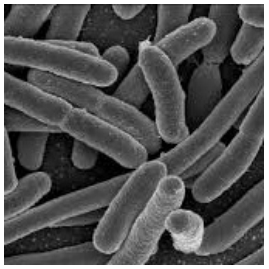
Jun 23, 2017

Bacterial Cryopreservation



DOI
https://dx.doi.org/10.17504/protocols.io.ihncb5e

Dr. Steven Wilhelm

The Aquatic Microbial E...

Steven W W Wilhelm
The University of Tennessee, Knoxville

DOI: https://dx.doi.org/10.17504/protocols.io.ihncb5e

Protocol Citation: Dr. Steven Wilhelm 2017. Bacterial Cryopreservation. **protocols.io** https://dx.doi.org/10.17504/protocols.io.ihncb5e

Protocol status: Working

Created: June 16, 2017

Last Modified: November 09, 2017

Protocol Integer ID: 6414

Keywords: bacterial cryopreservation

Abstract

Please contact Dr. Steven Wilhelm (wilhelm@utk.edu) for additional information regarding this protocol.

Preparation

1 Take a dense culture and transfer a volume of cells (usually 20 mL) into a sterile centrifuge tube

2 Spin down the culture at 4,500 rpm, 10 min
00:10:00

3 Decant supernatant into bacterial waste

4 Resuspend pellet into 2 mL of the appropriate growth medium

5 Vortex 10 sec

6 Split the 2 mL into two cryogenic vials by adding 1 mL to each vial

7 Invert to mix several times

8 Immediately place into -80ºC freezer

Transferring Cells

9 Remove cryogenic vials one at a time from the freezer

10 Using a sterile pipette tip, scrape a small amount of frozen cells and place into 20 mL sterile growth medium. Slowly move the cells to the appopriate growth conditions.
20 µL

11 Place cryogenic vial immediately back into the freezer