Announcements

Lab Practical (~30 min)

Pre-lab Lecture
  - Day 1 Overview
  - Intro to Amplifying DNA
  - Intro to Restriction Enzymes
  - Today in Lab
Announcements

• BE seminar series:
  – Thursdays at 4:05 pm in 32-141
  – Full schedule linked from BE website

• Maintaining our lab space

• Introducing... Naiyan, your TA for Module 1
Inverse Pericam

- **EYFP**
- **cP-EYFP**
- **linker**
- **calmodulin**
- **4 [Ca\(^{2+}\)]-binding sites**
- **bind M13**
Goal: Affect Binding Properties

Δ affinity ∝ \[ [\text{Ca}^{++}] \text{ bulk solution} \] 

Δ cooperativity ∝ \[ \frac{1}{\text{fluor binding}} \]

Vary [Ca^{++}], keeping [CaM] same
DNA Amplification Process

\[
\begin{align*}
\text{ds} & \rightarrow \text{ss} \\
\text{melt} & \\
\text{anneal} & \\
\text{extend} & \\
\text{primers} & \\
\end{align*}
\]
Designing Primers

TCGAT

mutation

A X CTA
Intro to Restriction Enzymes

- EcoRI
- GAATTC
- "sticky" overhangs
- G AATTC
- CTTAA
- palindromic DNA
- Type 2: blunt ends
- TACGAC
- ACCGACAT
- endonucleases cut DNA
Today in Lab

• Study inverse pericam at multiple levels
• Design primers
  – Amino acid change
  – Silent change
    ➔ add new restriction site
2nd primer: S101L
• For next time: get familiar with scope of module, OpenWetWare, read two papers
  – Read figures, skim text, then do a close read