hPSC Dissociation Buffer DXF

PromoCell

Instruction Manual

Product	Size	Catalog Number
hPSC Dissociation Buffer DXF	100 ml	C-41322

Product Description

The PromoCell hPSC Disscociation Buffer DXF is suitable for routine subculture of pluripotent stem cells (PSC). It supports clump- as well as single cell passage. The formulation is non-enzymatic, chemically defined and animal-free. The gentle mode of action allows for controlled colony disaggregation, which is critical for the successful culture of undifferentiated PSC.

Instructions for Use

Additional materials needed:

- Dulbecco's PBS, w/o Ca^{++}/Mg^{++} (Order-No. C-40232)
- hPSC Growth Medium DXF (Order-No. C-28060)
- hPSC-ECM DXF (Order-No. C-43070)

Optional (for single cell passage):

- Y-27632 (Order-No. PK-CA577-1596-1)

A. Clump passage (recommended for routine culture)

1. Aspirate culture medium and wash cells twice with Dulbecco's PBS, w/o Ca^{++}/Mg^{++}.
2. Add 200 µl/cm² of hPSC Dissociation Buffer DXF and incubate for 5 - 6 minutes in the incubator at 37°C and 5% CO_2.

Note: Colonies should appear bright and shiny. The single cells on the colony borders should begin to round up.

3. Carefully aspirate and discard the Dissociation Buffer and add 1 - 5 ml of fresh complete hPSC Growth Medium DXF. Using a serological pipet, flush the colonies away from the surface. Avoid flushing more than 4 - 5 times in order to maintain cell clumps of sufficient size. If the colonies do not detach, use a cell scraper.
4. Dispense cell clumps into new ECM-coated culture vessels with fresh hPSC Growth Medium DXF.
5. Expand cells according to your routine protocol.

B. Single cell passage

1. Aspirate culture medium and wash cells twice with Dulbecco's PBS, w/o Ca^{++}/Mg^{++}.
2. Add 200 - 300 µl/cm² of hPSC Dissociation Buffer DXF and incubate for 5 - 6 minutes in the incubator at 37°C and 5% CO_2.
3. Aspirate the Dissociation Buffer and replace by 50 - 100 µl/cm_2 fresh hPSC Dissociation Buffer DXF supplemented with 10 µM Y-27632 (100 mM stock in DMSO).
4. Detach the cells by flushing the culture surface several times using a serological pipet. Triturate by pipetting up and down an additional 5 - 10 times.
5. Spin the single cell suspension (5 min, 200 x g, room temperature).
6. Aspirate and discard the supernatant and resuspend the cells in fresh hPSC Growth Medium DXF supplemented with 10 µM Y-27632.

Note: Alternatively, resuspend the cells in a buffer of choice and use them for your experiments, e.g. immunostaining for flow cytometry analysis.

7. Plate the cells in an ECM-coated vessel and expand cells according to your routine protocol. Do not use Y-27632 for more than 24 hours after plating.

Storage and Stability

Store at 2 - 8°C immediately after arrival. If stored properly, the product is stable until the expiry date stated on the label.

Quality Control

All lots of PromoCell Cell hPSC Dissociation Buffer DXF are subjected to comprehensive quality control tests. Each lot is routinely tested with human iPS cells for function, absence of cytotoxicity, and physical parameters such as osmolarity and pH. Approved in-house lots are used as a reference.
In addition, all lots have been tested for the absence of microbial contaminants (fungi, bacteria).

Intended Use

This product is for *in vitro* research use only and not for diagnostic or therapeutic procedures. For safety precautions please see appropriate MSDS.

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