Pojma ‚biološki del‘ in ‚biokocka‘

Biological part, Biobrick

Standardiziran biološki del je funkcionalna enota DNA (nukleotidno zaporedje), ki zapisuje za neko biološko funkcijo.

Osnovni deli imajo samo eno funkcijo in jih ni mogoče razdeliti na manjše enote. Osnovne dele lahko sestavljamo in tako dobimo sestavljene dele.

‚Naprava‘ je poseben tip sestavljenega dela, ki v celici opravlja neko nalogo.

‚Biokocka‘ = standardni biološki del (Tom Knight, 2003)

SB temelji na hierarhičnem sestavljanju osnovnih delov.

Primeri biokock

Osnovni del – npr. lambda promotor 

Sestavljeni del – npr. RBS, zapis za RFP, terminator 

Naprava je sestavljeni del, ki v celici opravlja neko nalogo – npr. regulirana sinteza RFP pod kontrolo cI

Sistem je sestavljen iz več naprav in predstavlja ustrezno povezane genetske elemente, ki zagotavljajo tak celični fenotip, kakršnega smo si zamislili v načrtu. Primeri sistemov so (a) ustrezno prilagojene metabolične poti za sintezo neke nove snovi, ali (b) oscilatorji.

Fundacija BioBricks

„Biotehnologija v javnem interesu“

BBF so ustanovili 2006 in je ‚organizacija v javnem interesu‘. Cilj organizacije je, da se biološko inženirstvo izvaja na odprt in etičen način in v dobrobit vseh ljudi in planeta v celoti. Ustanovitelji BBF so bili znanstveniki in inženirji, ki so spoznali, da ima sintezna biologija potencial, da močno zaznamuje usodo ljudi in planeta, in ki so želeli, da to novo področje služi javnemu interesu.

Vizija BBF je, da bi prišlo do sodelovanja med znanstveniki in inženirji, da bi za delo uporabljali prosto dostopne standardizirane biološke dele, ki so varni, etični, cenovno učinkoviti in javno dostopni, s tem pa bi ustvarjali rešitve za probleme, s katerimi se srečuje človeštvo.

SB naj bi delovala za dobro v svetu in naj bi postala del arhitekture, medicine, bioremediacije, kmetijstva in drugih področij.

Ključni za uspeh SB v prihodnosti so mladi znanstveniki.

Biološko varnost (biosecurity & biosafety), bioetiko, skrb za zdravo okolje in trajnostno naravnanost je treba združiti z raziskovalnim delom in uporabnimi tehnologijami.

Člani BBF so inženirji, znanstveniki, odvetniki, izumitelji, učitelji, študentje, politiki in običajni državljani, ki si prizadevajo za uresničitev ciljev fundacije.

Registry of Standard Biological Parts

The Registry's Repository

The Registry's Repository contains DNA samples for thousands of parts, submitted by iGEM teams and labs. Last year, iGEM teams sent in samples for over 1500 parts.

Be sure to add your parts and send samples to the Registry so that they can be made available to the community!

add your part

send your sample

Registry News

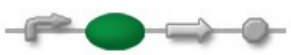
- Registry Release
- Registry 6.0
- Report Bugs
- Request Features
- News Archive
- Feature Box Archive

Other

- Registry API
- Safety
- Videos

Featured on the Registry

Ribosome Binding Sites

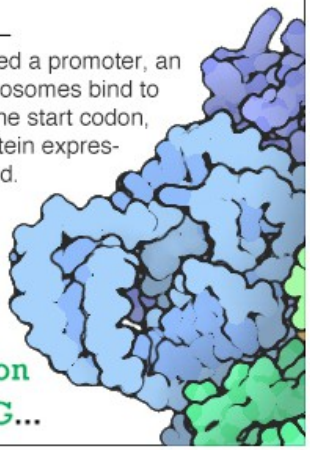


To make a protein in a cell you need a promoter, an RBS, a CDS and a terminator. Ribosomes bind to the RBS and begin translation at the start codon, which is ATG on the registry. Your choice of RBS will affect protein expression levels. RBSs are small so can be synthesized or assembled. We have have about 150 RBS parts on the Registry. The most used is BBa_E0034. There are many RBS collections on the Registry:

Anderson Collection | By expression level | *E. coli* | Eukaryotic

Promoter Ribosome binding site Start Codon

...TCTAGAGAAAGANNNGANNNACTAGATG...



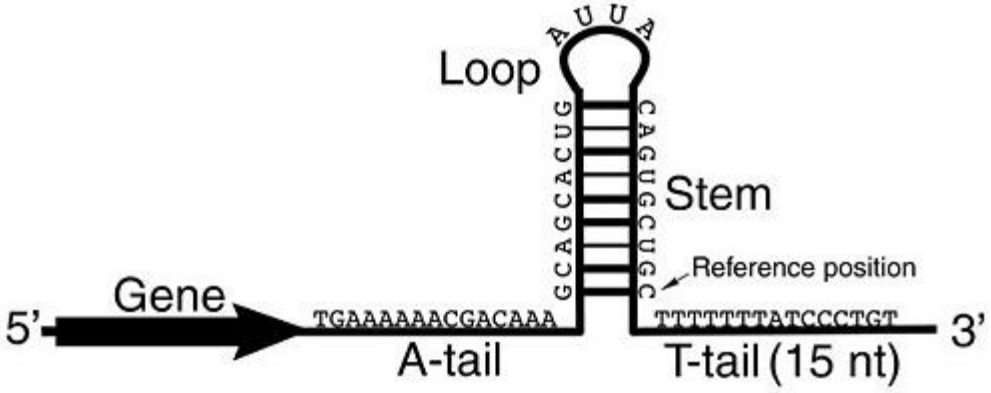
The iGEM Registry is a growing collection of genetic parts that can be mixed and matched to build synthetic biology devices and systems.

As part of the synthetic biology community's efforts to make biology easier to engineer, it provides a source of genetic parts to iGEM teams and academic labs.

You can learn more about iGEM Teams and Labs at iGEM.org.

Tipi osnovnih bioloških delov:

- Promotorji
- Vezavna mesta za ribosom
- Kodirajoča zaporedja
- Transkripcijski terminatorji



http://parts.igem.org/Catalog

Registry of Standard Biological Parts

Catalog

- Browse parts by type • devices by type
- Browse parts and devices by function • by chassis • by standard • or by contributor
- Browse chassis
- Browse user-supplied catalog pages - these pages have not undergone curation by the Registry but have been made by the Registry user community. Please feel free to add new catalog pages to this section.

Browse parts by type

Catalog	List	
		**Promoters (?)**: A promoter is a DNA sequence that tends to recruit transcriptional machinery and lead to transcription of the downstream DNA sequence.
		**Ribosome Binding Site/about (?)**: A ribosome binding site (RBS) is an RNA sequence found in mRNA to which ribosomes can bind and initiate translation.
		**Protein domains (?)**: Protein domains are portions of proteins cloned in frame with other proteins domains to make up a protein coding sequence. Some protein domains might change the protein's location, alter its degradation rate, target the protein for cleavage, or enable it to be readily purified.
		**Protein coding sequences (?)**: Protein coding sequences encode the amino acid sequence of a particular protein. Note that some protein coding sequences only encode a protein domain or half a protein. Others encode a full-length protein from start codon to stop codon. Coding sequences for gene expression reporters such as LacZ and GFP are also included here.
		**Translational units (?)**: Translational units are composed of a ribosome binding site and a protein coding sequence. They begin at the site of translational initiation, the RBS, and end at the site of translational termination, the stop codon.
		**Terminators (?)**: A terminator is an RNA sequence that usually occurs at the end of a gene or operon mRNA and causes transcription to stop.
		**DNA (?)**: DNA parts provide functionality to the DNA itself. DNA parts include cloning sites, scars, primer binding sites, spacers, recombination sites, conjugative tranfer elements, transposons, origami, and aptamers.
		**Plasmid backbones (?)**: A plasmid is a circular, double-stranded DNA molecules typically containing a few thousand base pairs that replicate within the cell independently of the chromosomal DNA. A plasmid backbone is defined as the plasmid sequence beginning with the BioBrick suffix, including the replication origin and antibiotic resistance marker, and ending with the BioBrick prefix.
		**Plasmids (?)**: A plasmid is a circular, double-stranded DNA molecules typically containing a few thousand base pairs that replicate within the cell independently of the chromosomal DNA. If you're looking for a plasmid or vector to propagate or assemble plasmid backbones, please see the set of plasmid backbones. There are a few parts in the Registry that are only available as circular plasmids, not as parts in a plasmid backbone, you can find them here. Note that these plasmids largely do not conform to the BioBrick standard.
		**Primers (?)**: A primer is a short single-stranded DNA sequences used as a starting point for PCR amplification or sequencing. Although primers are not actually available via the Registry distribution, we include commonly used primer sequences here.
		**Composite parts (?)**: Composite parts are combinations of of two or more BioBrick parts.

Registry of Standard Biological Parts

Composite parts (?): Composite parts are combinations of of two or more BioBrick parts.

Browse devices by type

We're in the process of developing new support for the specification of devices in the Registry. For the time being, please see the existing device tables below.

- **Protein generators (?)**:
- **Reporters (?)**:
- **Inverters (?)**:
- **Receivers and senders (?)**:
- **Measurement devices (?)**:

Browse parts and devices by function

*This section replaces the previous **Featured parts** pages.*

Biosafety: Parts and devices improving biological containment.

Biosynthesis: Parts involved in the production or degradation of chemicals and metabolites are listed here.

Cell-cell signaling and quorum sensing: Parts involved in intercellular signaling and quorum sensing between bacteria.

Cell death: Parts involved in killing cells.

Coliroid: Parts involved in taking a bacterial photograph.

Conjugation: Parts involved in DNA conjugation between bacteria.

Motility and chemotaxis: Parts involved in motility or chemotaxis of cells.

Odor production and sensing: Parts the produce or sense odorants.

DNA recombination: Parts involved in DNA recombination.

Viral vectors: Parts involved in the production and modification of Viral vectors.

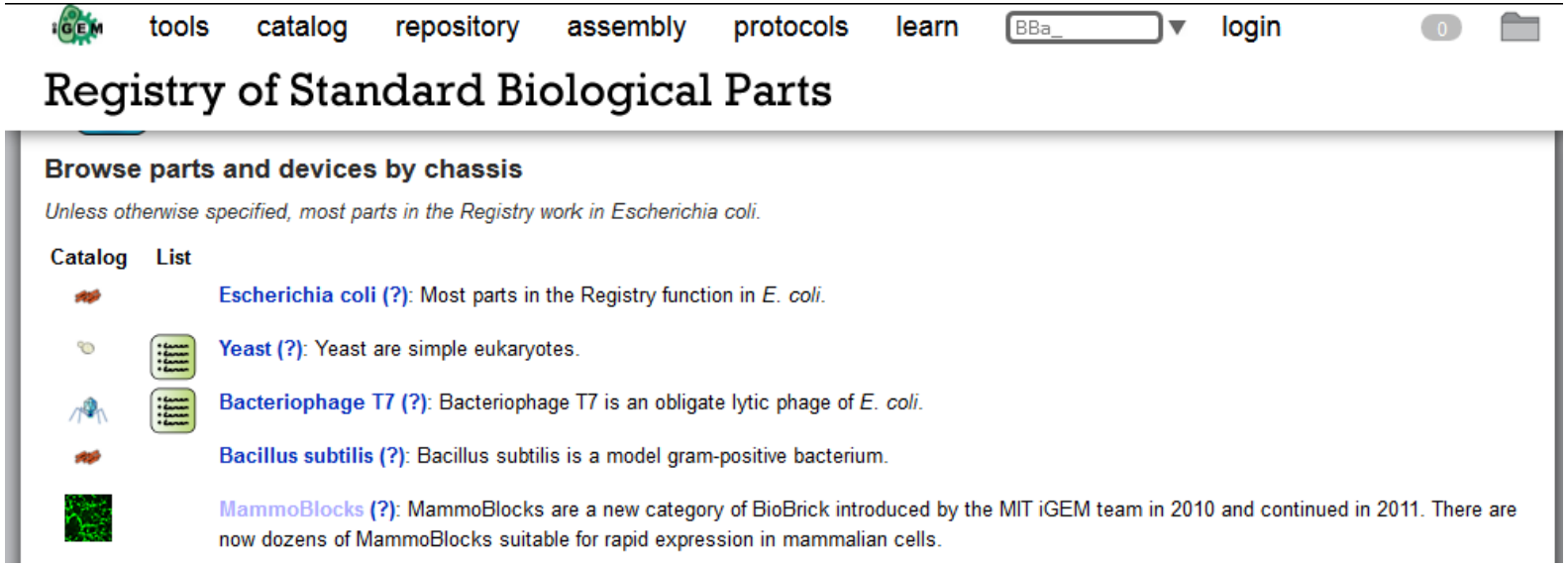
Categories

- biosafety (19)
- cds (700)
- chassis (1525)
- classic (2003)
- collections (0)
- direction (839)
- dna (133)
- function (1219)
- plasmid (244)
- plasmidbackbone (153)
- primer (42)
- promoter (667)
- proteindomain (420)
- rbs (156)
- regulation (787)
- ribosome (149)
- rnap (573)
- t3 (3)
- terminator (67)
- test (3)
- test1 (1)
- viral_vectors (115)

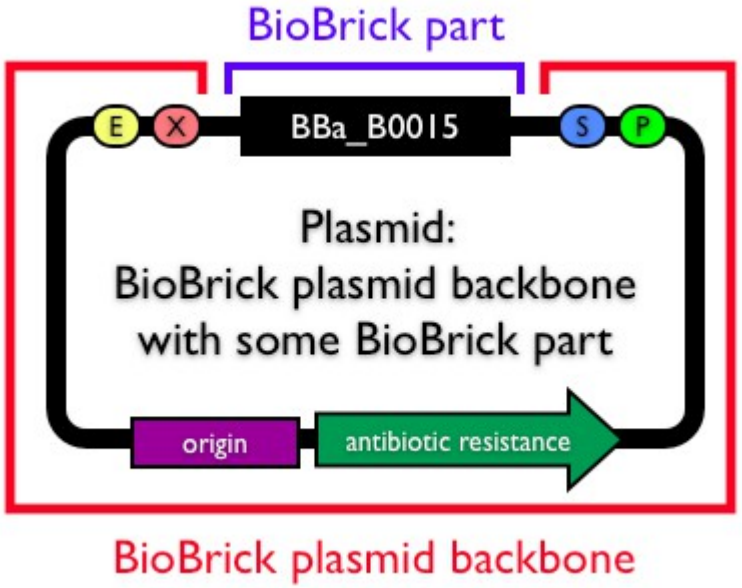
Šasija

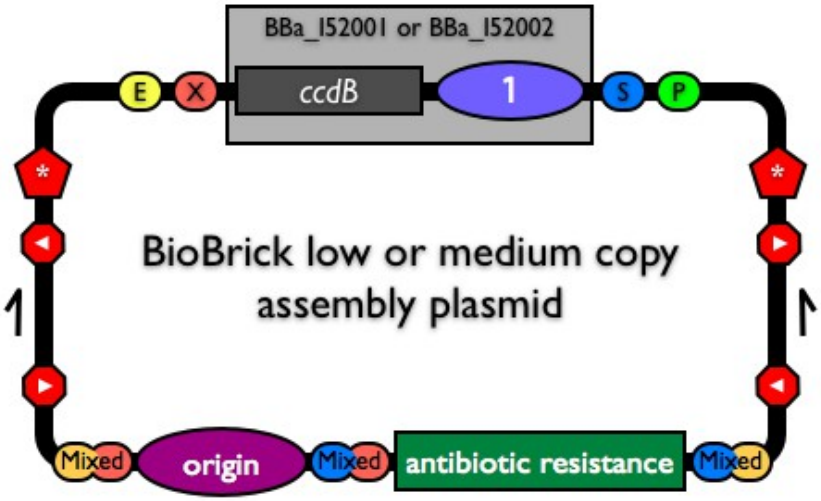
Chassis

‚Šasija' pomeni osnovo, na kateri delamo spremembe in je običajno enaka gostiteljskemu organizmu, ki ga uporabimo za izvedbo eksperimentov. Vendar pa z izrazom šasija opisujemo tudi brezcelične sisteme za npr. transkripcijo in translacijo.



Biokocke so praviloma vstavljene v plazmid (plazmidni skelet)





http://parts.igem.org/Plasmid_backbones/Assembly

Da bi lahko dele s pridom uporabili za sestavljanje različnih sistemov, morajo biti dobro raziskani in opisani.

Osnovne biološke dele povezujemo v sestavljene po točno določenih postopkih – standardih sestavljanja (*assembly standards*), ki so opisani v literaturi (ne vedno v člankih). Postopki so različni in so oštevilčeni, da je mogoče razlikovati med standardi sestavljanja, saj je od tega odvisno, kako fleksibilne so posamezne naprave in sistemi.

Assembly standard 10 (?): Assembly standard 10, or the original BioBrick assembly standard, was developed by Tom Knight in 2003. Most parts in the Registry comply with this assembly standard.

Assembly standard 23 (?): Assembly standard 23, or the Silver standard, is compatible with original BioBrick assembly standard and allows for in-frame assembly of protein domains.

Assembly standard 25 (?): Assembly standard 25, or the Freiburg standard, extends upon the original BioBrick assembly standard and allows for in-frame assembly of protein domains.

Assembly standard 21 (?): Assembly standard 21, also known as the BglBrick, BBb, or Berkeley standard, is optimized to enable in-frame assembly of protein domains.

Assembly standard 28 (?): Assembly standard 28, also known as the Lim lab standard or AarI cloning, is optimized for assembly of 3 parts into a vector simultaneously. Most parts that comply with Assembly standard 28 function in yeast.

Assembly standard 15 (?): Julie Norville has developed a new set of parts for assembly of fusion proteins.

Assembly standard 65 (?): The MIT iGEM 2010 and 2011 teams have developed a Gateway-based standard for assembly of mammalian promoters, genes, and expression vectors known as MammoBlocks.

Sestavljanje delov med seboj

Dele, pripravljene po istem standardu, lahko med seboj spajamo in ob tem ohranjamo njihov format, torej niso potrebne nobene predhodne prilagoditve posameznih delov.

Vsak del ima v vektorju svoj zgornji in spodnji rob (prefix, suffix), preko katerih poteka izrezovanje in kasneje zlepljanje.

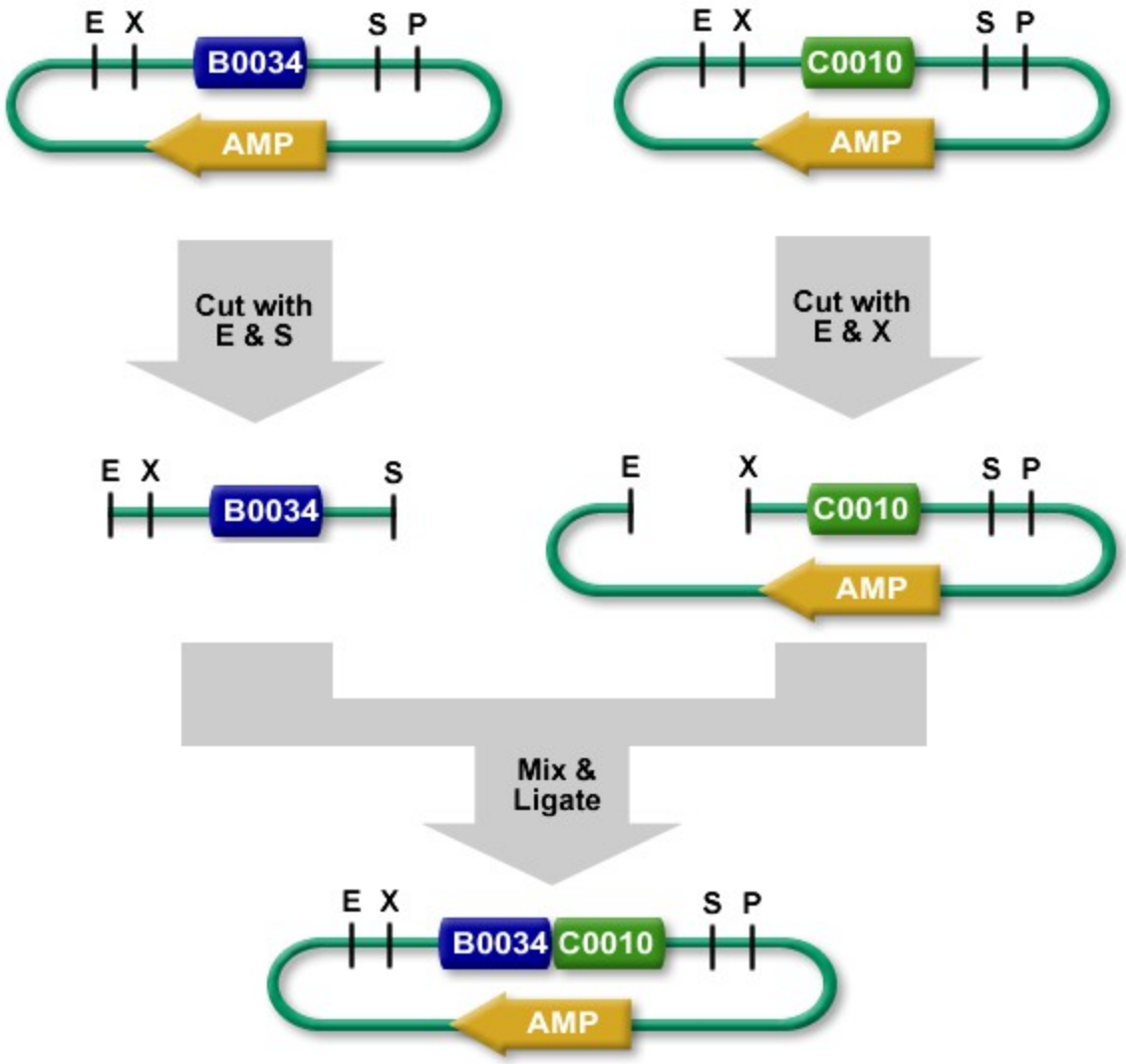
```
            Prefix                                          Suffix
5' - GAATTC GCGGCCGC T TCTAGA G ...part... T ACTAGT A GCGGCCG CTGCAG - 3'
     EcoRI    NotI      XbaI                  SpeI     NotI   PstI
```

Ko povezujemo dva dela, med njima ostane spoj (scar), kar je lahko problematično pri povezovanju dveh kodirajočih zaporedij, ki morata ostati v istem bralnem okviru. To je eden od razlogov, zakaj je bilo treba prvi standard (sestavljanje št. 10) nadgraditi.

Prvi del izrežemo z encimoma Eco/Spe, vektor z drugim delom pa režemo z Eco/Xba. Po končani ligaciji ostane med deloma spoj:

5‘- [part 1] TACTAGAG [part 2] -3'

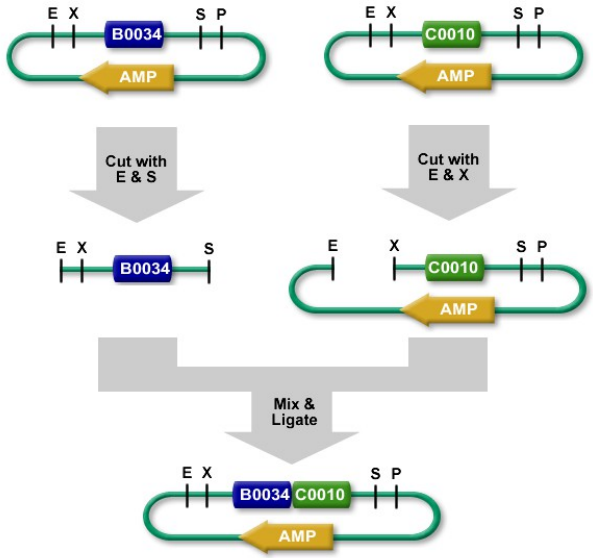
Standardni način sestavljanja (BioBrick10)



http://parts.igem.org/Help:Assembly

Restrikcijskih mest, ki jih uporabite za sestavljanje, ne smete imeti v zaporedju biokock!

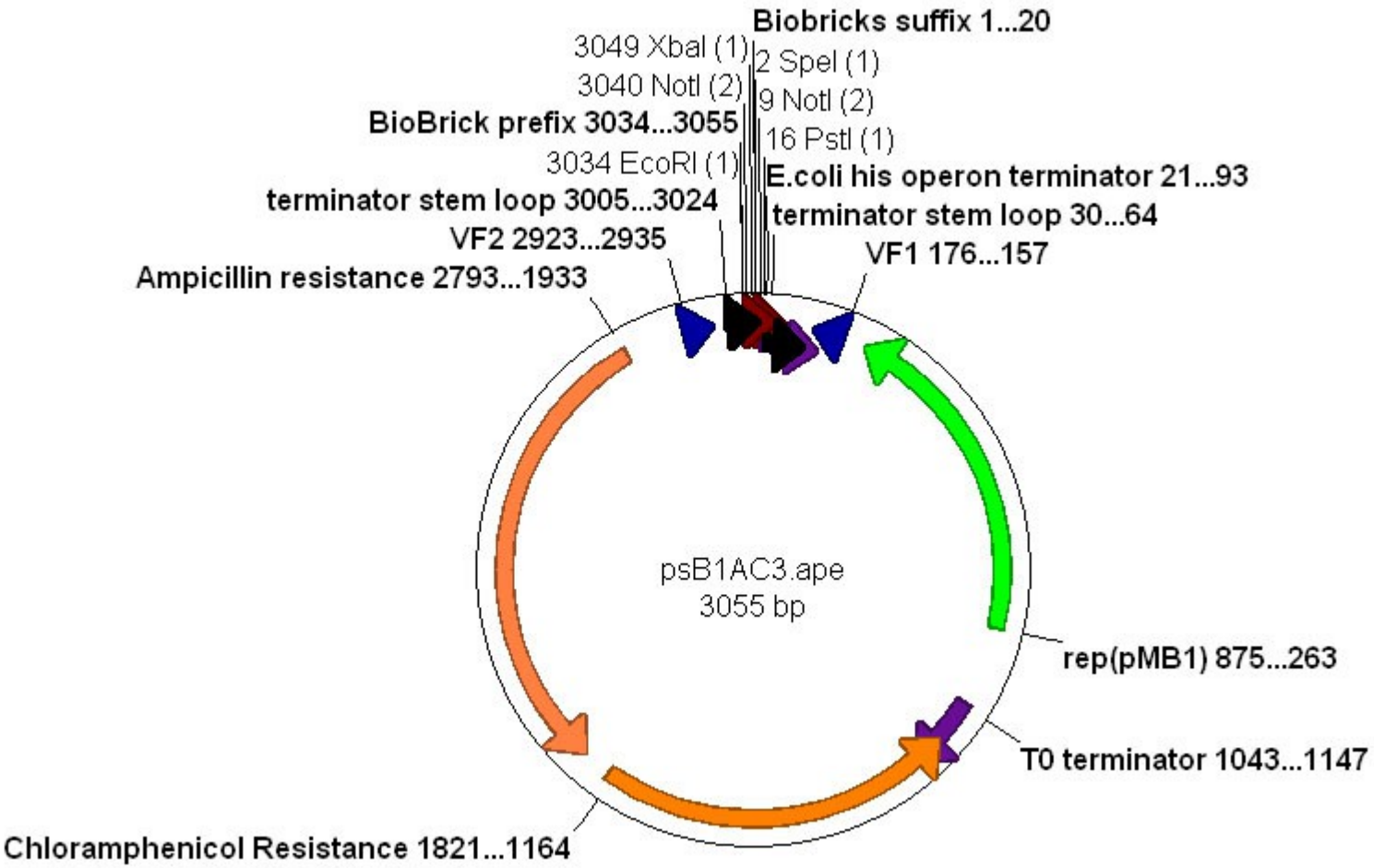
```
EcoRI: G/AATTC
XbaI:  T/CTAGA
SpeI:  A/CTAGT
PstI:  C/TGCAG
NotI: GC/GGCCGC
```



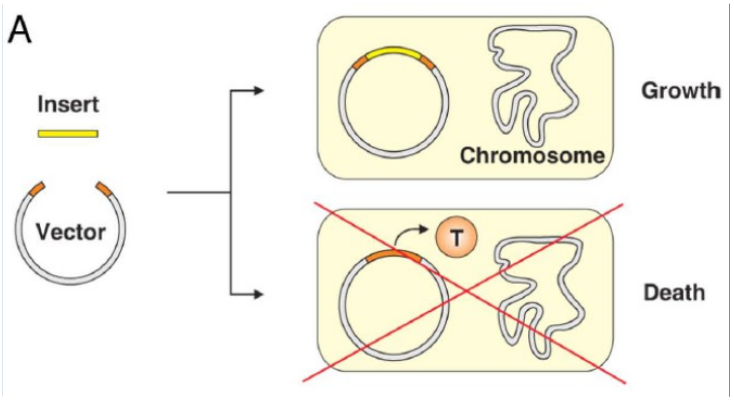

```
5’ *AATTC GCGGCCGC T TCTAGA G --Insert-- T A      3’
3’      G CGCCGGCG A ACATCT C --Insert-- A TGATC* 5’
   EcoRI   NotI       XbaI                 SpeI
```

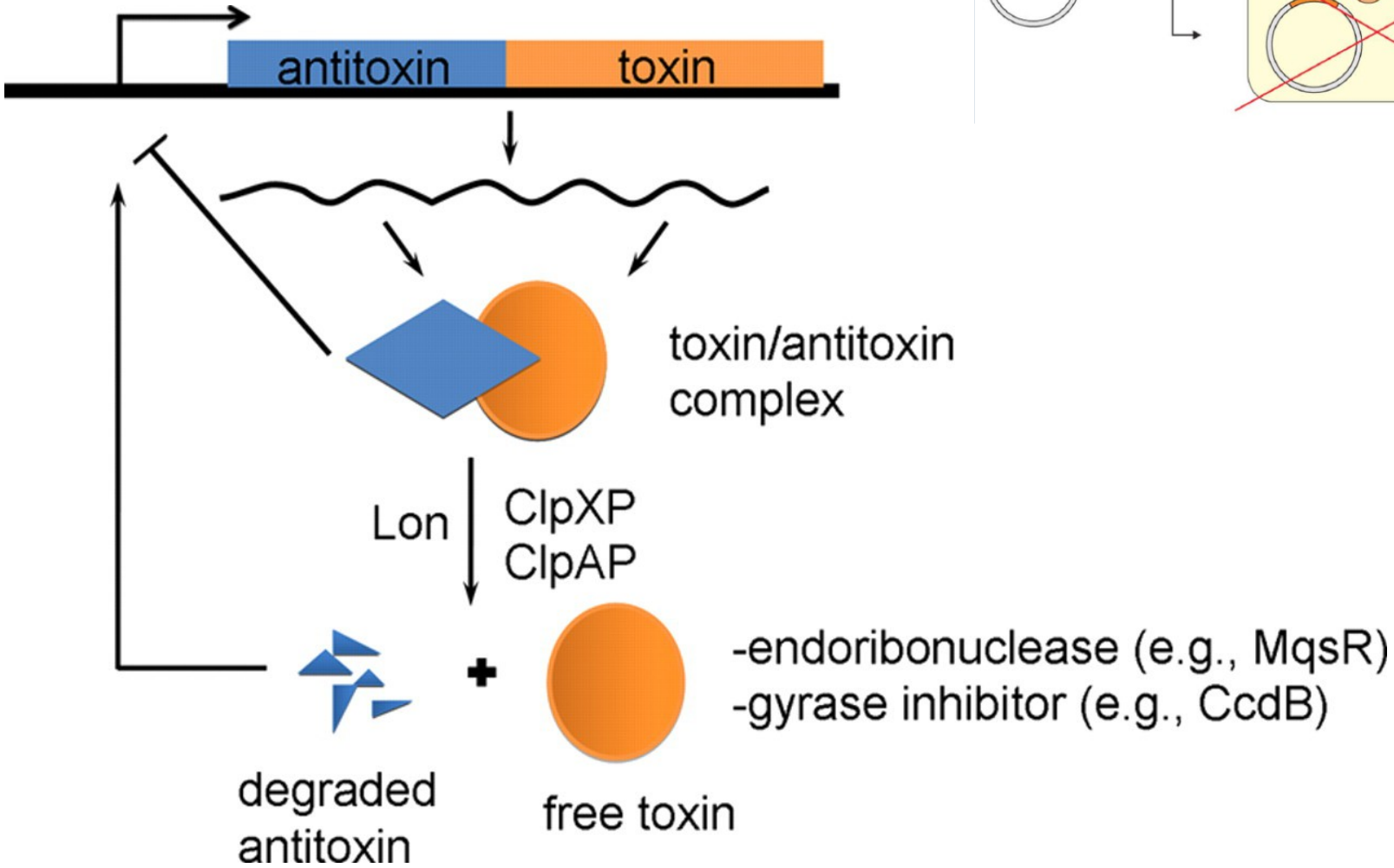
```
5’ --gca G                *CTAGA G---- 3’
3’ --cgt CTTAA*                T C---- 5’
         EcoRI              XbaI
```

```
5’ --gca G  *AATTC GCGGCCGC T TCTAGA G--insert--T A   *CTAGA G---- 3’
3’ --cgt CTTAA*  G CGCCGGCG A ACATCT C--insert--A TGATC*   T C---- 5’
         EcoRI       NotI       XbaI                 Mixed
```



pSB1AC3-P1010: v MCS je vstavljen zapis za CcdB (toksin)





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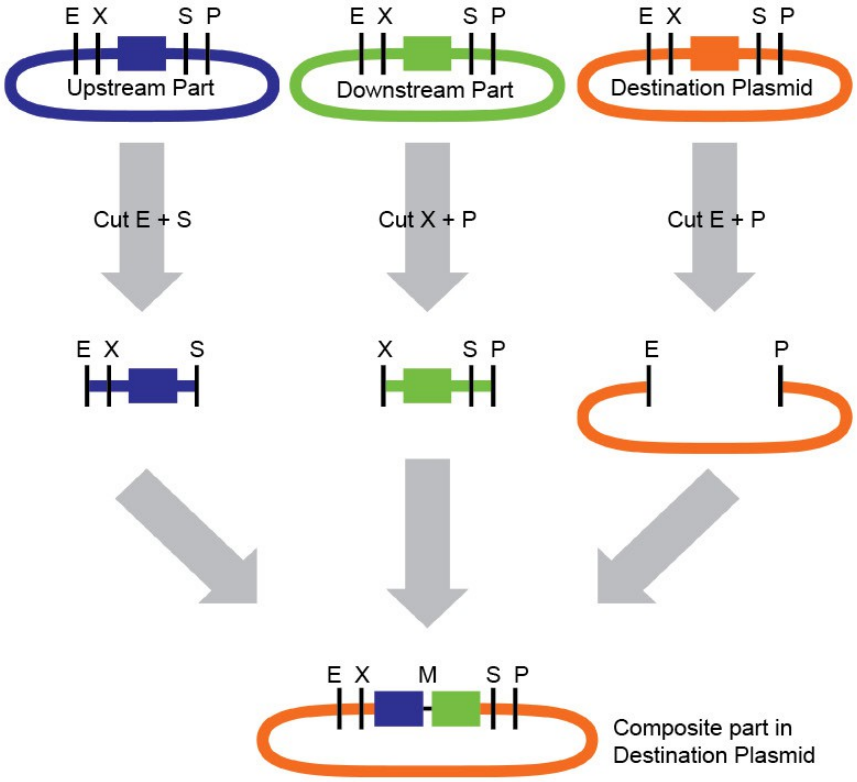
BioBrick™ Assembly Manual

ginkgobioworks™

NEW ENGLAND BioLabs Inc.

This manual describes the major steps of BioBrick assembly using BioBrick Assembly Standard 10. The input to the protocol is DNA for the two parts to be assembled and a destination plasmid. The manual includes protocols for the digestion of the three input DNA molecules and the ligation of the digested DNA to form a circularized plasmid containing the composite part. The product of the ligation reaction can be used to transform competent cells with the composite part. To read more about the BioBrick system and browse the BioBrick collection, visit the Registry of Standard Biological Parts at http://partsregistry.org.

E=EcoRI-HF™
X=XbaI
S=SpeI
P=PstI
M=Mixed site



BioBrick assembly overview

1 Start with two BioBrick parts and a BioBrick destination plasmid. The destination plasmid contains a toxic gene, *ccdB*, in the BioBrick cloning site and a different antibiotic resistance marker to the upstream and downstream parts.

2 Digest each of the parts with the appropriate restriction enzymes.

3 Mix the digests together and perform a ligation step. One of the ligation products formed will be the correctly assembled composite part in the destination plasmid. You can use the ligation mix to transform competent cells with the new composite part.

The BioBrick™ Assembly Kit from NEB and Ginkgo BioWorks has been designed for use with this manual. Download this manual from http//ginkgobioworks.com/support

Version 1.0