

# Datasheet

# **Freezing Medium**

# **Cell Culture Reagent**

Product	Description	Catalogue-No.	Size
Freezing Medium	Cryopreservation medium, with 10 % DMSO	P07-90010 P07-90050	10 ml 50 ml

## Product description

Freezing Medium is a cryopreservation medium for animal and human cells (adherent and suspension cells). It is supplemented with fetal bovine serum (FBS) and dimethyl sulfoxide (DMSO). This composition guarantees a high survival rate and excellent cell growth after thawing.

## Storage conditions

Storage conditions:	- 20°C
Stability:	2 years from date of production
Filling:	10 ml, 50 ml, other sizes on request

# Composition

Freezing Medium is based on DMEM and is supplemented with FBS and 10 % DMSO.

# Suitability

Freezing Medium is for the cryopreservation of human and animal cells.

## **Special Advantages**

Due to its composition with FBS and 10 % DMSO it guarantees high survival rate and high cell viability after the thawing process.

## Instructions for Use

- 1. <u>Freezing cells with Freezing Medium</u>
- For optimal results only vital cells in the log-growth phase should be used.
- Thaw Freezing Medium and store till using at 2-8°C.
- Trypsinate adherent cells, transfer the cells into the culture medium and centrifuge.
- Discard the supernatant and wash the cell pellet in PBS (without Ca<sup>2+/</sup>Mg<sup>2+</sup>).
- After an additional centrifugation step (100 200 g, 5 10 minutes) transfer the cells into PBS and determine cell number and cell vitality by trypan blue cell viability staining.
- Centrifuge the cells again and discard the supernatant.
- Transfer the cells into the cold Freezing Medium (5x10<sup>5</sup> 2x10<sup>6</sup> cells/ml Freezing Medium).
- Suspend the cells carefully mixing the suspension by repeated pipetting until there are no more cell clumps.
- Refill the cell suspension into labelled cryotubes (0,5 1,5ml/tube).
- Freeze the cells in an automatically or manually controlled cryo freezer. The optimal freezing rate is approximately 1°C/minute.
- Alternatively, put the tubes for 15 minutes into a refrigerator, so that the freezing medium can penetrate into the cells. After this step freeze the tubes at -20°C for 2 hours box and put them into the vapour phases of liquid nitrogen over night.
- Store cryotubes in a cryotank with liquid nitrogen.
- 2. Thawing cells



- Remove the cryotubes from the cryotank and thaw them as soon as possible in warm water (< 1 minute).
- Disinfect the exterior of the cryo tubes with alcohol, convict the cells under sterile conditions into a centrifuge tube with 10 ml growth medium and mix it carefully.
- Centrifuge the cells (150 200 g, 5 10 minutes).
- Discard the supernatant and recover the cells into the designated culture medium. Determine the cell viability by an appropriate method, like FACS or trypan blue cell viability staining.

## **Technical support**

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

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