

iGEM 2016 – SDU

Title: Bacterial freezing stock

Date issued: 2012.10.26

SOP number: SOP0005_v02

Review date: 2016.04.28

Version number: 02

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1. Purpose

To freeze new strains of bacteria in -80°C

2. Area of application

This procedure is valid for all *E. coli* strains

3. Apparatus and equipment

| Apparatus/equipment | Location (Room number) | Check points | Criteria for approval/rejection |
|---------------------|--|--------------|---------------------------------|
| Freezer (-80 °C) | Laboratory (class 1) – V18-404b-0 Laboratory (class 2) – V15-501b-2 | | -75 °C - -80 °C |
| Pipette p1000 | | | |
| 15 ml tube rack | Hallway storage (1. Floor) | | |
| Vortex | | | |

4. Materials and reagents – their shelf life and risk labelling

| Name | Components (Concentrations) | Manufacturer / Cat. # | Room | Safety considerations |
|-------------------|-----------------------------|-----------------------|----------------|-----------------------|
| Glycerol | 50 % | AppliChem | Anne Mette, RT | |
| Blue pipette tips | | Contact Lab-manager | Micro storage | |
| Freezing tubes | | Contact Lab-manager | Micro storage | |

5. QC – Quality Control

6. List of other SOPs relevant to this SOP

JMJ_SOP0001 ON culture of *E.coli*

7. Environmental conditions required

8. Procedure

- 8.1 Make ON culture
- 8.2 Add 300 μ L 50 % glycerol to freezing tubes
- 8.3 Autoclave the freezing tubes (if not already)
- 8.4 Vortex ON culture of bacteria
- 8.5 Add 900 μ L ON culture to each of the 3 freezing tubes with glycerol
- 8.6 Pipette up and down to mix
- 8.7 Check register sheet for number for the cultures and write on the tubes
- 8.8 Add the tubes to your collection of strains

9. Waste handling

| Chemical name | Concentration | Type of waste (C, Z...) | Remarks |
|------------------|---------------|-------------------------|---------|
| ON Culture | | Liquid bacterial waste | |
| Once use plastic | | GMO yellow waste | |

10. Time consumption

- Total-time 2h 15 min.
- Hands-on-time 15 min.

11. Scheme of development

| Date / Initials | Version No. | Description of changes |
|--------------------|-------------|---|
| 12.10.25 / MM | 01 | The SOP has been written |
| 13.01.03 / MM & TK | 01 | The SOP has been approved |
| 16.04.28/BKB | 02 | Changes in the volume used; from 250µL glycerol to 300µL From 750µL ON to 900µL. Added the steps; 8.1 Make ON culture 8.3 Autoclave. |
| 16.04.28/ PRA | 02 | The SOP has been approved |

12. Appendixes