Biobrick Safety Sheet

Risk level: 1

Plasmid: pSB3C5

Chassis: Escherichia coli (BW25113 strain

 $\Delta EnvZ$)

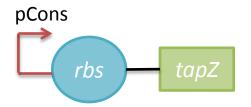


Diagram of the construction

BioBrick code: none for

the moment

Construction method

- Technic: overlapping PCR



Pcons (<u>J23119</u>)

Origin and initial function:

Parts J23119 is a constitutive promoter part isolated from a small combinatorial library.

E.coli: are bacteria commonly used in laboratories. Some strains are dangerous but most of them are Harmless.

Purposes in the system:

Expressing the tapz gene.

Size:

14 bp



Standard Elowitz RBS (Bba B0034)

Origin and initial function:

This rbs is hosted in E. Coli. It does not code for a protein and does not increase the risk level.

Purposes in the system:

It allows the transcription of tapz gene.

Size:

47 bp

Coding sequence rsmA

Alternate name: carbon storage regulator

Origin and initial function:

TapZ is a chimeric receptor, constituted by the fusion of the periplasmic part of Tap and the cytoplasmic part of EnvZ. This two receptor shares a similar structure. Tap is a low-abundance receptor involved in chemotaxis and detecting small molecules (di- and tripeptides). EnvZ activates the transcription of two genes, ompC and ompF, in response to osmotic pressure through a kinase activity and the phosphorylation of the transcription factor ompR.

<u>Purposes in the system:</u>

Detecting di-and tri-peptide in the medium and activated the transcription of gene under dependency of the pompC promoter.

Size:

1554 bp

Feedback

Theoritical interactions:

- For the moment we do not know what would happen if the microorganism were scattered outside of the laboratory. Therefore the question to answer is: in which environment can this microorganism live?

The environment in which it has been used and the consequences:

| Environment | Consequences |
|--|--|
| This biobrick is only used in a biology laboratory of level 1 for the moment | We do not know if there is any consequences. Theoretically there would be no dangerous effect. |

Safety issues:

- For the moment we do not know what would happen if the microorganism were scattered outside of the laboratory. Therefore the question to answer is: in which environment can this microorganism live?

Tests to do in order to answer safety issues:

- test organism's survival in sewers.
- check organism's presence in the researchers' bodies. What are the consequences?

Limitation:

- Because no tests have been done in a different environment than a biology laboratory of level 1, the use of those microorganisms should be forbidden in other environnements until a study proves that the risk is low enough.
- when using this microorganism good laboratory practice must be followed

characterization:

put here the information about the functioning of the BioBrick and experimental results.

- Manson MD, Blank V, Brade G, Higgins CF. Peptide chemotaxis in E. coli involves the Tap signal transducer and the dipeptide permease. Nature. 1986 May 15-21;321(6067):253-6.
- Cai SJ, Inouye M. EnvZ-OmpR interaction and osmoregulation in Escherichia coli. J Biol Chem. 2002 Jul 5;277(27):24155-61.
- Weerasuriya S, Schneider BM, Manson. MD. Chimeric Chemoreceptors in Escherichia coli: Signaling Properties of Tar-Tap and Tap-Tar Hybrids. J Bacteriol. 1998 Feb;180(4):914-20.
- Baumgartner JW, Kim C, Brissette RE, Inouye M, Park C, Hazelbauer GL. Transmembrane signalling by a hybrid protein: communication from the domain of chemoreceptor Trg that recognizes sugar-binding proteins to the kinase/phosphatase domain of osmosensor EnvZ. J Bacteriol. 1994 Feb;176(4):1157-63.

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