

Notebook

Date: 25/8/2021

Experiment: Digestion of the gene pNRD and the plasmid pBR-A.

Person in charge: Isis Lanzoni Vargas

Description: The digestion of the gene pNRD was made using the restriction enzymes EcoRI and PstI using the **DNA Digestion** protocol. Due to the small amount of gene the reaction was made in the same tube in which the gene was contained.

Results: Results are expected in further steps.

Date: 26/8/21

Experiment: Digestion of the genes pBAD, REC1, REC2, OC1 and OC2 and ligation of the gene pNRD into the plasmid pBR-A.

Person in charge: Jose Lizano Bolaños

Description: The digestion of the genes pBAD, REC1, REC2, OC1 and OC2 were made using the restriction enzymes EcoRI and PstI using the **DNA Digestion** protocol. The reactions were made into PCR tubes.

The ligation of the gene pNRD into the plasmid pBR-A was made following the steps of the protocol **DNA Ligation**.

Results: Results are expected in further steps.

Date: 31/9/21

Experiment: Transformation of the gene pNRD into *E. coli* DH5a and ligation of the genes pBAD, REC1, REC2, OC1 and OC2 into the plasmid pBR-A.

Person in charge: Jose Lizano Bolaños

Description: Transformation of the plasmid pBR-A with the gene pNRD into *E. coli* DH5a using heat shock following the protocol **E. coli Transformation**. The antibiotic used in this procedure was ampicillin.

The ligation of the genes pBAD, REC1, REC2, OC1 and OC2 into the plasmid pBR-A were made following the steps of the protocol **DNA Ligation**.

Results: Results of the transformation were checked on 1/9/21, there were no colonies on the plate.

Date: 2/9/21

Experiment: Transformation of the genes pBAD, REC1, REC2, OC1 and OC2 into *E. coli* DH5a.

Person in charge: Jose Lizano Bolaños

Description: Transformation of the plasmid pBR-A with the genes pBAD, REC1, REC2, OC1 and OC2 into *E. coli* DH5a using heat shock following the protocol **E. coli Transformation**. The antibiotic used in this procedure was ampicillin.

Results: Results of the transformation were checked on 2/9/21, there were no colonies on the plate.

Date: 14/9/21

Experiment: Digestion of the genes pBAD, REC1, REC2, OC1 and OC2.

Person in charge: Jose Lizano Bolaños

Description: The digestion of the genes pBAD, REC1, REC2, OC1 and OC2 were made using the restriction enzymes EcoRI and PstI using the **DNA Digestion** protocol. The reactions were made into PCR tubes.

Results: Results are expected in further steps.

Date: 15/9/21

Experiment: Ligation of the genes pBAD, REC1, REC2, OC1 and OC2 into the plasmid pBR-A.

Person in charge: Isis Lanzoni Vargas

Description: The ligation of the genes pBAD, REC1, REC2, OC1 and OC2 into the plasmid pBR-A were made following the steps of the protocol **DNA Ligation**.

Results: Results are expected in further steps.

Date: 17/9/21

Experiment: Transformation of the genes pBAD, REC1, REC2, OC1 and OC2 into *E. coli* TOP10

Person in charge: Jose Lizano Bolaños

Description: Transformation of the plasmid pBR-A with the genes pBAD, REC1, REC2, OC1 and OC2 into *E. coli* TOP10 using heat shock following the protocol ***E. coli* Transformation**. The antibiotic used in this procedure was ampicillin.

Results: The results of the plates were checked on the date 18/9/21. There were colonies on the plates of the genes pBAD, OC1 and OC2.

Date: 21/9/21

Experiment: Colony PCR of the colonies from the transformations of the genes pBAD, OC1 and OC2.

Person in charge: Jose Lizano Bolaños

Description: A colony PCR was performed following the protocol **Colony PCR**, using the polymerase RedTaq and the primers V2F and VR.

Results: The results were checked on the date 23/9/21 performing a DNA agarose electrophoresis described on the protocol **DNA Digestion** in the section of "Confirmation". There was no amplification of the expected genes.

Date: From 10/10/21 to 18/10/21

Experiment: *In silico* characterization of the proteins from parts BBa_K3917001 (Counter) and BBa_K3917003 (Overcount system)

Person in charge: Jose Lizano Bolaños

Description: For each protein from the systems BBa_K3917001 (Counter) and BBa_K3917003 (Overcount system) except for the fluorescent proteins some *in silico* analysis were done. The prediction of the protein structure was performed with Swiss-Model tool. The prediction of the protein motifs was performed with My Hits Motif Scan Results.

Results: The results of the predictions can be consulted in **Results**