before plating the cells in to 10ml of Smooth Muscle Cell Growth Medium (cAP-24) in a T25 flask, and the cells should be incubated in a 37°C CO2 incubator for overnight. The medium should be changed on the next day.

1. Subculture Protocol:

- A) Rinse the cells in T25 flask with 5ml DPBS (**Room Temperature**, **<u>RT</u>**) twice.
- B) Add 2ml of Trypsin/EDTA (<u>RT</u>) (Invitrogen Catalogue number: 25300-062) into T25 flask (make sure the whole surface of the T25 flask is covered with Trypsin/EDTA), and gently dispose the Trypsin/EDTA solution **within 10 seconds** with aspiration.
- C) Leave the T25 flask with the cells at <u>RT</u> for 1-2 minute (the cells will normally come off the surface within 1 minute, monitor the cell under microscopy).
- D) Suspend the cells with 20ml of Smooth Muscle Cells Growth Medium and then split cell suspension into 2 x T25 flasks (10ml each, and the cells are subcultured at 1:2 ratio)

2. Cell culture protocol (proliferating):

- A) Smooth Muscle Cell Growth Medium should be changed every other day.
- B) The cells normally become confluent within 5-6 days (when split at 1:2 ratio).

Although primary cells are tested pathogen-free, investigators should handle these cells with caution and treat all animal cells as potential pathogens, since no test procedure can completely guarantee the absence of infectious agents.