

How to complete a BioBrick Safety Sheet

1 Put the name of the biobrick used in the registry

2 Put a diagram of the biobrick construction using symbols at the end of this manual

3 Give plasmids on which the BioBrick has been added. Several plasmids are possible

4 Give chassis used. Several chassis are possible

5 Make a brief description of how the construction was made and focus on the biological parts used and created.

6 For each part of the biobrick give the name, the origin, its usage and its size. As far as its origin is concerned, if it is a part composed of different other parts, explain the origin of each part. It is also important to explain their natural function. If there is any BioBrick already registered set up a link with the BioBrick web page.

Biobrick Safety Sheet

Biobrick name : none for the moment

Plasmid : pSB1A3

Chassis: *Escherichia coli* (BW25113 strain)

Diagram of the construction

pLAC 0-1

rsmY

Construction method

- Technic: Gibson Assembly
- Biobricks:
 - pLAC comes from [BBa_113601](#)
 - rsmY comes from iGEM Grenoble 2011 team work

Promoter pLAC 0-1 ([BBa_R0011](#))

Origin and initial function :

This promoter is a hybrid one made up of two natural promoters. It consists of the phage lambda promoter P(L) which activates the pathogenicity by increasing the transcription. The phage lambda destroys *E. coli* using a toxin which destroys the membrane. In this regulatory region, instead of the *ci* binding site, there is *lacO1* (from *E. coli* LacI operon). *LacO1* is an operator from lactose operon, it binds LacI (the lac repressor) which is released upon complexation with IPTG. The inducer.

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

Phage lambda: is an *E. coli* virus without any pathogenicity towards humans.

Purposes in systems :

It allows a rsmY strong promotion that can still be induced by IPTG and repressed by LacI.

Size :

55 bp

Coding sequence rsmY

rsmY (name in the registry)

Origin and initial function :

rsmY is an iRNA from *Pseudomonas aeruginosa*.

Pseudomonas aeruginosa: it is a level 2 pathogenic bacterium that leads to nosocomial infection. It is ubiquitous.

Purposes in systems:

It is as a part of a riboswitch system : RsmA-rsmY. The RsmA protein inhibits the translation which is recovered thanks to the rsmY iRNA, when it binds to the RsmA protein. It is used to show that the sensor has detected something.

Size :

170bp

Example of explanation

Another part deals with the feedback

Feedback

Theoretical interactions :

We do not notice any theoretical potential effects for this BioBrick.

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This part deals with potential effects that could occur and that are underline by the theory. Therefore here you should put effects that could occur theoretically.

environment in which it has been used and consequences :

Environment	consequences
This biobrick is only used in a biology laboratory of level 1 for the moment	The construction has not been built yet. Therefore we do not know if there is any consequences. Theoretically there would be no dangerous effect.

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For this part list all environments in which the organism has been used and consequences that happened. If different kind of organism in terms of chassis used or plasmid used, do not forget to define about which construction you are talking about.

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?

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Sum up the currently state in safety and write the questions that have to be answered to improve the safety use of this BioBrick

Tests to do in order to answer safety issues :

- test organism's survival in sewers.
- check organism presence in the researchers' bodies.

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Write tests and experiences that should be done in order to answer on questions above.

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 others environments until a study proves that the risk is low enough.
- when using this microorganism good laboratory practice must be followed

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Thanks to information given previously, give in which ways this BioBrick has to be used or not.

characterization :

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

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in order to give more information on its use, put all the information about the functioning of the BioBrick and experience results

- Lederberg E. (January 1950). Lysogenicity in Escherichia coli strain K-12 , *Microbial Genetics Bulletin*, v.1, pp. 5-8
- Lederberg J. (1953). Genetic Studies of Lysogenicity in Escherichia Coli . *Genetics* 38. 51-64. [on line]. (august 2012). available on [PMID 17247421](https://pubmed.ncbi.nlm.nih.gov/17247421/)
- St-Pierre F, Endy D (2008). Determination of cell fate selection during phage lambda infection. *Proc. Natl. Acad. Sci : U.S.A.* 105. [on line] (August 2012). Available on <http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2605630/?tool=pmcentrez>
- (2011) Bacteria An introduction to Earth's largest family, PDF generated using the open source mwlib toolkit. [on line] (August 2012). Available on <http://www.scribd.com/doc/74972710/Bacteria>
- Pseudomonas Genome Database . [on line] (August 2012). Available on <http://v2.pseudomonas.com/>
- Brenni A, Lory S. (May 2009). Determination of the regulon and identification of novel mRNA targets of Pseudomonas aeruginosa RsmA. [on line] (August 2012). Absattract available on <http://www.ncbi.nlm.nih.gov/pubmed/19426209>

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At the end do not forget to write references used and also the name of the team who puts the risk level on the BioBrick Safety Sheet, together with the date of the last update.

Author: (iGEM Grenoble 2012)

Symbols to use



RBSs



promoter



Coding sequence

Biobrick Safety Sheet

BioBrick name : put the name of the biobrick in the registry

Adding on a plasmid : name of them

Chassis used : Name of theme

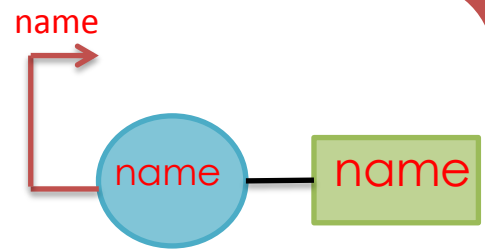


Diagram of the construction

Construction method

Put here the way the construction is built and give the different biological parts used during the construction

Promoter



:

name(name registry with the link to the web page)

Origin and initial function :

Give the origin of the promoter. More precisely give the origin of the promoter in terms of organisms and what is the goal in the nature.

Purposes in the system :

Give its usage in the system

Size :

complete

RBS

name

:

Name and link to the part registry

Origin and initial function :

complete

Purposes in systems :

complete

size:

complete

Coding sequence

Name and part registry link

Origin and initial function :

complete

Purposes in the system :

complete

Size :

complete

Feedback

Theoretical interactions :

Thanks to the theoretical knowledge that you have, give the potential bad effects that would be present depending the way the BioBrick is used.

environment in which it has been used and consequences :

Environment	consequences
complete	Complete (add lines if necessary)

Safety issues:

- complete

Tests to do in order to answer safety issues :

- complete

Limitation :

-complete

characterization :

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

- Put references

Author : put the name of the team that complete this sheet

Update: put the current date