iGEM2013 – Microbiology – BMB – SDU

Title: Restriction digest **Date issued:** 2013.06.19

SOP number: SOP00012_v01 Review date: 2013.06.18

Version number: 01 Written by: ASF

1. Purpose

To digest DNA-pieces with restriction enzymes

2. Area of application

Cloning

3. Apparatus and equipment

Apparatus/equipment	Location (Room number)	Check points	Criteria for ap
		•	
		•	
		•	
		•	
		•	
		•	
		•	

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

Name	Components	Supplier / Cat. #	Room (hallway storage)	Safety co
Purple pipette tips		Contact lab-manager	Micro storage	

Green pipette tips	Contact lab-manager	Micro storage	
Eppendorf tubes	Contact lab-manager	Micro storage	
Distilled water	Contact lab-manager	Micro storage	
Fast digest green buffer	Agilent Technologies	Freezer at 1. Floor	
Enzyme 1	Agilent Technologies	Freezer at 1. Floor	
Enzyme 2			
PCR products			

5. QC – Quality Control

6. List of other SOPs relevant to this SOP

7. Environmental conditions required

8. Procedure

- 1. Mix the reagents from the table below
- 2. Mix the restriction mixture in an eppendorf tube by pipetting up and down
- 3. Leave for 30 min. at 37°C (no shaking!)
- 4. Immidiately load the restriction mixture in the gel
- 5. Run the gel and cut out and purify correct sized bands according to SOP0014.

Reagents	μL
Gel cut-out	XμL
Enzyme 1	1 μL
ex. ecoR1	
Enzyme 2	1 μL
ex. pst1	
Buffer	3 μL
Destilled	To total volume
Water	of 30 μL

9. Waste handling

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

10. Time consumption

11. Scheme of development

Date / Initials	Version No.	Description of changes
13.06.19 / PRA	01	The SOP has been written
13.06.26 / PRA	01	The SOP has been approved

12. Appendixes

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