Applied StemCell, Inc.
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Datasheet

DR4 MEF Cells, P3, Mitomycin-C-treated (DR4 Mouse Embryonic Fibroblast Cells)

Product Information

Specifications

Catalog Number	Cells per Vial	Number of Vials
ASF-1023	4 x 10^6	1
ASF-1024	4 x 10^6	5
ASF-1025	2 x 10^6	1
ASF-1026	2 x 10^6	5

Description

MEF cells serve as feeder cells that support the growth of undifferentiated mouse or human embryonic stem cells (ESCs) and induced pluripotent stem cells (iPSCs). MEF cells are isolated from 13.5-day old mouse embryos and should be used at early passages. Before use as feeder cells, MEF cells must be mitotically inactivated by γ-irradiation or mitomycin-C treatment.

DR4 MEF cells are derived from mice that are genetically engineered with 4 drug-resistant genes, neomycin, hygromycin, puromycin and 6-thioguanine.

Background: 129P2/OlaHsd, 129S4/SvJae, BALB/c and C57BL/6

Passage

P3

Treatment

Mitomycin C

Shipping

Dry ice

Storage and Stability

Store in liquid nitrogen freezer immediately upon receipt. This product is stable for at least 6 months from the date of receiving when stored as directed.

Biosafety Level

BSL-1

Safety Precaution

PLEASE READ BEFORE HANDLING ANY FROZEN VIALS. Please wear the appropriate Personal Protection Equipment (lab coat, thermal gloves, safety goggles and a face shield) when handling the cells. Handle the frozen vials with due caution. Please be aware that the following scenario can occur: Liquid nitrogen can leak into the vials when the vials are submerged in liquid nitrogen. Upon thawing, the liquid nitrogen returns to the gas phase, resulting in a dangerous build-up of pressure within the vial. This can result in the vial exploding and expelling not only the vial contents but also the vial cap and plastic fragments of the vial.

Restricted Use

This product is for research use only and not intended for human or animal diagnostic or therapeutic uses.

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Media and Material

Medium (I)

Component	Concentration
DMEM (High Glucose)	
FBS	10%
Nonessential amino acids	0.1 mM
Sodium Pyruvate	1 mM
L-Glutamine	2 mM

Suggested plating density (II)

Dish Size	Surface Area*	Working volume	MEF per dish / well
100 mm	55 cm^2	11 - 16.5 mL	1.7 - 2.8 x 10^6
60 mm	21 cm^2	4.2 - 6.3 mL	0.65 – 1.1 x 10^6
35 mm	9 cm^2	1.8 - 2.7 mL	0.27 – 0.45 x 10^6
T25	25 cm^2	5 – 7.5 mL	0.75 - 1.25 x 10^6
T75	75 cm^2	15 – 22.5 mL	2.25 - 3.75 x 10^6
T175	175 cm^2	35 – 52 mL	5.25 - 8.75 x 10^6
6-well	9.5 cm^2	1.9 - 2.9 mL	0.29 – 0.48 x 10^6
12-well	3.8 cm^2	0.8 - 1.2 mL	0.11 – 0.19 x 10^6
24-well	1.9 cm^2	0.4 - 0.6 mL	57,000 – 95,000
48-well	0.95 cm^2	0.2 - 0.3 mL	22,500 – 47,500
96-well	0.32 cm^2	100 - 200 µL	9,600 – 16,000

**Approximate growth surface areas. Numbers can vary between plastic ware from different suppliers*

Protocol

1. Remove a vial of frozen cells from liquid nitrogen and place it onto dry ice for 5 minutes before thawing it at 37ºC water bath. As soon as the majority of the content of the vial thawed, transfer it to a conical tube containing 10x volume of pre-warmed medium.

2. Spin at 1000 rpm for 5 minutes, discard medium, resuspend the cells in growth medium and plate them at an appropriate density in a gelatin-coated tissue-culture dish (generally 25,000-50,000 cells/cm^2, Media and Material II). Optimal density is to be determined by the user for specific applications.

   Tip: make sure coating with 0.1-0.2% Gelatin solution in 37 degree incubator for at least 30 min.