

### **Datasheet**

# Stempan DMEM

# **Complete Medium for Embryonic Stem Cells**

Product	Description	Catalogue-No.	Size
Stempan DMEM	Complete medium for embryonic stem cells	P08-50500	500 ml

#### **Product description**

Stempan is a medium for in vitro cultures of mouse embryonic stem cells in which an undifferentiated state of the cells should be maintained. Additional use of leukaemia inhibitory factor (LIF) is necessary.

#### Storage conditions

Storage: 2-8°C

Stability: 6 month from date of production Size: 500 ml, other sizes on request

#### Composition

Stempan consists of a balanced mixture of salts, amino acids, vitamins, trace elements, hormones and is supplemented with selected fetal bovine serum (FBS). All ingredients of Stempan are tested for ES-culture-suitability before manufacturing the complete medium.

#### Suitability

Cultivation of mouse embryonic stem cells while maintaining an undifferentiated state of the cells. FOR RESEARCH USE ONLY!

Not approved for human or animal diagnostic or therapeutic procedures.

#### Special advantages

Mouse embryonic stem cells (ES-cells) are pluripotent cells. They can be retrieved from blastocysts and transferred into a permanent cell culture. Cultivation of ES-cells with Stempan could be done on fibroblast feeder layers or coated culture plates.

#### Instructions for use

Primary embryonic fibroblasts are generally used as feeder-cell-layers. The fibroblasts cell type STO in combination with LIF (leukaemia inhibitory factor) could be used as an alternative whereas there is the disadvantage of a shortened cell lifetime of primary fibroblasts. Before the cells could be used as feeder-layer-cells they must be inactivated by Mitomycin C or gamma irradiation (50 Gy).

ES-cells are fast growing cells which should be kept in optimal cell density to prevent cell differentiation. As the cells are clustered tightly within the cell colonies the differentiation of single cells is nearly impossible.

The cell culture should be supplied with new medium every day and subcultivated every 2-3 days. ES-cells should be kept in high-density growth conditions and converted at a confluence of 70-80%. Cells in large colonies have the tendency to differentiate.



Attention: Stempan must be supplemented with LIF (Leukaemia inhibitory factor, 10ng/ml) to avoid cell differentiation.

## Coating of culture plates

Culture plates should be filled with sterile gelatine solution (0.1%) in order to coat the whole surface area of the culture plate. Alternatively use feeder cell layer.

#### Preparation of medium

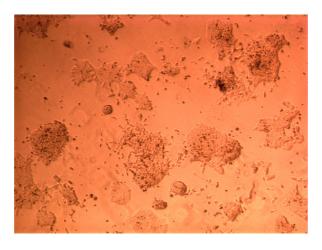
After warming Stempan to 37°C add LIF in a concentration of 10 ng/ml.

#### Detachment of ES-cells

Remove medium from 70% confluent ES-Cell culture. Rinse cell layer with DPBS (without Mg<sup>2+</sup>/Ca<sup>2+</sup>) and cover cell layer with trypsin/EDTA solution (0,05%/0,02%). Remove trypsin/EDTA after 2-3 minutes. Observe detachment of cells with a microscope. Resuspend ES-cells in Stempan after complete detachment from culture plates (serum inactivates residual trypsin) and pipette vigorously to dissolve clustered cells (but not to single cells).

#### Cell splitting

Fill gelatine coated culture plates with prepared Stempan and add cell suspension. Split ratio should be adjusted between 1:4 to 1:8, depending on growth rates. Incubate cells at 37°C and 5% CO2 in an appropriate incubator.



Mouse cells in Stempan

#### **Technical Support**

For technical support, questions or remarks please contact your local PAN-Biotech partner or the technical department of PAN-Biotech via email (info@pan-biotech.com) or phone +49-8543-601630.