

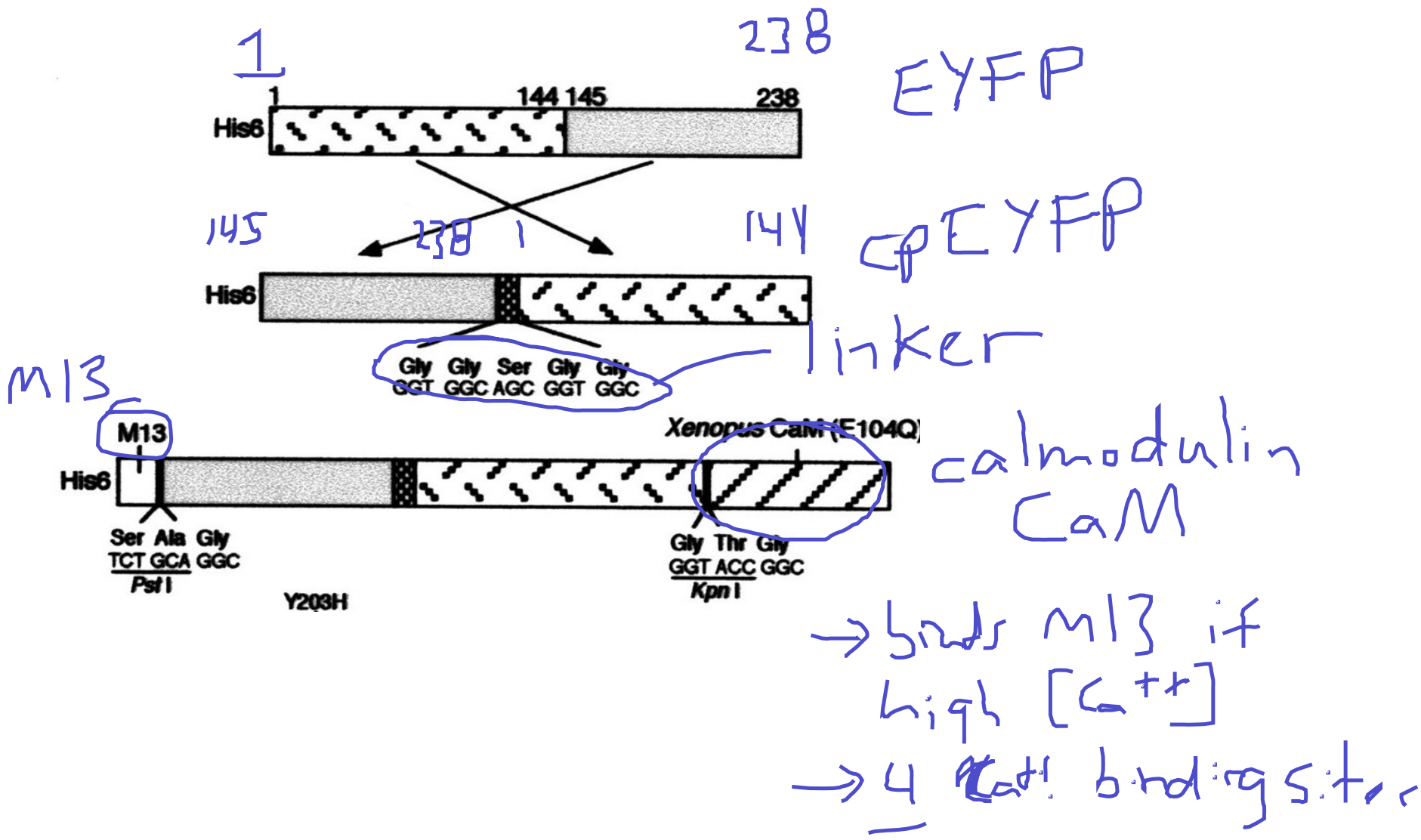
- Announcements
- Pre-lab Lecture
 - ❖ Module 2: design overview
 - ❖ Primer design for mutagenesis
 - ❖ Intro to restriction enzymes
 - ❖ Today in Lab: M2D1

Announcements

- Module 1 drafts returned March 22nd/23rd
- Revisions due April 5th/6th (Thu/Fri)
- Draft letter grade may increase by up to 1 $\frac{1}{3}$
- Indicate (highlight, etc.) revisions made

- Introducing... Fahim, your TA for Module 2

Inverse pericam



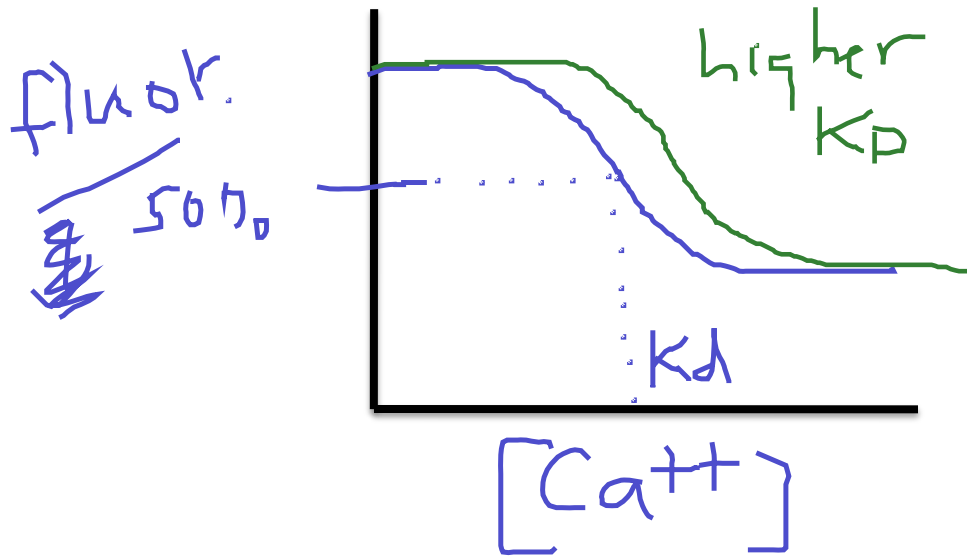
Goal: affect binding properties

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

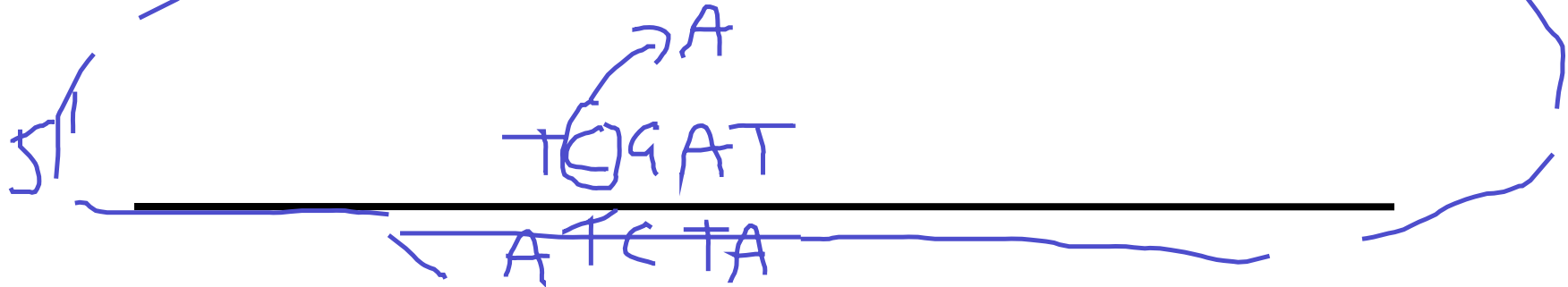
fluor. \propto binding

affinity \downarrow

Cooperativity \uparrow

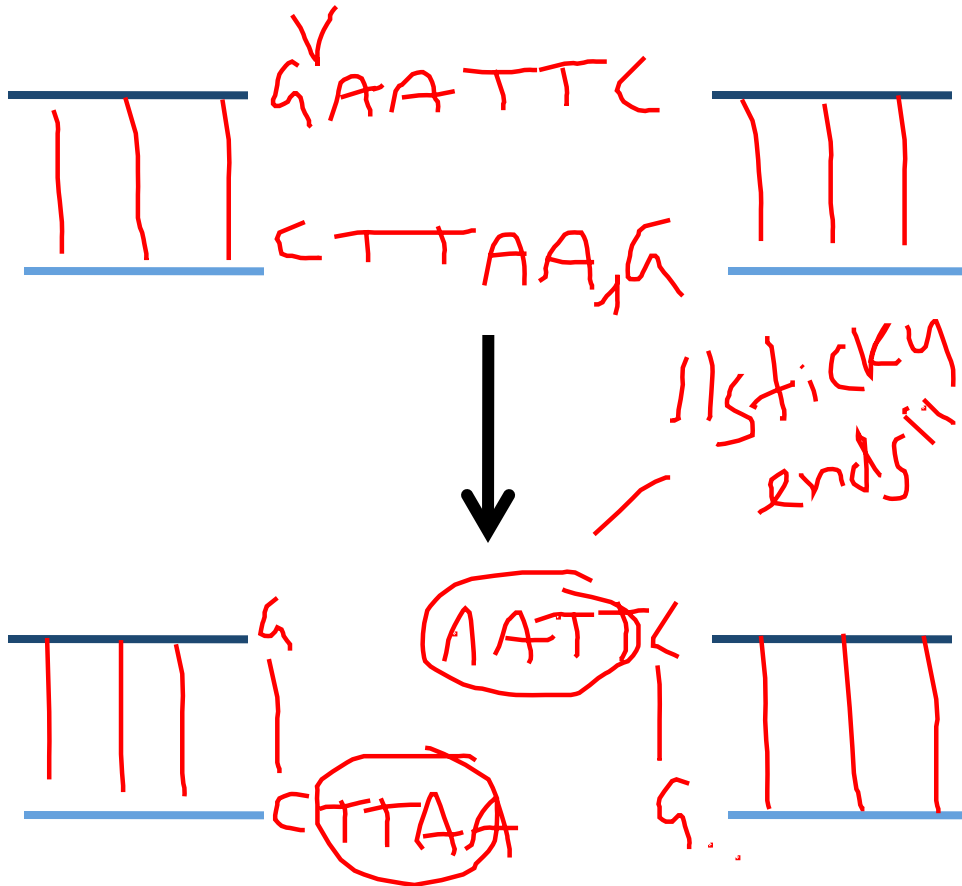


Designing mutagenic primers



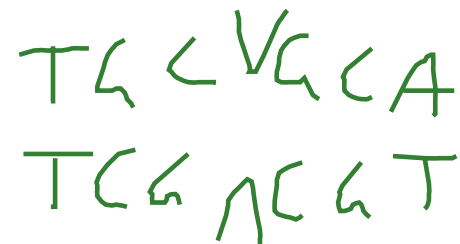
Intro to restriction enzymes

EcoRI



endonucleases
→ cuts DNA

can be used
→ in ligations
→ for analysis
palindromic sites
also blunt cut



Today in Lab: M2D1

- Study inverse pericam at multiple levels

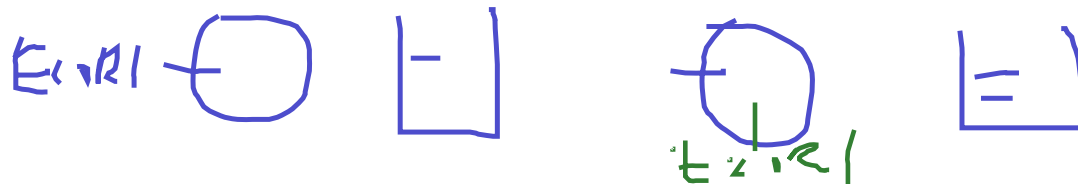
- Design primers

- Amino acid change

- Silent change

SM

make new, unique, restriction site



- Choose positive control: E67K, T79P, M124S
- For next time: read two papers
 - Focus on Nagai; other one is for some history/context
 - Time in class on D2 to re-read your assigned section