



## Mouse Embryonic Fibroblast (MEF) Cells (P3)

### Cat. Numbers:

ASF-1013	ASF-1213	ASF-1223	ASF-1113
ASF-1015	ASF-1215	ASF-1225	ASF-1115

### 1. At what density should I plate the MEF cells to support hESC or mESC culture?

The suggested MEF density is  $3 - 5 \times 10^4$  cells/cm<sup>2</sup> surface area. Table 1 gives the recommended cell density for a variety of cell culture vessels.

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

**Table 1:** Approximate growth surface areas and recommended MEF cell number for cell culture. Numbers can vary between plastic ware from different suppliers.

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## 2. How many times can I passage untreated MEF cells?

To properly support ESC maintenance MEF cells should not be passaged beyond P5. It is generally recommended to use feeder cells at P3. (Our mitotically inactivated MEFs are all P3).

## 3. How long can the MEF cells be used to support ES cell pluripotency after plating?

It is best to use the MEF feeder cells within one week after plating. Do not use them after more than 10 days and passage your ES cells to a fresh feeder layer if the MEF cells are more than 2 weeks old.

## 4. What is the mouse strain background for each of your MEF products?

DR4 MEF: Mixed (129/SvJae, 129/OlaHsd, BALB/c, C57BL/6, and DBA)

Neo MEF: Mixed (129/svj/sv, C57BL/6 and CD1)

CF-1 MEF: CF-1

## 5. Are the MEF cells pathogen tested?

All of our cell products are tested for pathogens. Our standard sterility tests include Mycoplasma spp., Sendai virus, Mouse hepatitis virus, Pneumonia virus of mice, Minute virus of mice, Mouse parvovirus (MPV1, MPV2, MPV3), and Theiler's murine encephalomyelitis virus.

## 6. How should I store MEF cells?

MEF cells can be stored at -80°C for 1 to 2 months. For long term storage, MEF cells should be kept in liquid nitrogen.

## 7. Do you offer volume discount?

Yes, we do. Please email ([info@appliedstemcell.com](mailto:info@appliedstemcell.com)) or call (408-773-8007) us for details.

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