System Engineering

20.109(F11)

M2D6 lecture

11.01.11

Ceiling for engineering complex biological systems?

Some explanations:
- Not enough parts?
- Insulation?
- Rules for composition?

Figure from **Nat Rev Mol Cell Biol (2009)** 10(6):410-422

The C-dog

DNA sequence → amino acid sequence of protein

http://publications.nigms.nih.gov/thenewgenetics/images/ch1_trans.jpg

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Beyond the C-dog

protein stability?
protein activity?
cross-talk with other systems?

http://publications.nigms.nih.gov/thenewgenetics/images/ch1_trans.jpg

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How could a change in protein stability affect β-gal?

	# autophos Cph8	Kinasing rate	P-tase rate	Amount OmpR+P
Wild type	5 "units"	3 "units"	2 "units"	1

SDS-Polyacrylamide Gel Electrophoresis (SDS-PAGE)

Loading dye has glycerol, SDS, reducing agent

Samples boiled before loading

SDS-Polyacrylamide Gel Electrophoresis (SDS-PAGE)
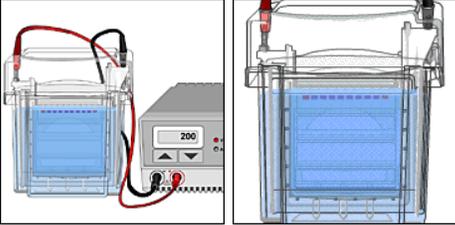
http://www.bme.gatech.edu/vcl/SDS_PAGE/Background/Introduction.htm

Detection through Western Analysis

Gel Electrophoresis

Transfer Membrane
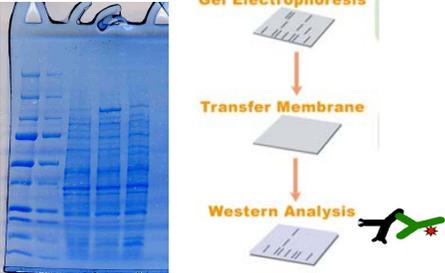
Western Analysis

http://www.genscript.com/product_001/western_application/grp_id/60065/op/detail/Uvrag_Antibody_Analysis.html

Beyond the C-dog
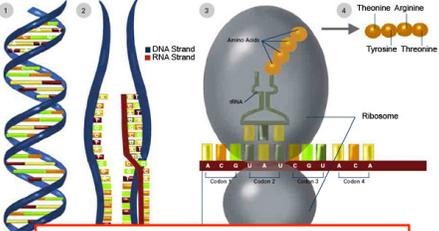
protein stability?
protein activity?
cross-talk with other systems?

http://publications.nigms.nih.gov/thenewgenetics/images/ch1_trans.jpg

Possible mutant "phenotypes"

β-galactosidase Activity (Miller Units)

- dark
- light

WT, M1, M2, M3, M4, M5

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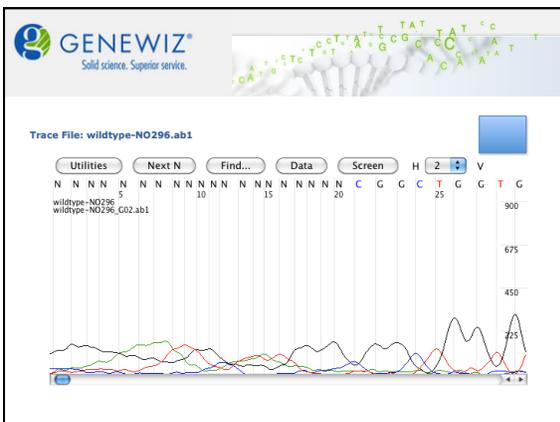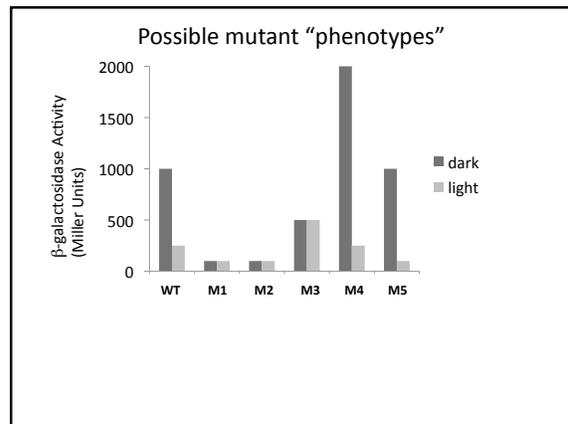
Summary

	# autophos Cph8	Kinasing rate	P-tase rate
Wild type	5 "units"	3 "units"	2 "units"
P+	5 "units"	3 "units"	3 "units"
Unstable	0 "units"	0 "units"	0 "units"