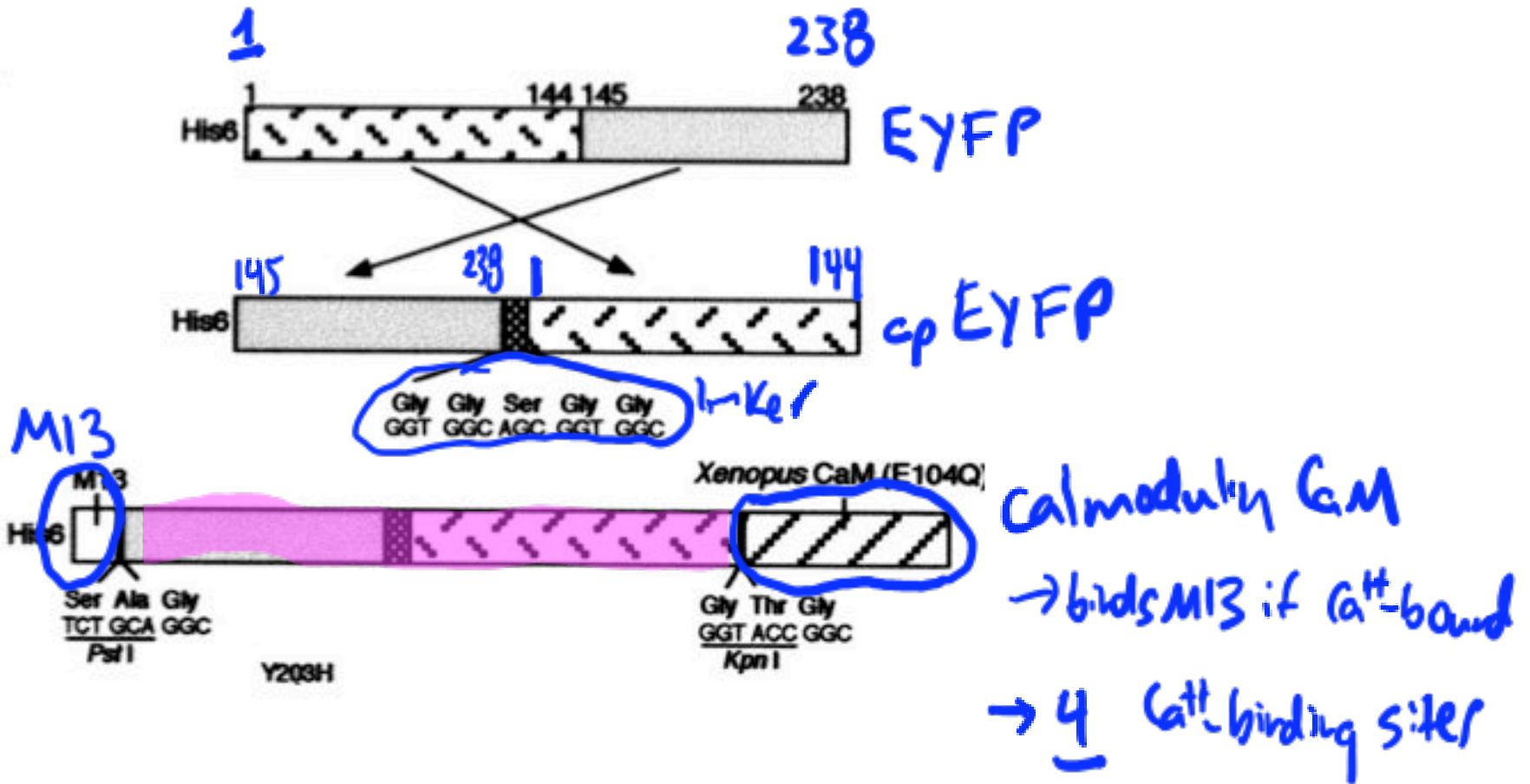


- Announcements
- Pre-lab Lecture
  - ❖ Module 2: Design Overview
  - ❖ Primer design for mutagenesis
  - ❖ Intro to Restriction Enzymes
  - ❖ Today in Lab: M2D1

# Announcements

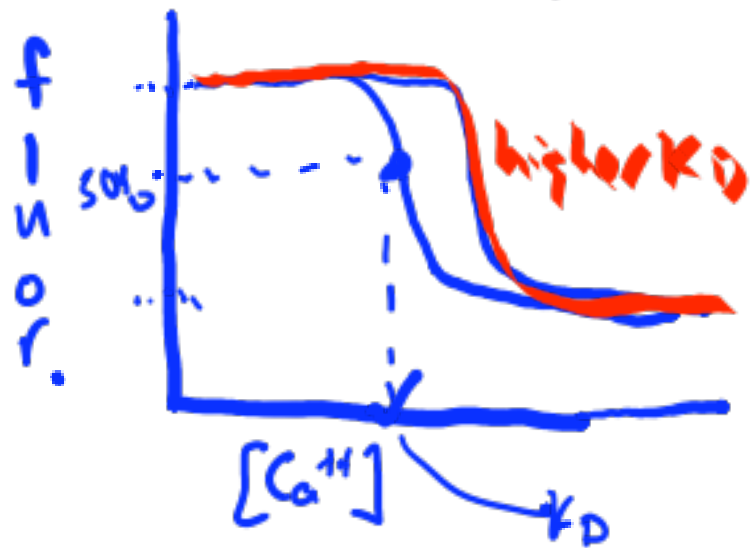
- iGEM competition  
info session Mar 11<sup>th</sup> (R) @ 5 or 6pm <igem.org>  
56-402
- Introducing... Xiaosai, your TA for Module 2

# Inverse Pericam



# Goal: Affect Binding Properties

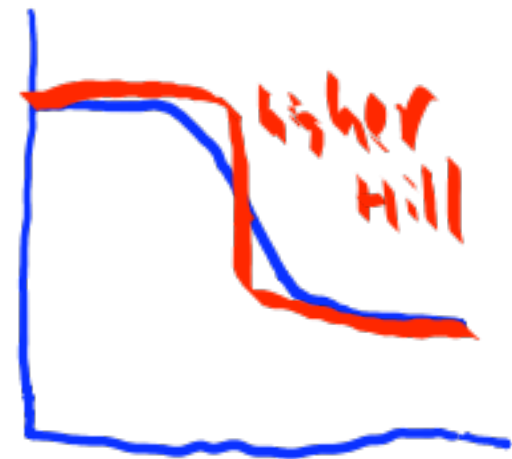
$\Delta$ affinity  $\downarrow$



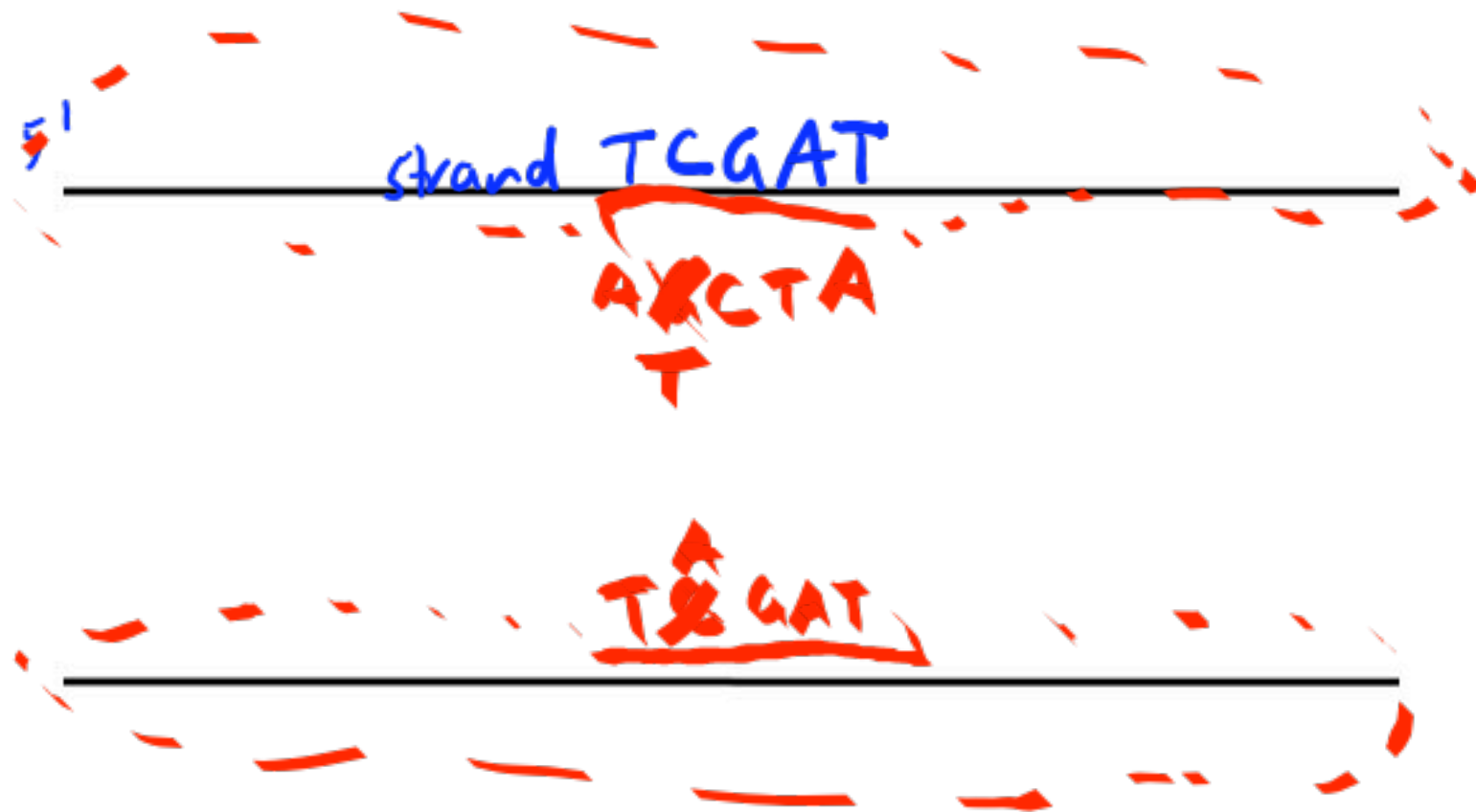
vary  $[Ca^{2+}]$ , keep  $[CaM]$  constant

fluorescence  $\uparrow$   
binding

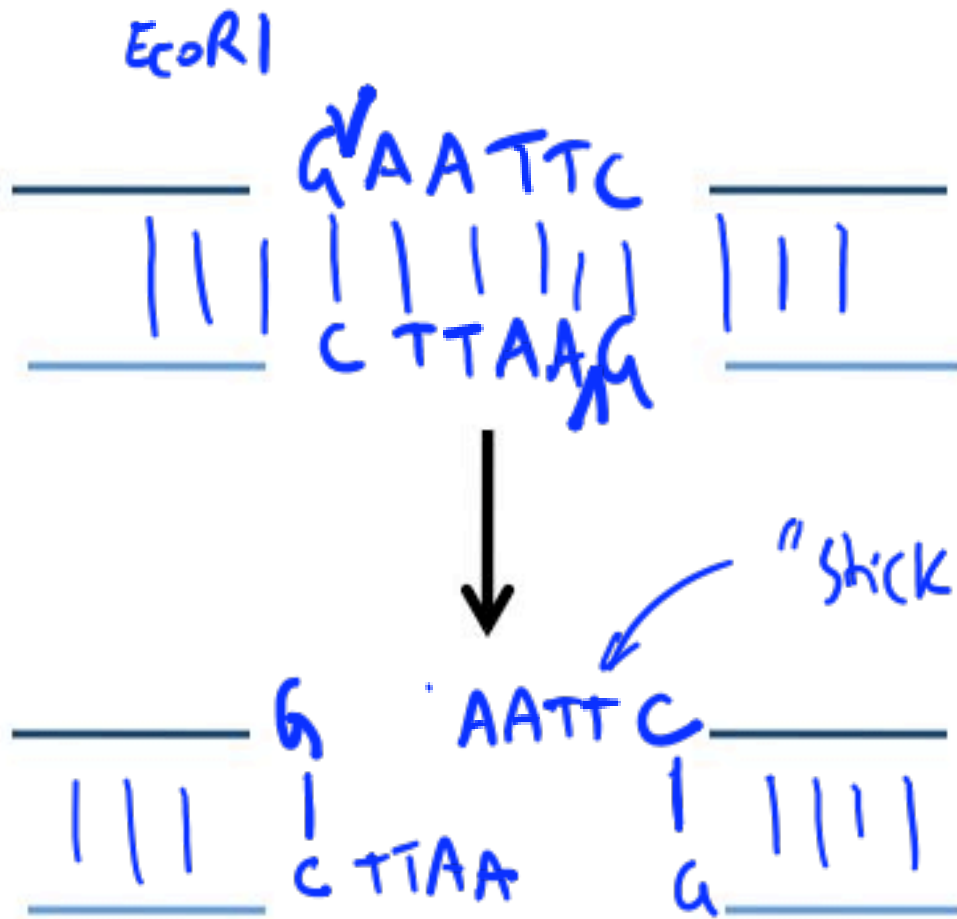
$\Delta$ cooperativity  $\uparrow$



# Designing Mutagenic Primers



# Intro to Restriction Enzymes



endonucleases

→ cuts DNA




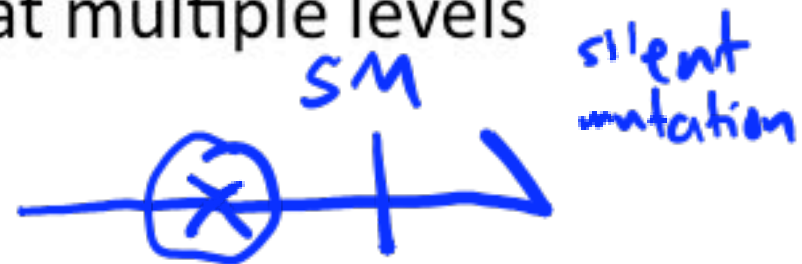
"sticky ends" can be used in ligations

palindromic DNA

also type 2  
leave blunt ends

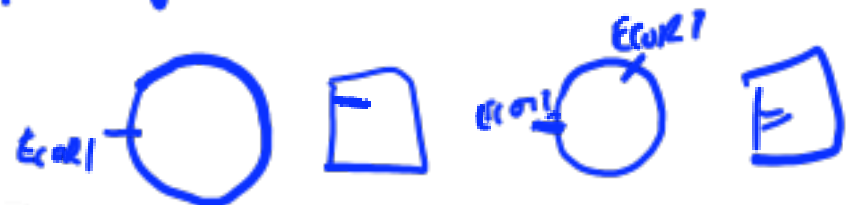
# Today in Lab

- Study inverse pericam at multiple levels
- Design primers
  - Amino acid change 



- Silent change **make new, unique restriction site**

example: S101L  
exp. → M124S



- For next time: begin reading two papers
  - Focus on Nagai; other one is for some history/context
  - Time in class on D2 to finish