



Cryopreservation

Cell Cryopreservation with CELLBANKER[®] 2

CELLBANKER 2 (Cat. # CB031) is a serum-free cell cryopreservation medium for mammalian cultured cells (see Table 1 on page 2 for cell types). This formulation minimizes batch-to-batch variability and facilitates stable cryopreservation and high cell viability after thawing.

Freezing

For optimal results, cells for cryopreservation should be healthy and growing in log phase. Standard freezing procedures may be substituted.

1. Examine and ensure that the cell culture is free of contamination, the cells are healthy, and the cells are at an appropriate density.
2. If necessary, detach the cells from the plate using standard procedures (e.g., scraping, trypsin). Perform a cell count to determine the viability and cell number.
3. Gently pellet cells by centrifugation at 1,000-2,000 rpm for 3-5 minutes at 4°C. Aspirate the supernatant.
4. Gently suspend the cells with CELLBANKER 2 cryopreservation medium; use 1 ml for 5×10^5 - 5×10^6 cells.
5. Dispense 1 ml aliquots of the cell suspension in cryopreservation vials that have been labeled with the cell line name, cell concentration, passage date, and other essential information.
6. Place the vials directly at -80°C for storage. If necessary, transfer the frozen vials to a liquid nitrogen storage tank after the vials have been frozen for at least 24 hours.

Thawing

1. Remove the frozen vial from storage and quickly thaw in a 37°C water bath.
2. Immediately mix 1 ml of cells with 10 ml of complete cell culture medium.
3. Gently pellet the cells by centrifugation at 1,000-2,000 rpm for 3-5 min. at 4°C. Aspirate the supernatant.
4. Gently suspend the cells with an appropriate volume of complete cell culture medium and plate in a culture flask.
5. Culture the cells according to standard protocols.

Table 1. Cell Types and Cell Lines Known to be Compatible with CELLBANKER 2

Cell type	Description
293	Sheared human Ad5 DNA-transformed cell line
293T	Human cell line expressing SV40 large T antigen
32D	Murine myeloid cell
3LL	Murine Lewis lung cancer cell
A10	Thoracic aortic smooth muscle of an embryonic rat
A2781	Glioblastoma cell
Ac2F	Rat liver cell
AtT20	Murine anterior pituitary cell
Ba/F3	Murine Pro-B cell
BHK-21	Baby hamster renal fibroblast cell
C2C12	Mouse myoblast cell
C3H10T1/2	Murine embryonic fibroblast cell
Caco-2	Human colonic adenocarcinoma cell
CHL/1U	Neonatal Chinese hamster lung cell
CHO	Chinese hamster ovary cell
CHO-K1	Chinese hamster ovary cell
Colo203	Human colonic adenocarcinoma cell
COS1	African green monkey kidney cell
COS7	African green monkey kidney cell
CTL2-2	Murine T-cell
DLP-1	Human colonic adenocarcinoma cell
DT40	Chicken B lymphocyte cell
DU145	Human prostate carcinoma cell
EBV-transformed B cells	
EJ-1	Human bladder carcinoma
ELM-D	Murine erythroblastic leukemia cell
HeLa	Human uterine cervical carcinoma cell
Hep3B	Human hepatoma cell
Hepal-6	Murine hepatoma cell
Hepatocytes	
HepG2	Hepatocellular carcinoma cell
HL-60	Human promyelocytic leukemia cell
HT-2	Murine cell
Huh-7	Human hepatocarcinoma cell
JM404	
Jurkat	Human T-cell line
K-562	Human chronic myelogenous leukaemia cell
Kato-III	Human stomach carcinoma
Keratinocytes	
KM12-LX	Human chronic myelogenous leukemia cell
L5178Y	Murine lymphoma cell
L929	Murine fibroblast cell
LM	Murine fibroblast cell
LNCap	Human prostate adenocarcinoma cell
Lymphocytes	
MCF-7	Human metastatic mammary carcinoma cell
MDCK	Madin-Darby canine kidney cell
Microvascular Endothelial Cells	
Molt-4	Human T-cell
NB-1/GOTO	Human neuroblastoma
NCI-H441	Human lung adenocarcinoma epithelial cell
NIH3T3	Mouse embryonic fibroblast cell
P388	Murine leukemia cell
P3/x63-Ag8.U1	Murine myeloma cell
PC12	Rat pheochromocytoma cell
Periodontal ligament fibroblasts	
Periodontal ligament membrane fibroblast cells	
Raji	Human B-cell line
RAW264.7	Murine macrophage cell
Sf9	Insect cell
SK-N-MC	Neuroepithelioma cell
SN12C	Renal carcinoma cell, Subclone: PM6,Clone8, MM3
Stomach carcinoma cells	
THle3	Human liver cell
T24	Human bladder carcinoma
Vero	African green monkey kidney epithelial cell
WEHI3B	Murine myeloid leukemia
WiDr	Human colon adenocarcinoma cell