

- Announcements
- Lab Quiz
- Pre-lab Lecture
 - ❖ Library construction
 - ❖ Registry of Std Bio Parts
 - ❖ BP analogue from EE
 - ❖ Today in Lab (Mod 2 Day 3)

Announcements

- Next time: come to *lab* first to set up exp.
(we'll go together to 16-336 for j. club talks)
- HW comments

$$1 \text{ Miller Unit} = 1000 * \frac{(Abs420 - (1.75 * Abs550))}{(t * v * Abs600)}$$

might be out of range

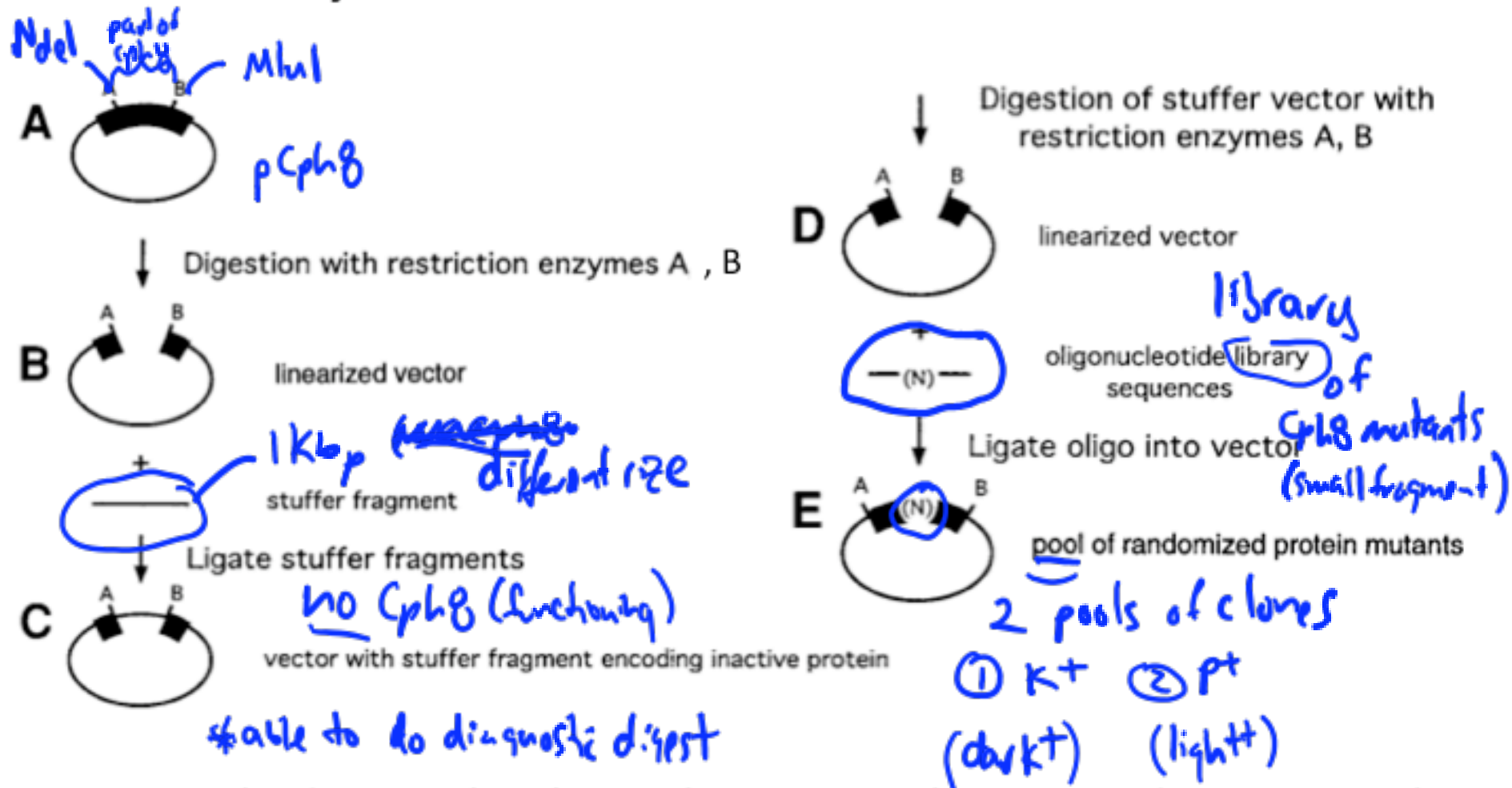
small measurement errors

v = cell culture volume in mL, 0.1 in our assay; t = time in min

* expect \approx same (take dilution into account)

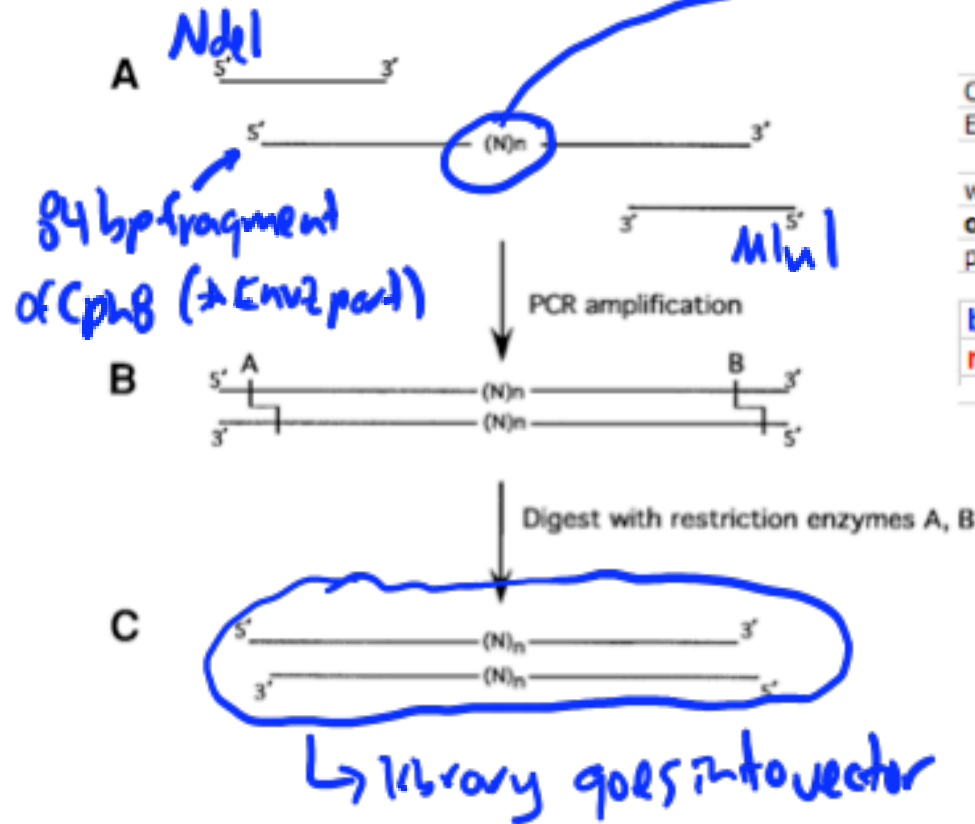
* no error (not absolute Δ , r.t.)

Library construction: macro view



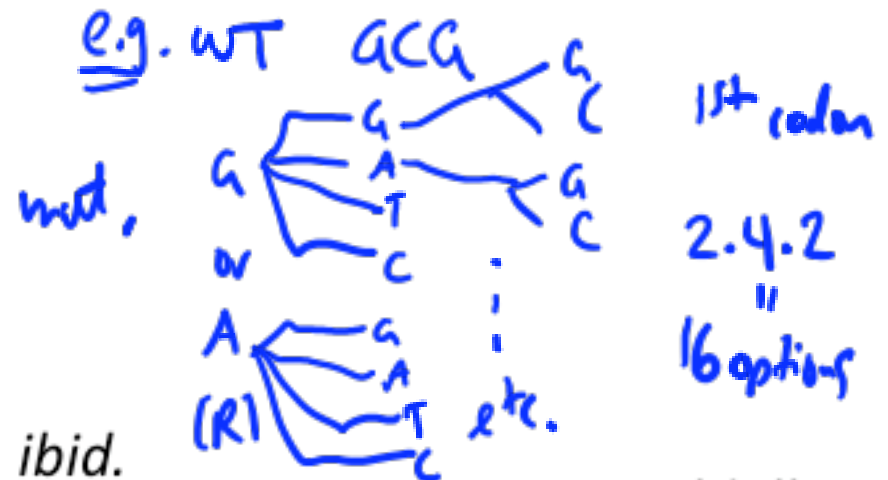
From *Methods in Molecular Biology* 231:65 by JL Sneed & LA Loeb
(Chapter 9, "Random Oligonucleotide Mutagenesis")

Library construction: micro view



Semi-randomized 15bp = 50aa

Cph1/EnvZ	A553	G554	V555	S556	H557
EnvZ	A239T	G240E	V241G	S242D	H243A
wt seq	GCG	GGG	GTA	AGT	CAC
oligo seq	RNS	RNS	RNS	RNS	SNW
poss aa	Val	Val	Val	Val	Val
	Ala	Ala	Ala	Ala	Ala
blue = K-P+	Asp	Asp	Asp	Asp	Asp
red = K+ P-	Glu	Glu	Glu	Glu	Glu
	Gly	Gly	Gly	Gly	Gly
	Tle	Tle	Tle	Tle	Tle



From *Methods in Molecular Biology*, *ibid.*

From N. Kuldell

Registry of Std Bio Parts

Goal: design light generator
that functions in *E. coli*

Browse parts by type

Catalog

List



Promoters (?): A promoter is
of the downstream DNA seq.



RBS (read)
Ribosome Binding Sites (?)
can bind and initiate translati



Protein domains (?): Protein
up a protein coding sequence
target the protein for cleavag



Protein coding sequences
Note that some protein codin
protein from start codon to st
also included here.



Translational units (?): Tran
They begin at the site of tran

levels of abstraction

Browse parts and devices by function

This section replaces the previous *Featured parts* pages.



Biosynthesis: Parts involved in the producti



Cell-cell signaling and quorum sensing: F



Cell death: Parts involved in killing cells.



Coliroid: Parts involved in taking a bacterial

Browse parts and devices by standard

Unless otherwise specified, most parts in the Registr

EX-SP
Assembly standard 10 (?): Assembly standard 10, i
comply with this assembly standard.

Assembly standard 23 (?): Assembly standard 23, i

Images from parts.mit.edu

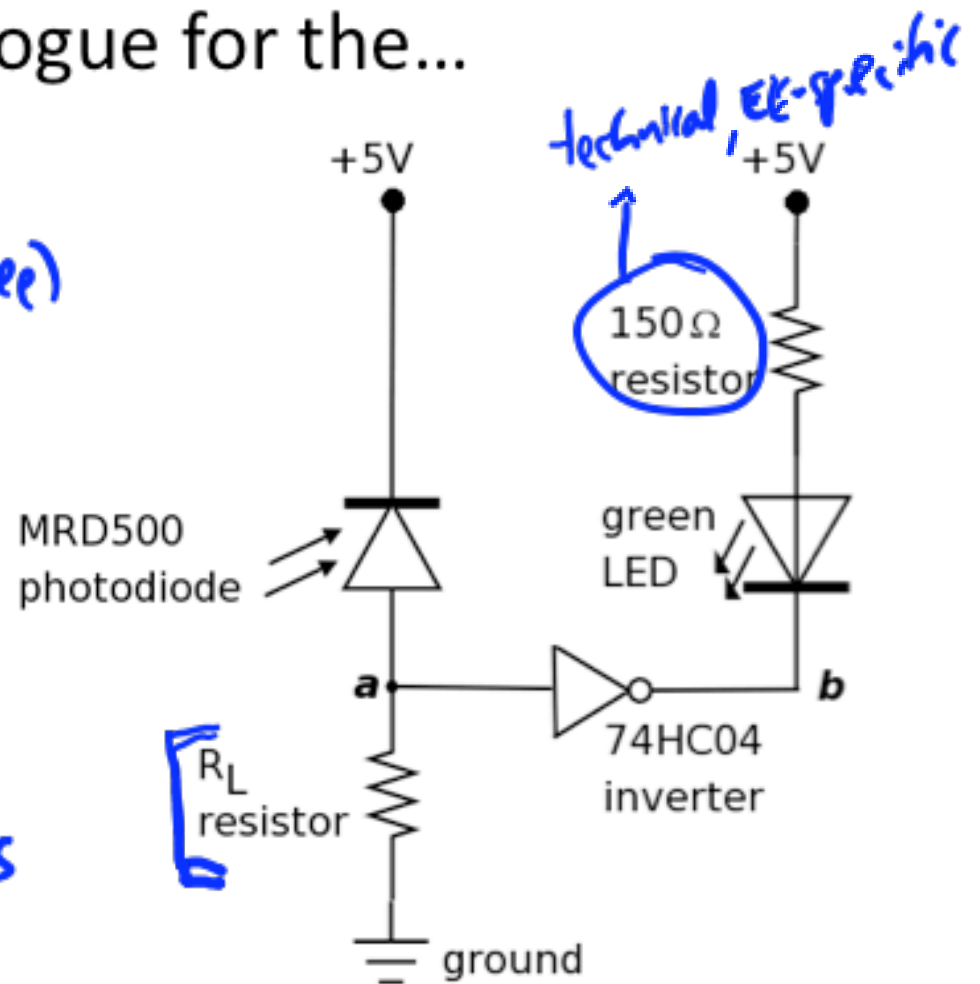
EE analogue to BP system

What is the genetic analogue for the...

- Photodiode? C_{ph}
- LED? $I_{ac} Z$ (+ subnode to sep)
- Inverter? $D_{mp} R$

Goal: vary R_L – what is the outcome?

- calculations
- empirical
- limit cases
- $R \sim 0$
- $R \sim \infty$



Today in Lab

Observe/document *coli*roid from last time



Set up library screen

→ choose Kt or Pt DNA library

1. electroporate

2. plate cells



incubate 1 hr*



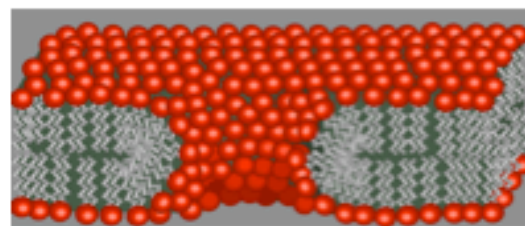
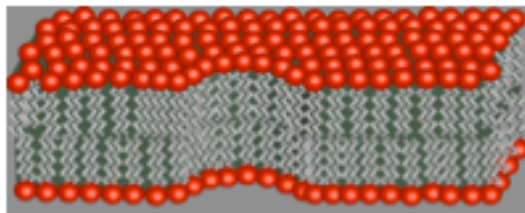
Explore EE analogue

Explore parts.mit.edu

has PCB, lacZ

but no wt pCphB

*transfer to SOC medium
(has extra sugar, etc.)



Membrane model from Wikimedia Commons, public domain image