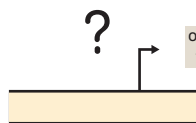


## 1. Design Reporters

unknown regulatory sites



obtain sequence  
from database

ACTGGAATT...

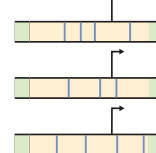


computationally design  
promoter variants

GCTGGTATT...  
ACGGGAATA...  
ACTTCAACT...

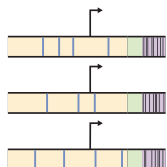
add primer pairs

order oligo pool



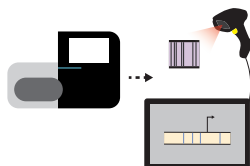
## 2. Library Cloning

add random barcode

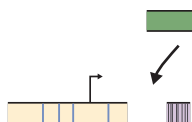


sequencing

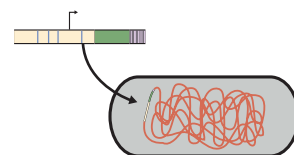
barcode mapping



add reporter gene

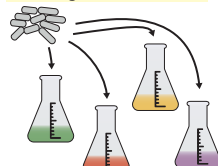


integrate reporters into genome

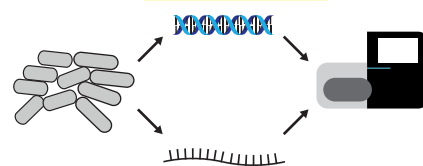


## 3. Growth and Sequencing

grow library in  
various growth conditions

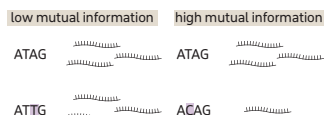


targeted sequencing on  
gDNA and RNA



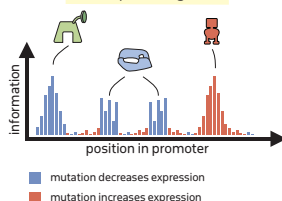
## 4. Identify Binding Sites

compute mutual information



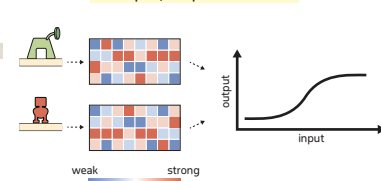
obtain information  
footprint

identify binding sites



compute energy  
matrices

fit thermodynamic models  
for input/output functions



## 5. Identify Transcription Factors

*in vitro* binding assay

knockout strains

DNA chromatography &  
mass spectrometry