Digestion

Name: Date:

Continue from Date
Name
Experiment
Project Name:

Procedure

1. add H_2O (38µl-DNA)
2. 5 µl NEB4 buffer (stored at iGEM's, -20°C)
3. 5 µl 10x BSA (used 1:10 diluted sample stored at iGEM's, -20°C)
4. DNA (500 ng) (Vector:Insert ratio 1:3 in following Ligation)
5. 1 µl restriction enzymes (stored at iGEM's, -20°C)
6. heat for 1-2 hours 37°C (6 hours if time)
7. heat for 20 minutes 80°C (inactivation of enzymes)
8. keep at 4°C if you cannot continue

Measured DNA-concentration with Nanodrop to calculate the volume of DNA to do the digestion:

Sample Name	DNA concentration (µg/µl)

Restriction enzymes you need to cut the vector, insert1 and insert 2:

Components	**Vector (µl)**	**Insert1 and 2 (µl)**
DNA (500ng)		
BSA (10x) (5µl)		
NEB4 Buffer (5µl)		
Enzyme 1 (1µl)		
Enzyme 2 (1µl)		
H_2O (38 µl- DNA)		
In total 50 µl		

Documentation:

Why are you doing this experiment? Where are the samples stored? Antibiotica resistance, vector used etc.

Run a gel to verify if the part is cut out.