iGEM2015 – Microbiology – BMB – SDU	
Title: Purification of RNA	Date issued:
SOP number: SOP0005	Review date:
Version number: v01	
	Written by:

Purpose

To purify RNA from bacterial cells.

Area of application

Measurement of transcriptional activity

Apparatus and equipment

Apparatus/equipment	Location (Room number)	Check points	Criteria for
			approval/rejection
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Materials and reagents – their shelf life and risk labelling

Name	Components	Supplier / Cat. #	Room (hallway storage)	Safety considerations

QC – Quality Control

List of other SOPs relevant to this SOP

Environmental conditions required

Procedure

- 1. Collect samples
- 2. RNA purification day 1
 - a. Remove samples from freezer and keep them on ice.
 - b. Resuspend samples in 150 μl of cold solution 1
 - c. Immediately transfer 150 μ l of solution 2 (not cold)
 - d. Immediately transfer the 300 μl to 1,5 ml tubes (Phase Lock Gel) containing 600 μl phenol and 300 μl chloroform.
 - The phenol/chloroform solution can be prepared before resuspension of samples. Pellet Phase Lock Gel before use.
 - e. Mix by hand
 - f. Heat solutions for 3 min at 85° C with shake.
 - g. Place tubes 5 minutes on ice.

- h. Spin for 3 minutes (max spin).
- i. Transfer supernatant (top phase) to a new 1,5 ml eppendorf tube containing 30 μ l 3 M Na-Ac pH 4,5 and 900 μ l 96 % EtOH
 - i. When preparing the solution, use ice cold 96 % EtOH and keep the NaAc-ethanol solution on ice until use.
- j. Store at -20° C.
- 3. Day 2 Wash and determine concentration

a.

Waste handling

Chemical name	Concentration	Type of waste (C, Z)	Remarks

Time consumption

- Total-time xx hours.
- Hands-on-time x hour.

Scheme of development

Date / Initials	Version	Description of changes
	No.	
	01	The SOP has been written

Appendices