Cryopreserve cells

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Abstract

Cryofreezing cells for preservation.

Citation: Bao Thai Cryopreserve cells. **protocols.io**
dx.doi.org/10.17504/protocols.io.mtsc6ne
Published: 25 Jan 2018

Protocol

Step 1.

Grow cells to 100% confluency in a 10cm dish, which can make 5 cryotubes with 1 mL each.

Step 2.
Trypsinize cells (1.5 mL trypsin in a 10mm plate).

Step 3.
Incubate at 37C for 5 minutes.

Step 4.
Resuspend cells completely off the plate and add 5mL of DMEM media containg FBS. Transfer to a 14 mL tube.

Step 5.
Centrifuge tubes at 100xg for 15 minutes.

Step 6.
Decant supernatant carefully.

Step 7.
Resuspend cell pellets completely in 5 mL of DMEM containing 5% DMSO.

Step 8.

Add 1 mL of the solution containing cells into each cryogenic tube.

Step 9.
Freeze cells slowly by putting the tubes in between tightly sealed styrofoam boxes at -80C for 2 days.

Step 10.
Transferred tubes to liquid nitrogen for long term storage.