

Cryopreservation medium **LaboBanker**

Serum-containing Medium	Package	Code No.
LaboBanker 1	100 ml x 1 20 ml x 4	BLB-1 BLB-1S
Serum-free Medium		
LaboBanker 2	100 ml x 1 20 ml x 4	BLB-2 BLB-2S

1 Product Description

Series of Labobanker are cryopreservation media which are ready-to-use and complete with no additives. Labobankers can cryopreserve many kinds of cultured cells with minimal damage from freezing and thawing steps. Suspended cells in Labobankers can be directly frozen in -80°C , without store in -20°C freezer. Labobanker 1 contains 10%DMSO and serum, which are recommended to cryopreserve the cells cultured in serum-containing medium. Labonaker 2 contains 10%DMSO but serum-free, which are recommended to cryopreserve the cells cultured in serum-free medium or Sf9 cells cultured in serum-containing medium.

2 Storage and Stability

Labobanker 1 and 2 should be stored at 2 to 8°C . Before unsealing, they can be stored at -20°C . To prevent from deterioration, frequent freeze-thaw should be avoided.

3 Quality Control

3.1 Sterility	Sterile
3.2 Mycoplasma	Not detected
3.3 pH	7.0 - 8.5 (Room temperature)
3.4 Endotoxin	< 20 EU/ml
3.5 Performance test	> 80% (Jurkat cells, CHO cells)

4 Product Use

4.1 Freezing Protocol

1. Remove the medium from the culture dish and wash the cells by PBS.
2. After removing PBS from the culture dish, add Trypsin/EDTA-PBS and incubate the cells for 5min at RT.
3. Add the new medium with FBS to the culture dish and gently isolate the cells from the culture dish by pipetting.
4. Transfer the cell suspension to centrifuge tube and count the cell numbers.
5. Centrifuge cell suspension at approximately 200 to 300 x g for 5 minutes and remove the supernatant.
6. Resuspend the pellet in cold (2 to 8°C) Labobanker at 5×10^5 to 5×10^6 cells/ml.
7. Divide this cell suspension into cryotubes at 0.5 to 1.0 ml/tube.
8. Leave the cell in the cryotubes on ice for 5 minutes.
9. Directly freeze the cell in a deep freezer at -80°C .
10. In case of long-term presevation, transfer the frozen cells at -80°C into a liquid nitrogen (-196°C) on the following day.

4.2 Thawing Protocol

1. Set the water bath at 37 °C (or culture temperature) and the centrifuge at room temperature.
2. Prepare 10 times volume of new culture media as the frozen cell suspension for washing the thawed cells and another new culture media for cell culture. Place them in a 37°C (or culture temperature) water bath.
3. Warm the frozen cells in the water bath rapidly until ice is completely thawed.

Table 1 Guidelines for Temperature and Required Time

Water Temperature (= Culture Temperature)	Volume of Cell/Labobanker mixture	Required time for thawing
27°C (for Sf9 cells)	0.5 ml 1.0 ml	2 min 00 s - 2 min 20 s 2 min 15 s - 2 min 35 s
37°C (for Mammalian cells)	0.5 ml 1.0 ml	1 min 40 s - 2 min 00 s 1 min 55 s - 2 min 10 s

4. Transfer the thawed cells into the warm media and mix gently. And leave it for 1 to 2 min at room temperature.
5. Centrifuge it at room temperature at approximately 100 to 200 x g for 5 minutes and remove the supernatant.
6. Gently resuspend the cells using warm media and transfer to culture dish.
7. Culture the cells into the recommended environment.

5 Caution

1. For research use only.
2. Don't preserve cells to use for medical care.
3. Don't use these products to preserve the human body.
4. Perform a prior validation study with your cells.

Provider



JUJI FIELD INC.

NihonbashiHoncho 1-Chome Bldg. 4F, 1-9-13
NihonbashiHoncho, Chuou-ku, Tokyo, 103-0023, Japan
TEL : +81-3-3516-0700
FAX : +81-3-3516-0701
E-mail : info@juji-field.co.jp

Manufacturer

KURABO
KURABO INDUSTRIES LTD.

Advanced Technology Division, Bio-medical Department