CHEM-E8125 – Synthetic Biology

BioBricks

Group 5:

Ville Takio

Micaela Lemström

Cecilia Hansson

Thi Lam

Gastric cancer inhibitor

Situation:

• 5ft most common cancer and 3rd most common cause of cancer death globally

Problem:

• Chronic alcohol consumption increases the risk of gastric cancer

What:

• Acetaldehyde classified as group 1 carcinogen to humans

How:

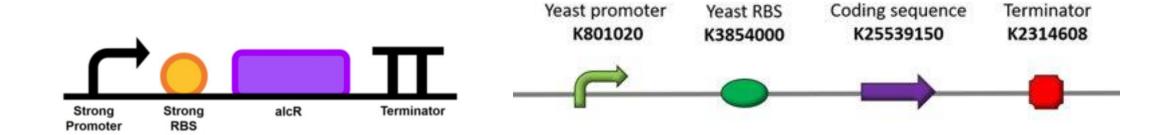
• Alcohol stimulates the uptake of carcinogens and enhances the aldehyde level

Solution:

Parts and how they work

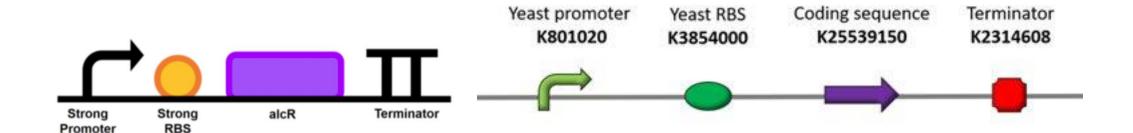
A NOT gate is a type of logic gate that produces an output that is the opposite of its input. In synthetic biology, this can be achieved using a regulatory protein that blocks the expression of a gene of interest in the absence of a specific input signal. Here's how to assemble a NOT gate with *S. cerevisiae* as the host organism:

- Clone the regulatory protein coding sequence (BBa_K2539150) downstream of the promoter (BBa_K801020) using standard molecular cloning techniques. This will create a transcriptional unit that is controlled by the promoter.
- Clone the gene of interest (BBa_K2539150) downstream of a second promoter (alcA) regulated by the regulatory protein (AlcR). This will create a second transcriptional unit that expresses the acetaldehyde dehydrogenase.
- Clone the terminator (BBa_K3854000) downstream of both transcriptional units to ensure that transcription stops at the end of the gene.
- Transform the resulting plasmid into yeast cells using standard transformation techniques.



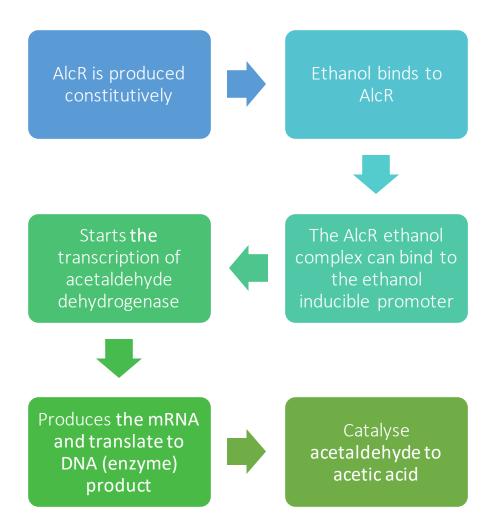
How does the construct work

- When ethanol is absent, the regulatory protein, which is constitutively expressed does not bind to the inducible promoter's operator, thus no transcription of the acetaldehyde dehydrogenase.
- When ethanol is present, it will bind to the activator protein and enable its binding to the promoter, allowing transcription
 of the acetaldehyde dehydrogenase.





How the system works



If no ethanol is present --> no production of acetaldehyde dehydrogenase

Design

AlcR Expression Construct Coding sequence Yeast promoter Terminator Yeast RBS K2539300 K801020 K25539150 K3854000 K2314608 alcR Terminator Strong Strong Promoter RBS

AlcR = regulatory protein, which senses ethanol and binds to the yeast ethanol inducible promoter

The promoter is constitutive promoter.

Ethanolinducible (CCACC) Kozak sequence

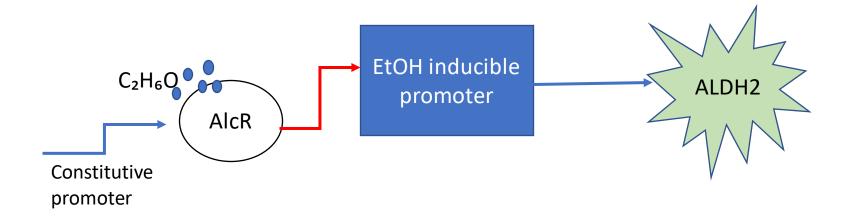
The ribosome binds here and starts transcription --> translation.

Acetaldehyde Dehydrogenase **Terminator Tmini**

Truth table

AND-gate truth table

Input (AlcR)	Input (EtOH)	Output (ALDH)
1	0	0
1	1	1
0	1	0
0	0	0



References

- https://pubmed.ncbi.nlm.nih.gov/28538665/
- https://pubmed.ncbi.nlm.nih.gov/32861308/