

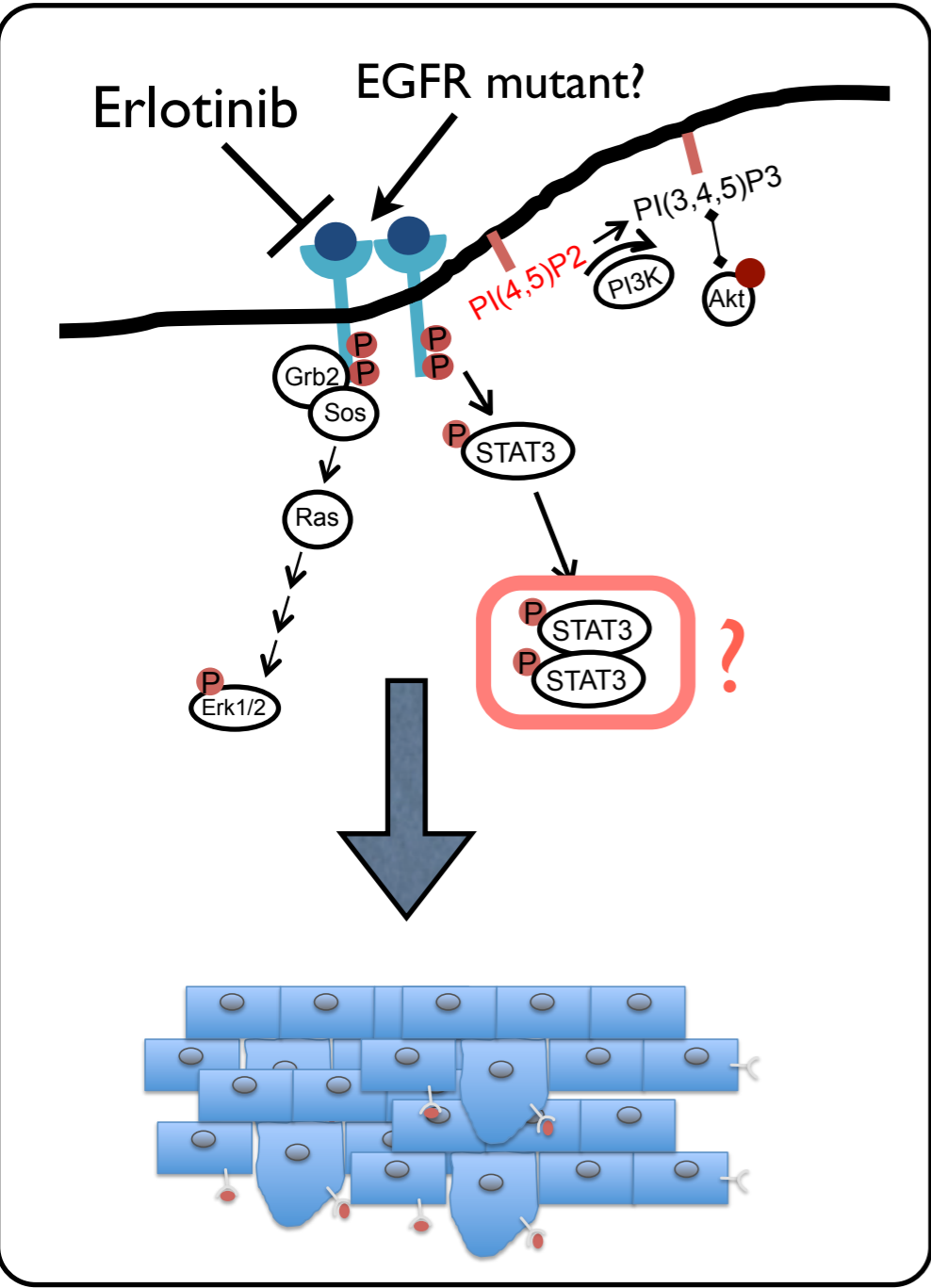
Module 2: Systems Engineering (M2D7)

- The robot is a go
- Summary thus far -- let's look at some WB data
- $6 \times 6 \times 2 (\times 3)$ = a lot of data to visualize
- How will we compare across conditions?
- Guest speaker + Data analysis 'office hour' next lecture

~ 35 min

Module 2 overview:

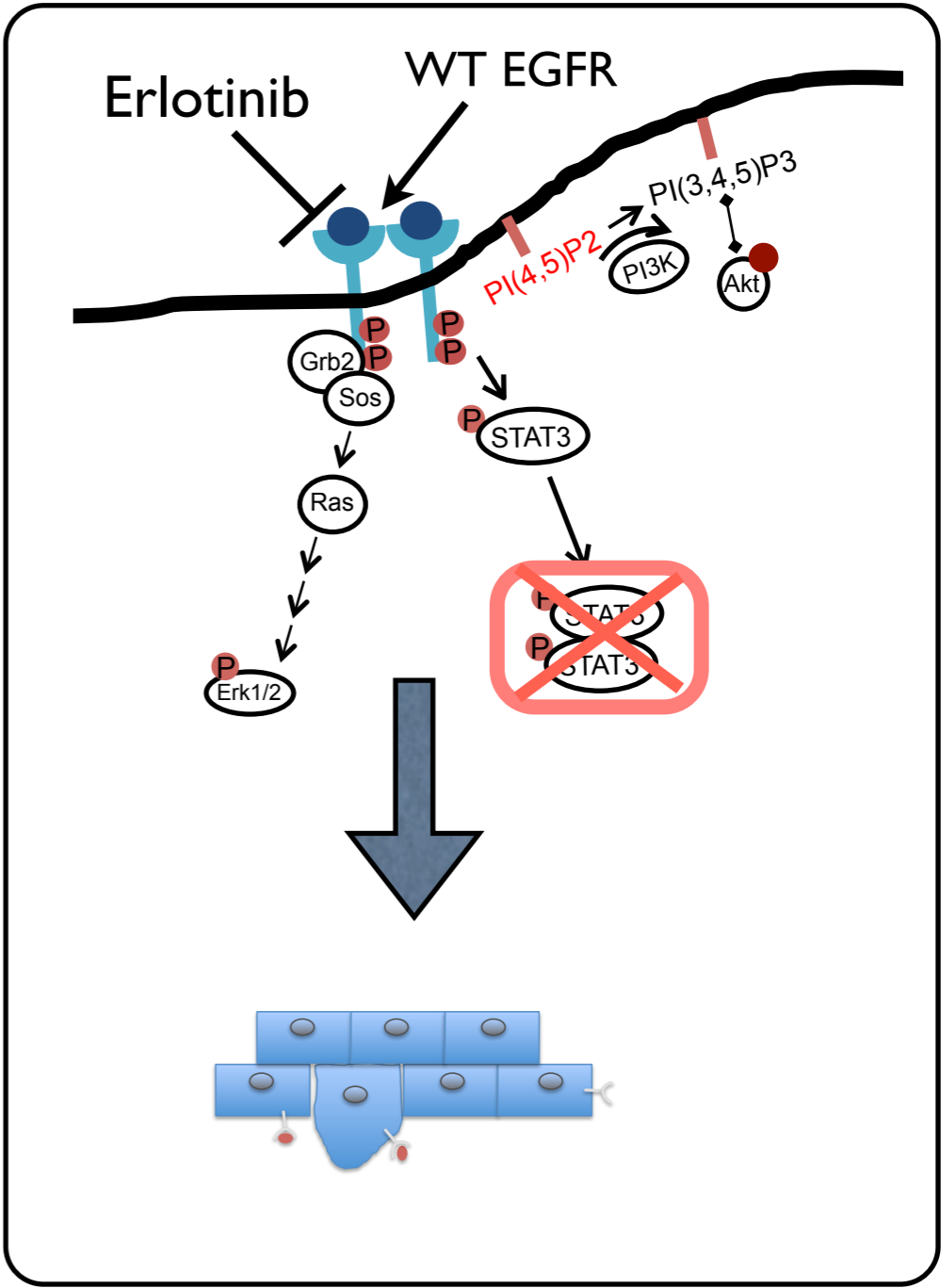
★ WB ★



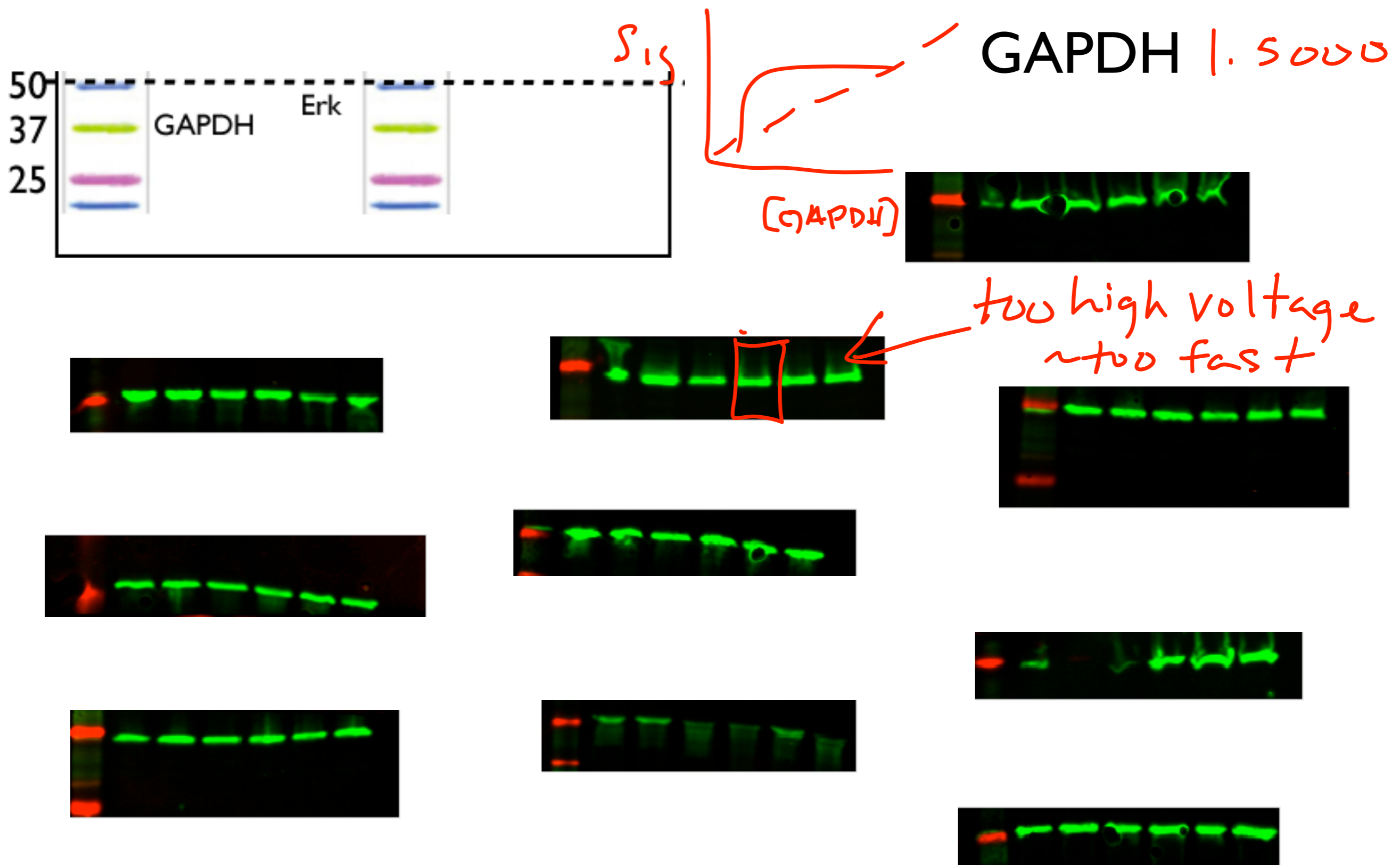
Mutation Analysis

WB Analysis

Viability Analysis

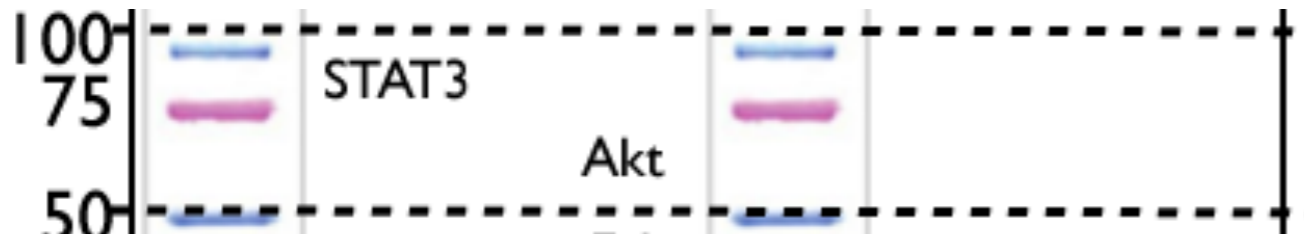


Semi-quantitative analysis: Your experiment

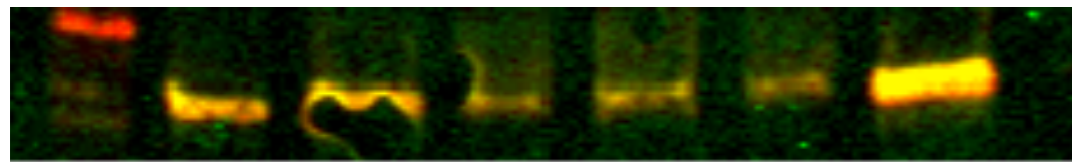


Semi-quantitative analysis: Your experiment

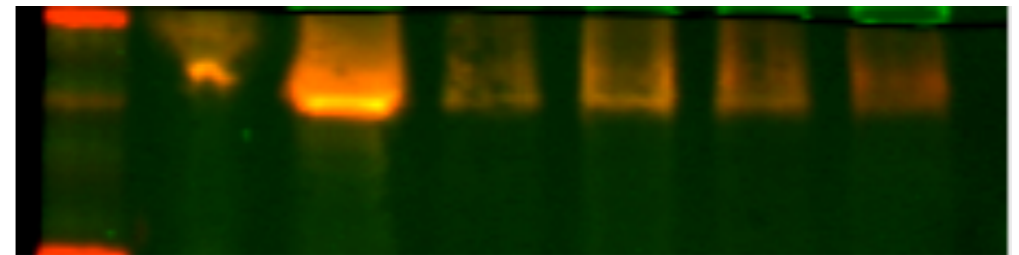
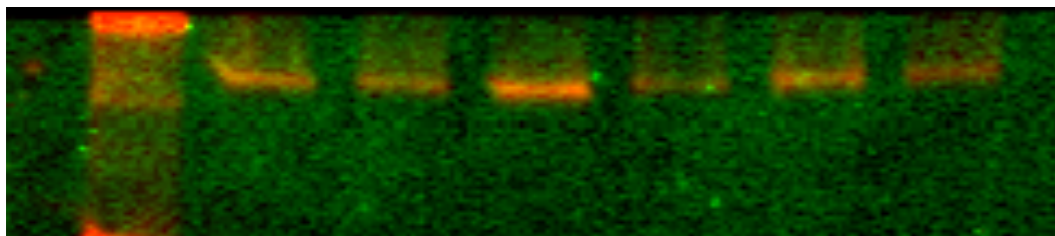
STAT3



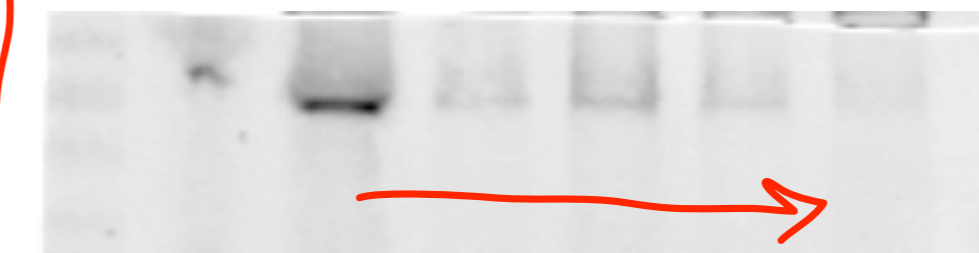
Qualitative -
Erlotinib doesn't



Δ PSTAT3

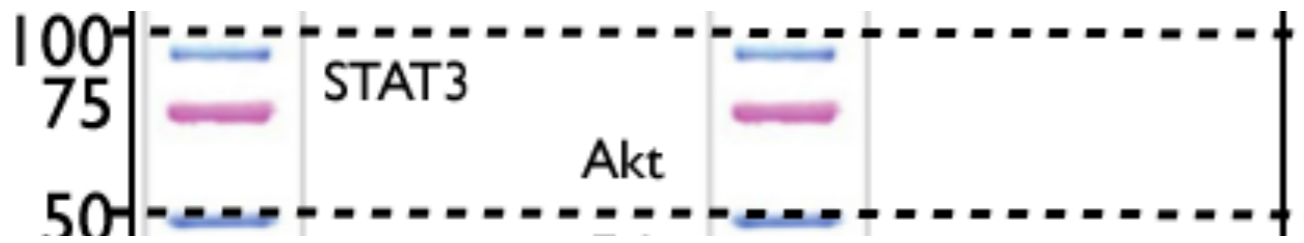


Total ^{Red}

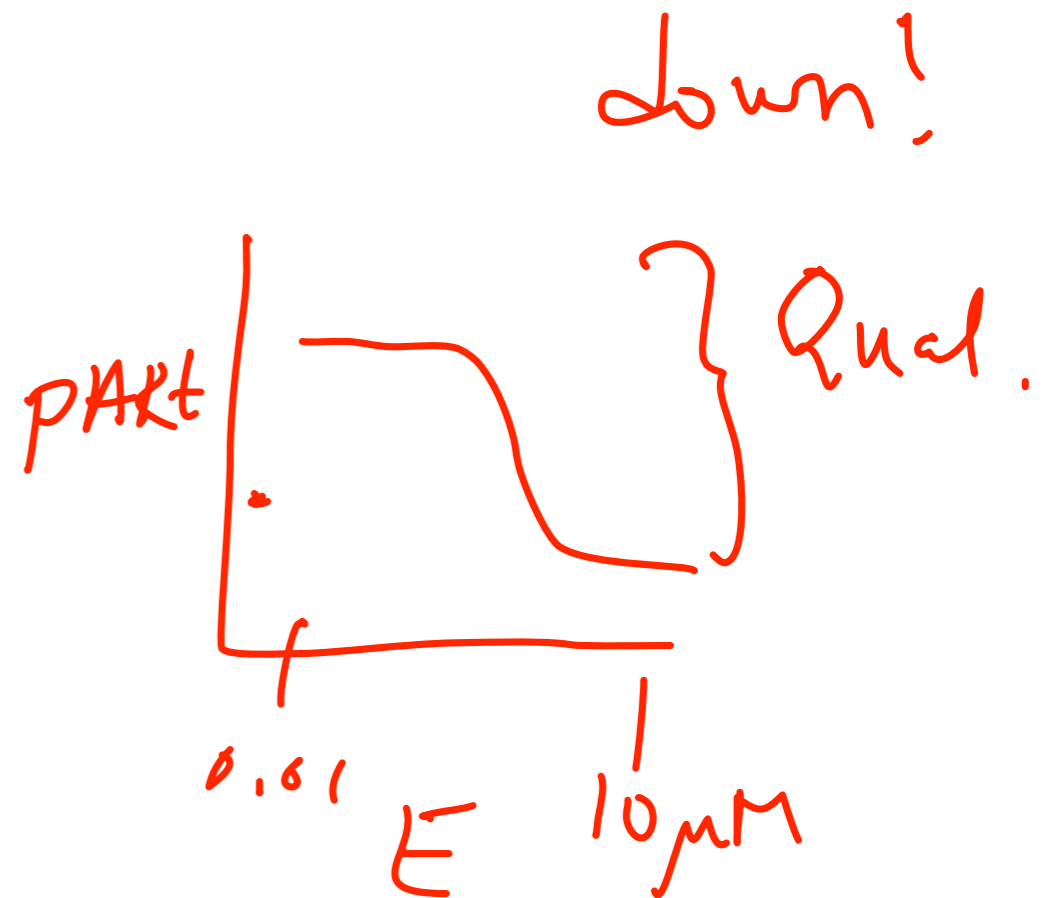
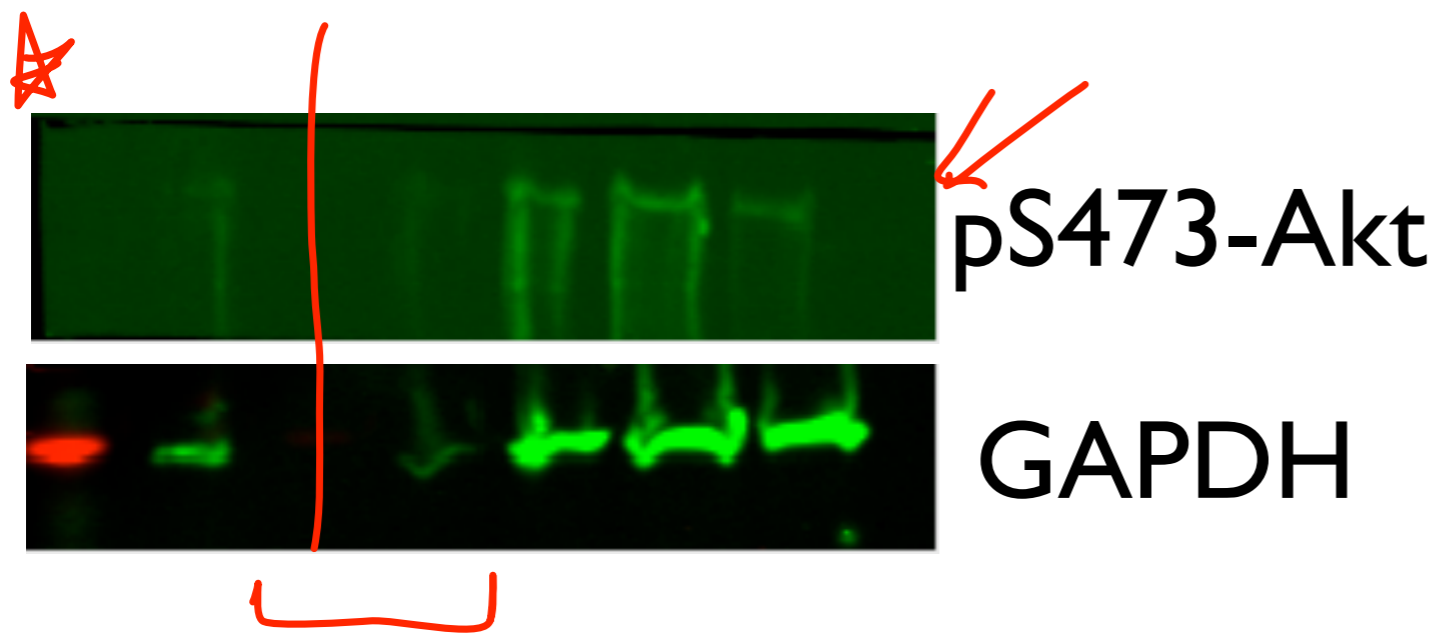
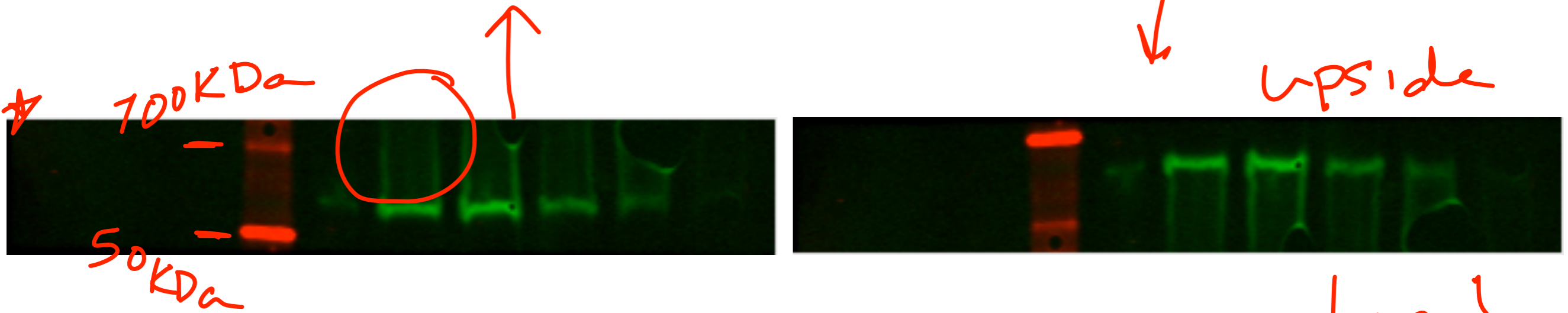


pY705
^{Green}

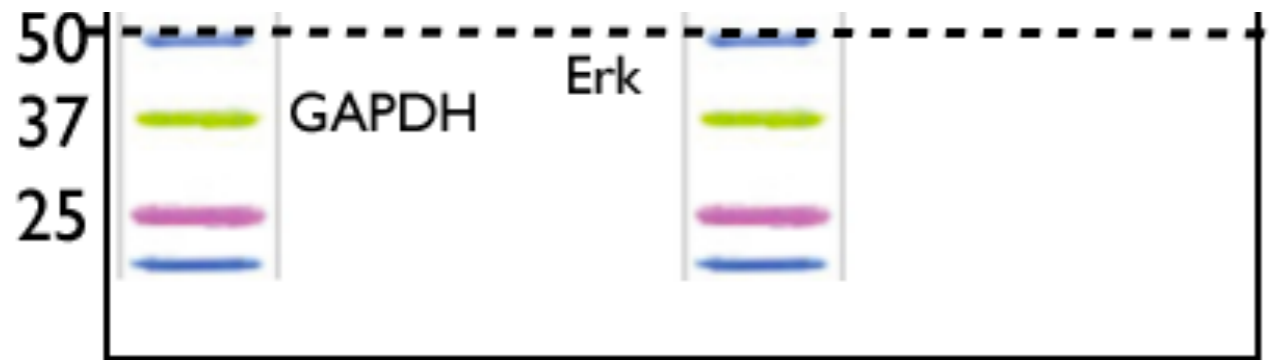
Semi-quantitative analysis: Your experiment



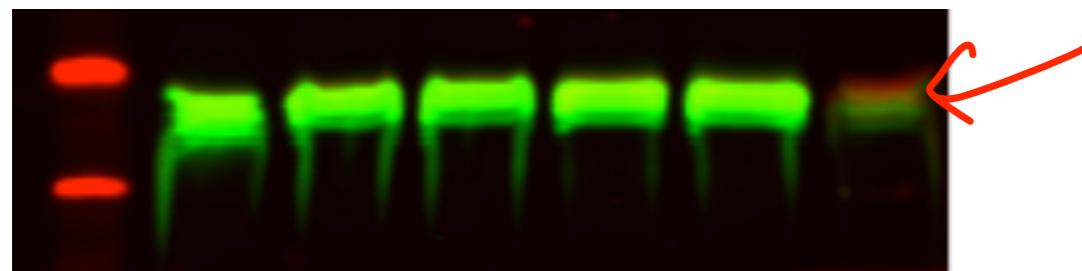
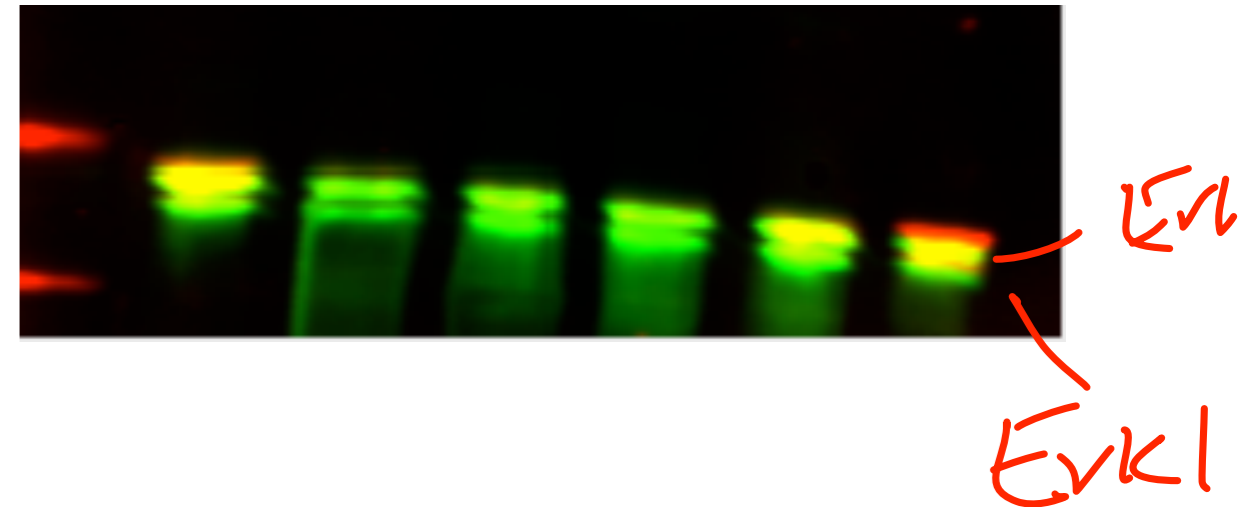
P Akt - green
↳ upside



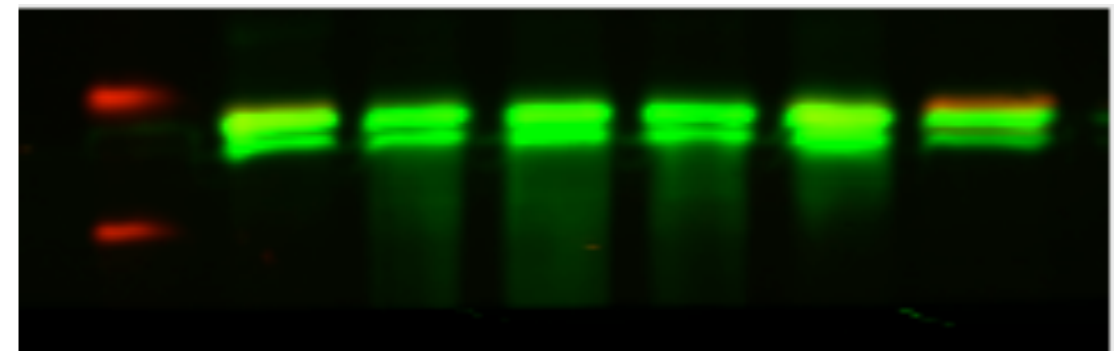
Semi-quantitative analysis: Your experiment



Erk



2 TBD
d

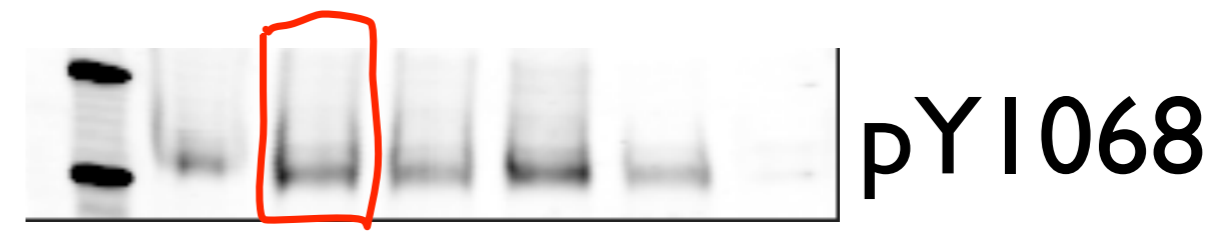
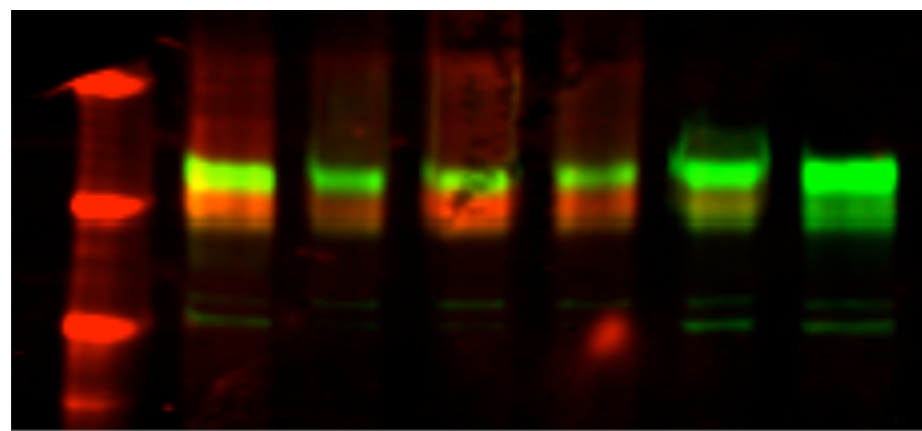
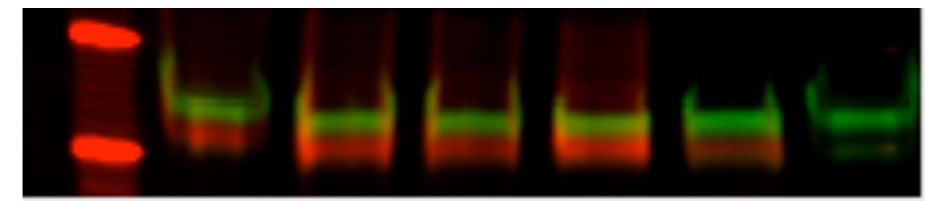


ppERK - green
ERK - Red

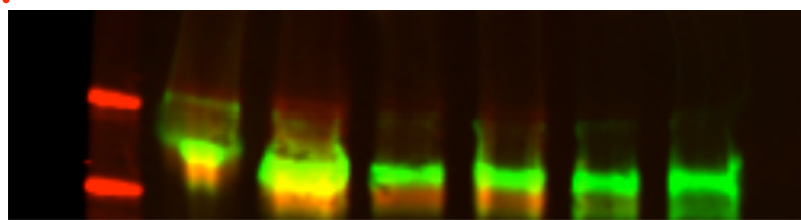
Semi-quantitative analysis: Your experiment



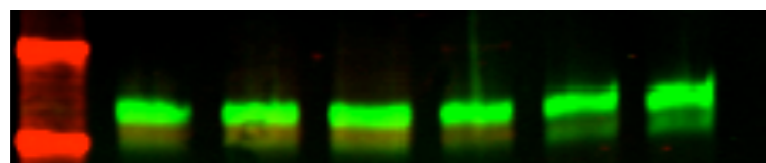
total - green EGFR
pY1068 - red



↪

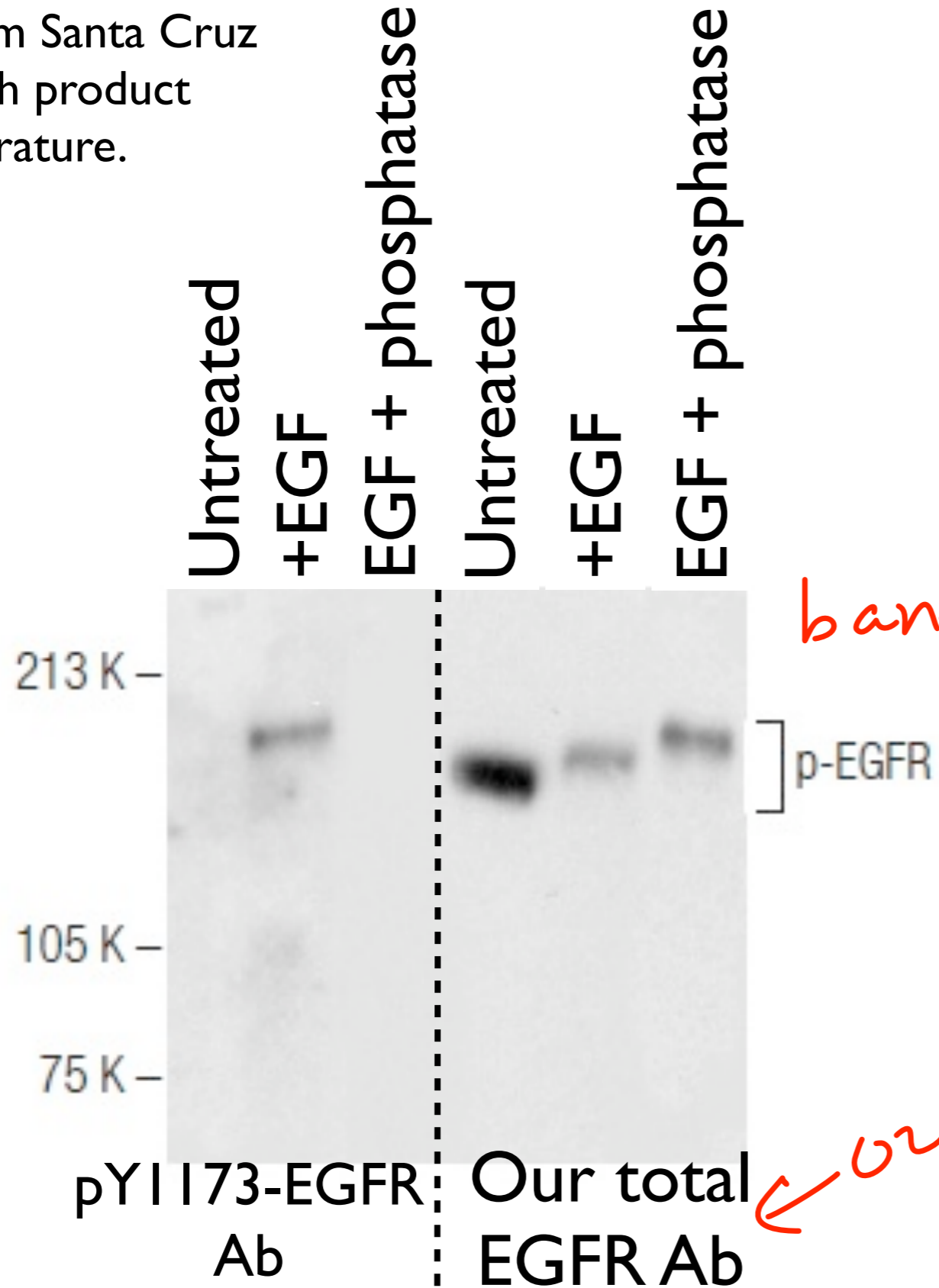


• pH! ← salt imbalance
 ↖ lab water



Semi-quantitative analysis: Your experiment

Image from Santa Cruz
Biotech product
literature.

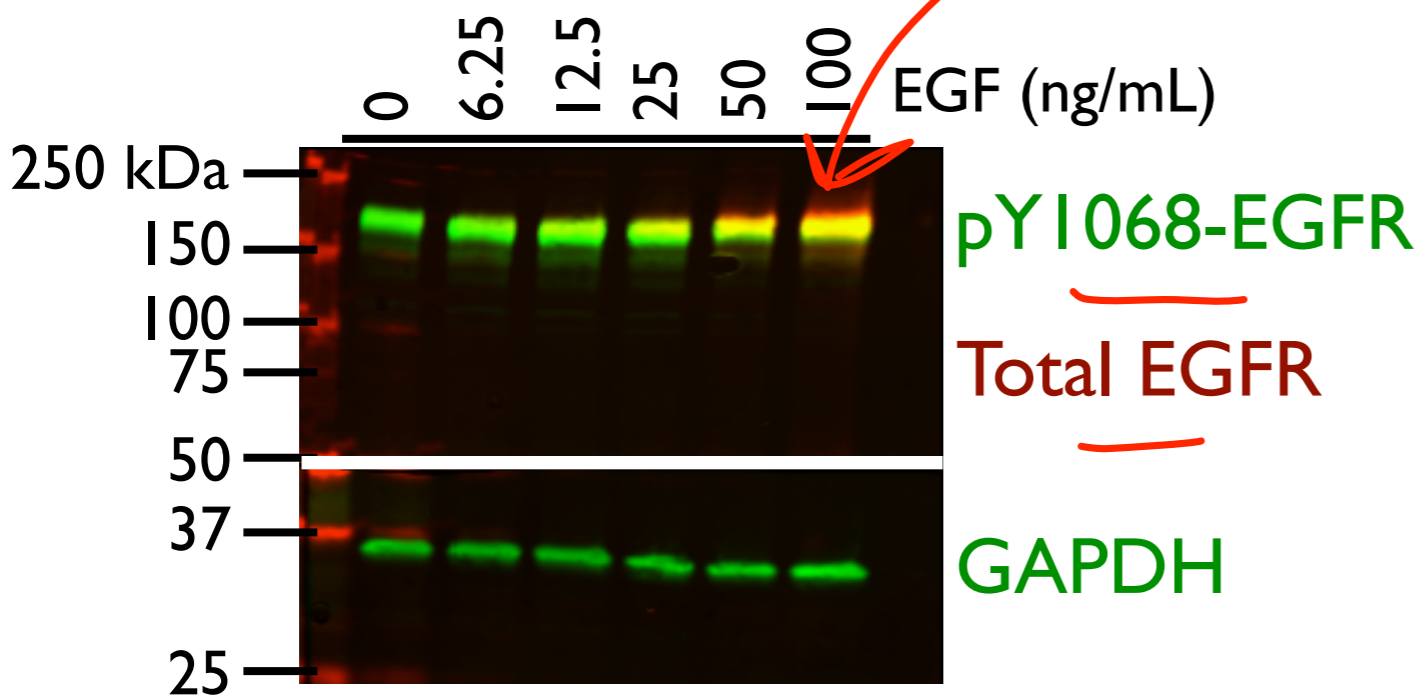


band shifts are "OK" -
but need to
understand why!

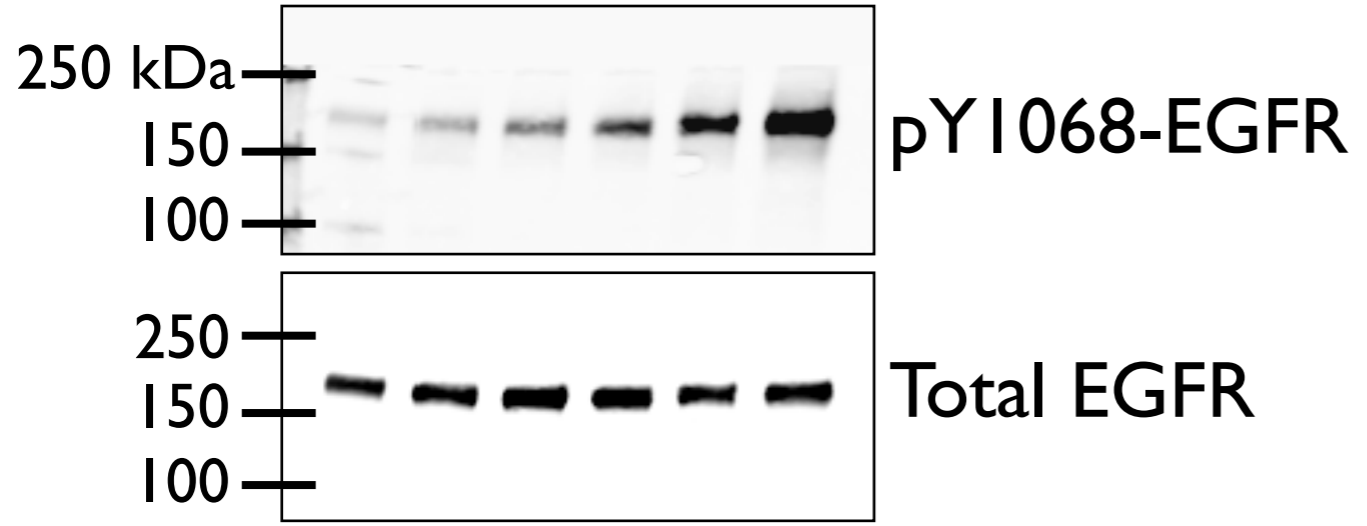
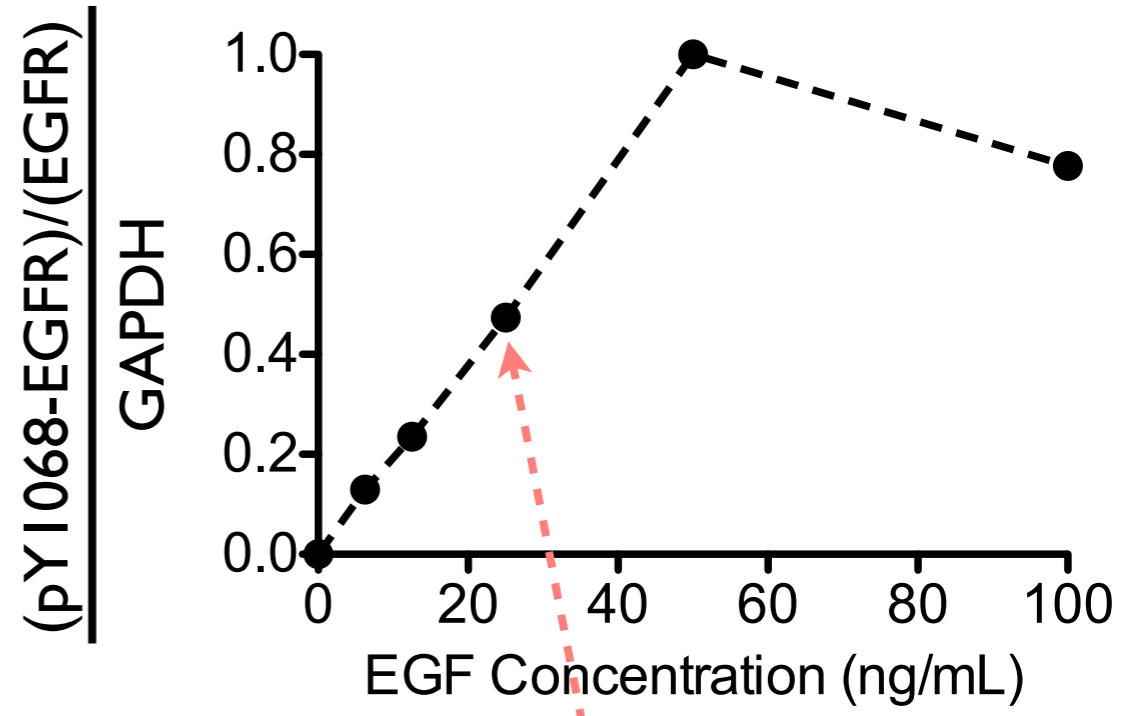
← ours

Semi-quantitative analysis: Western blot

Same cells → different buffer



Densitometric Analysis



*Warning \$\$\$

SKOV3

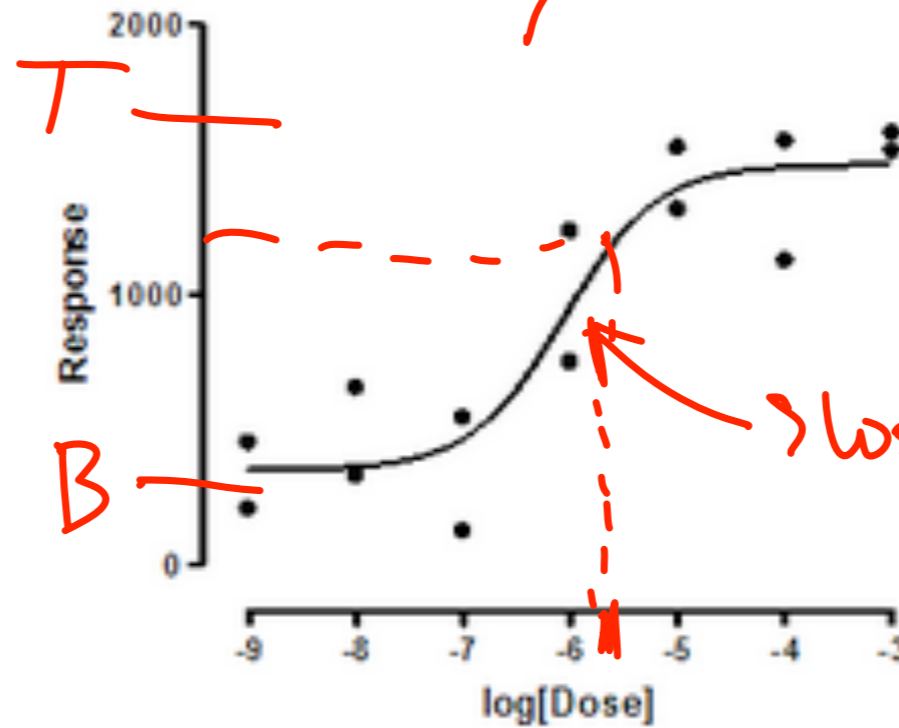
Non-linear curve fitting: Logistic Fit

How to quantify:

Logistic fit

4 parameters

Non-linear
curve fitting



Dose-Response

|| growth factor

|| culturing conditions

slope

Hill coeff.

$$Y = \text{Bottom} + (\text{Top} - \text{Bottom}) / (1 + 10^{((\text{LogEC50} - X) * \text{HillSlope}))})$$

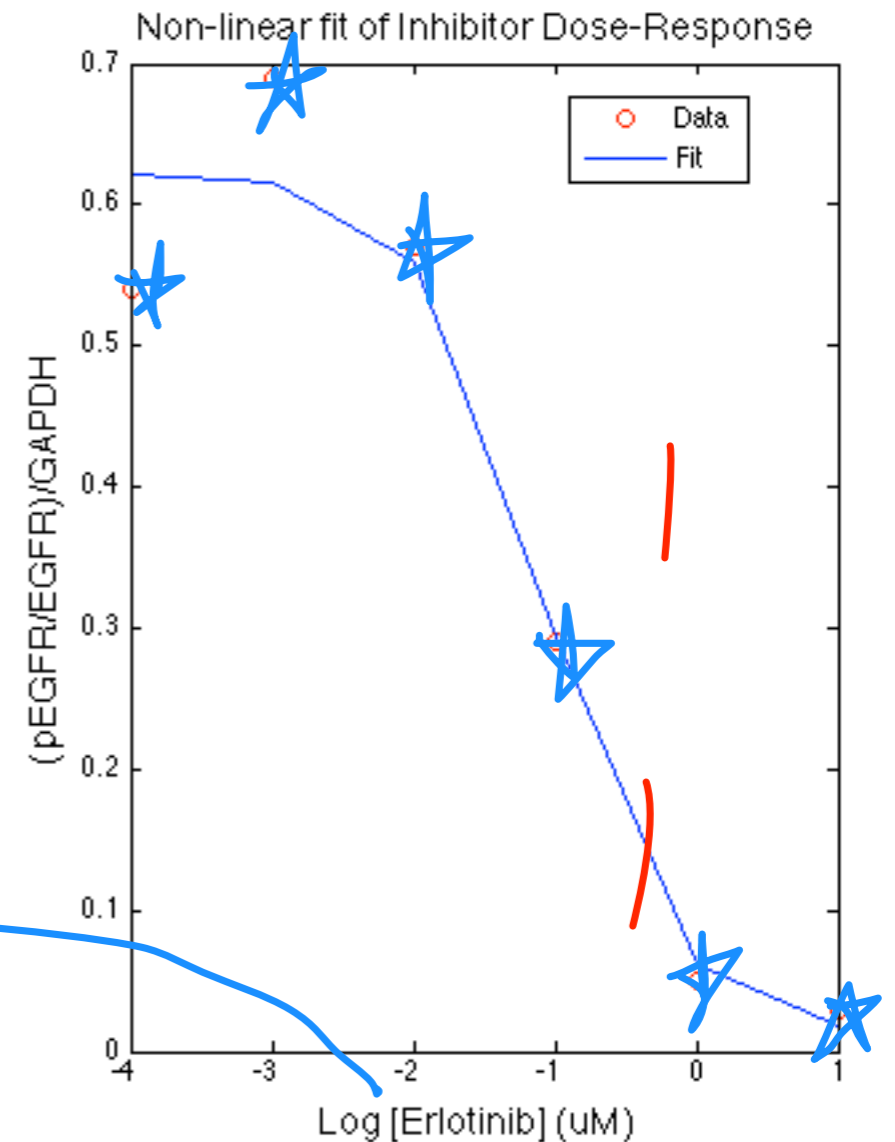
Relative

Non-linear curve fitting: Logistic Fit

How to quantify:

assume Hill = 1

$\log[X]$
~~long~~ ←



$$Y = \text{Bottom} + \frac{\text{Top} - \text{Bottom}}{1 + 10^{-(X - \text{IC}_{50})}}$$

Log IC50

Non-linear curve fitting: Logistic Fit

How to quantify:

$$r^2$$

$$r = y - \hat{y}$$

$$SS_{res} = \sum (y - \hat{y})^2$$

$$SS_{total} = \sum (y - \bar{y})^2$$

$r^2 =$ goodness of fit =

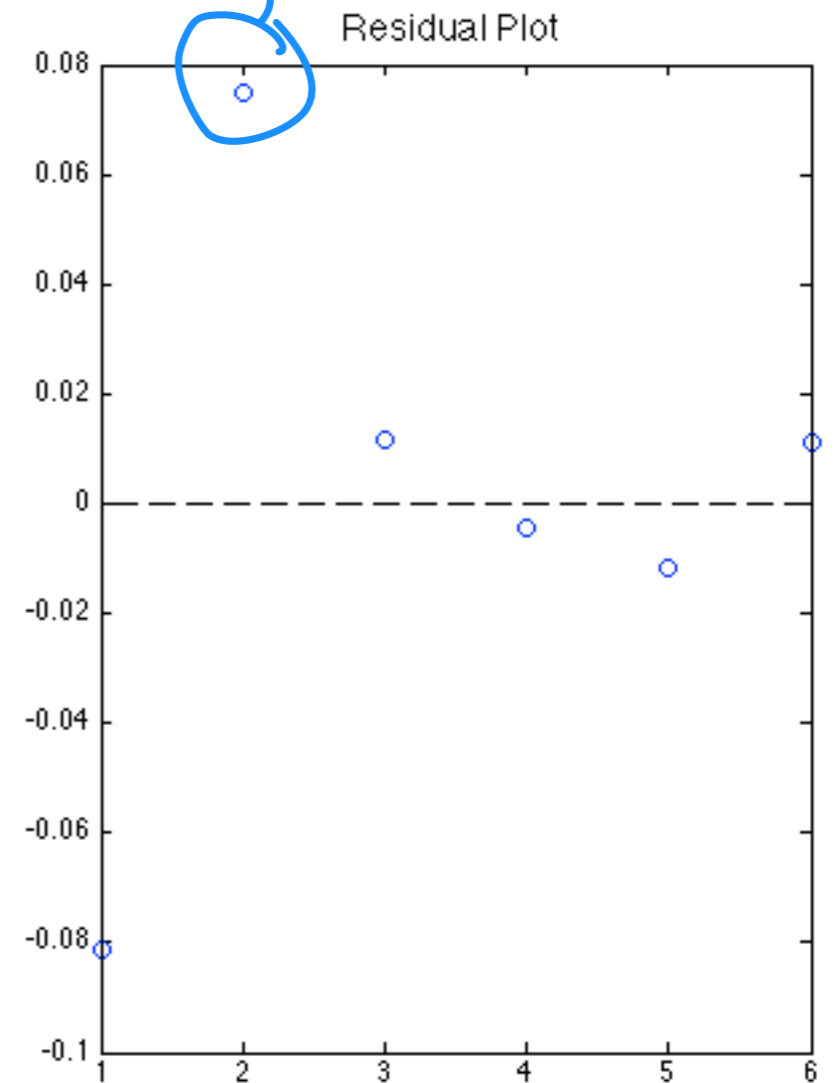
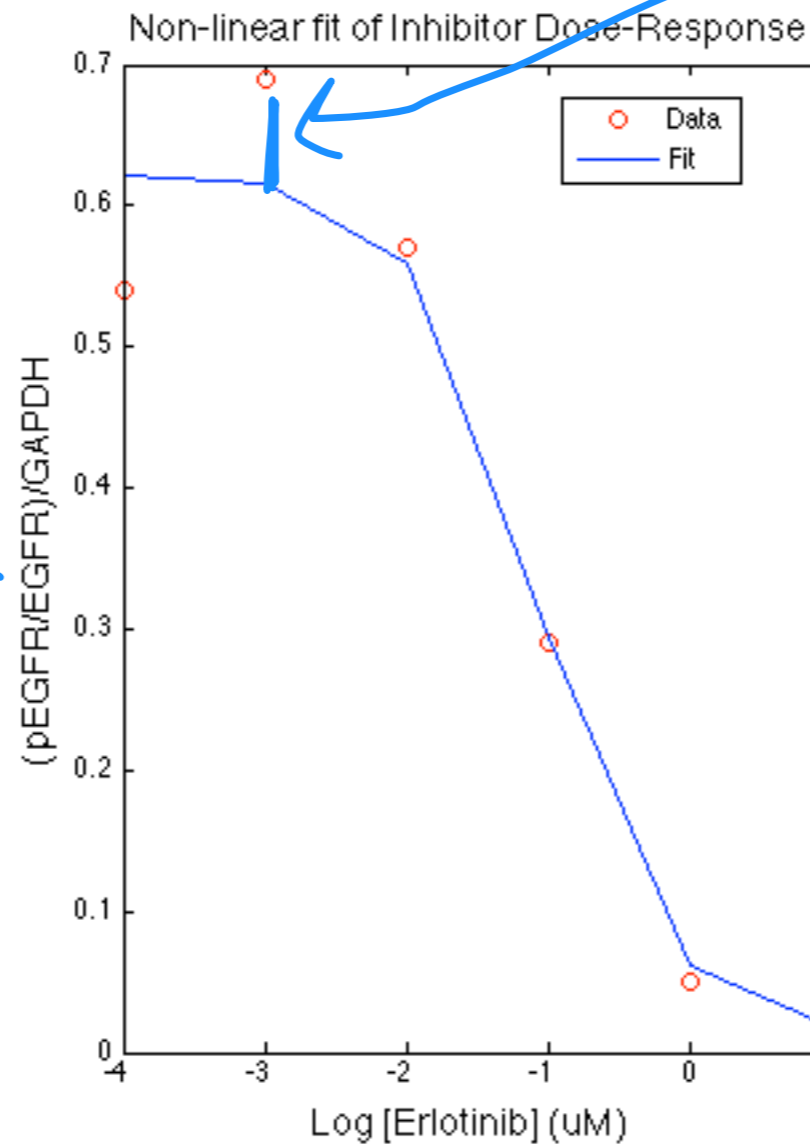
$$1 - \left(\frac{SS_{res}}{SS_{total}} \right)$$

$$Y = \text{Bottom} + (\text{Top} - \text{Bottom}) / (1 + 10^{-(X - IC50)})$$

$r^2 = 1$ perfect

$r^2 = 0$ v. bad

$r^2 > 0.5$



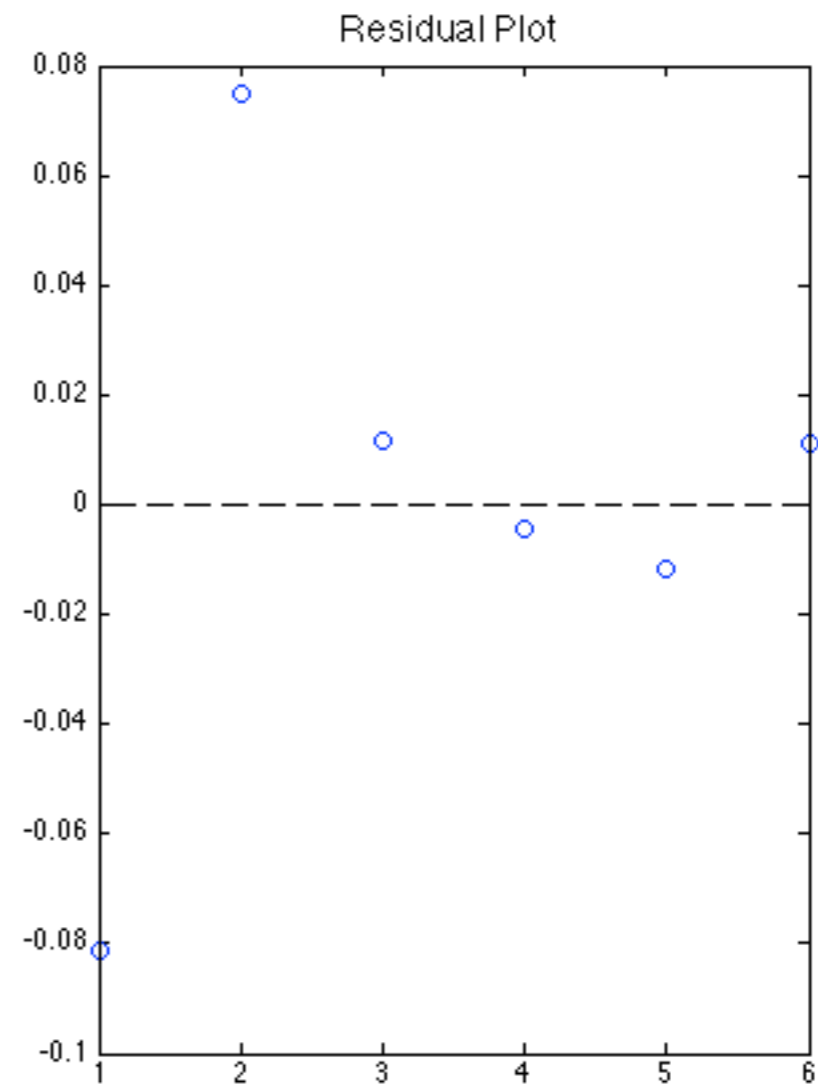
Non-linear curve fitting: Logistic Fit

How to quantify:

Estimated IC50 from nlinfit is: 0.085619 μM

85.6 μM

The lower 95 percent CI bound is: 0.016751 μM
The upper 95 percent CI bound is: 0.437613 μM



IC50 \rightarrow [E]

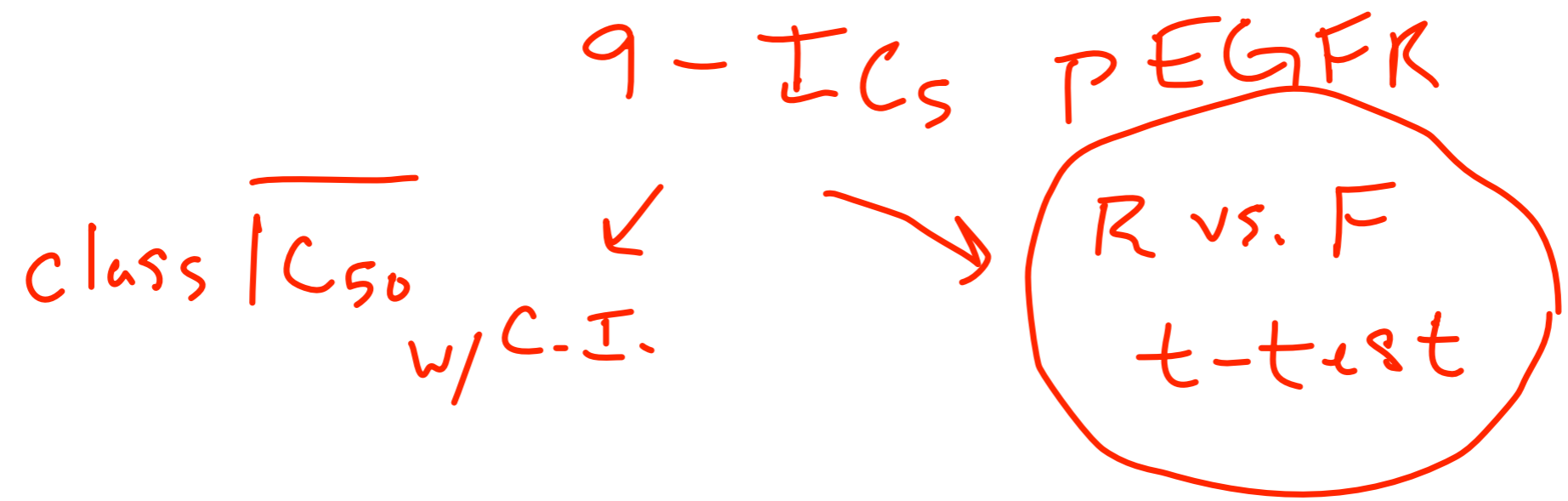
~~IC50~~

$$Y = \text{Bottom} + (\text{Top} - \text{Bottom}) / (1 + 10^{-(X - \text{IC50})})$$

What are the interesting comparisons?

WB → How does IC_{50} w.r.t. PEGFR
compare protein X?

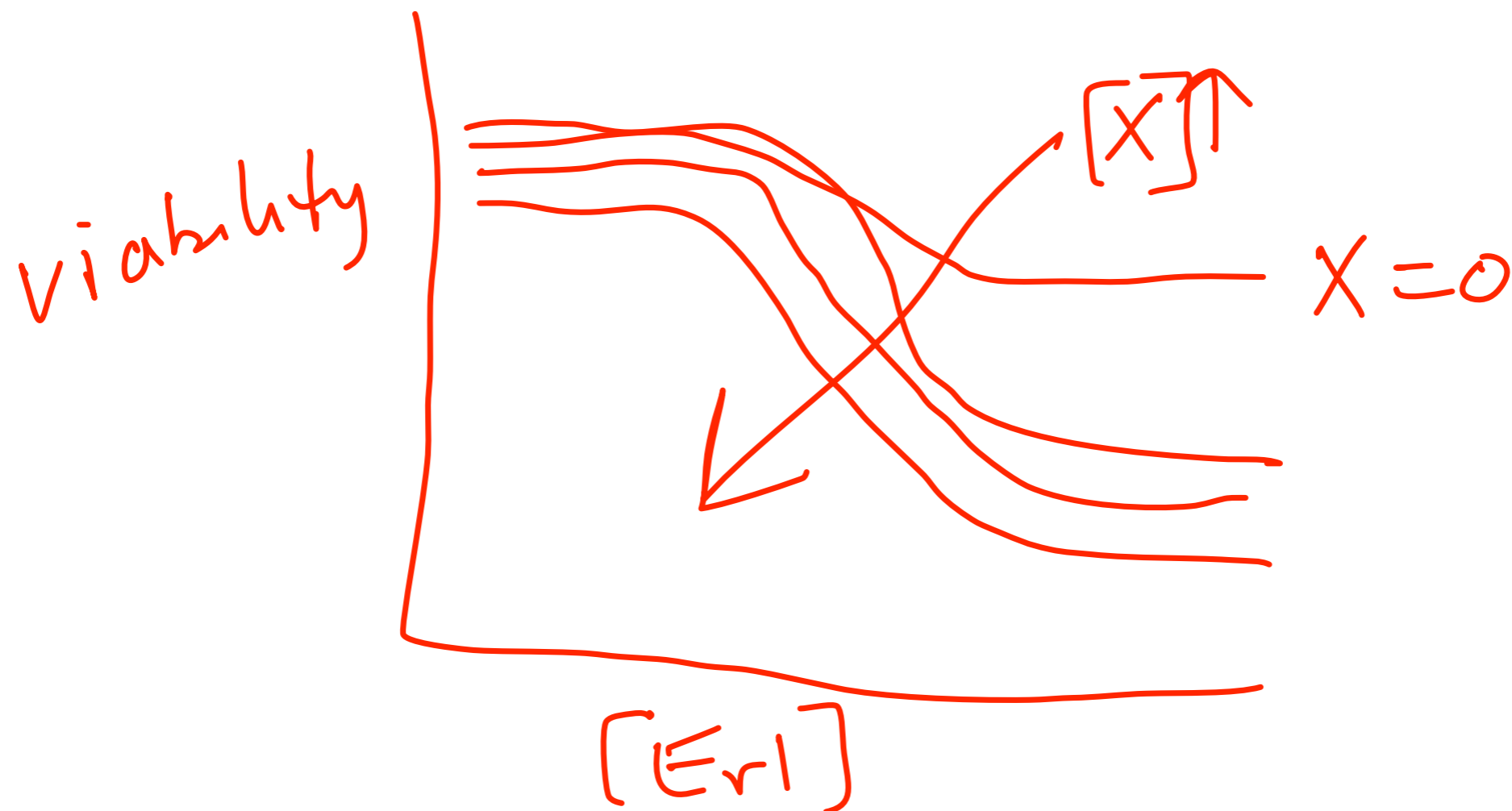
How consistent are the IC_{50}
across class?
days?



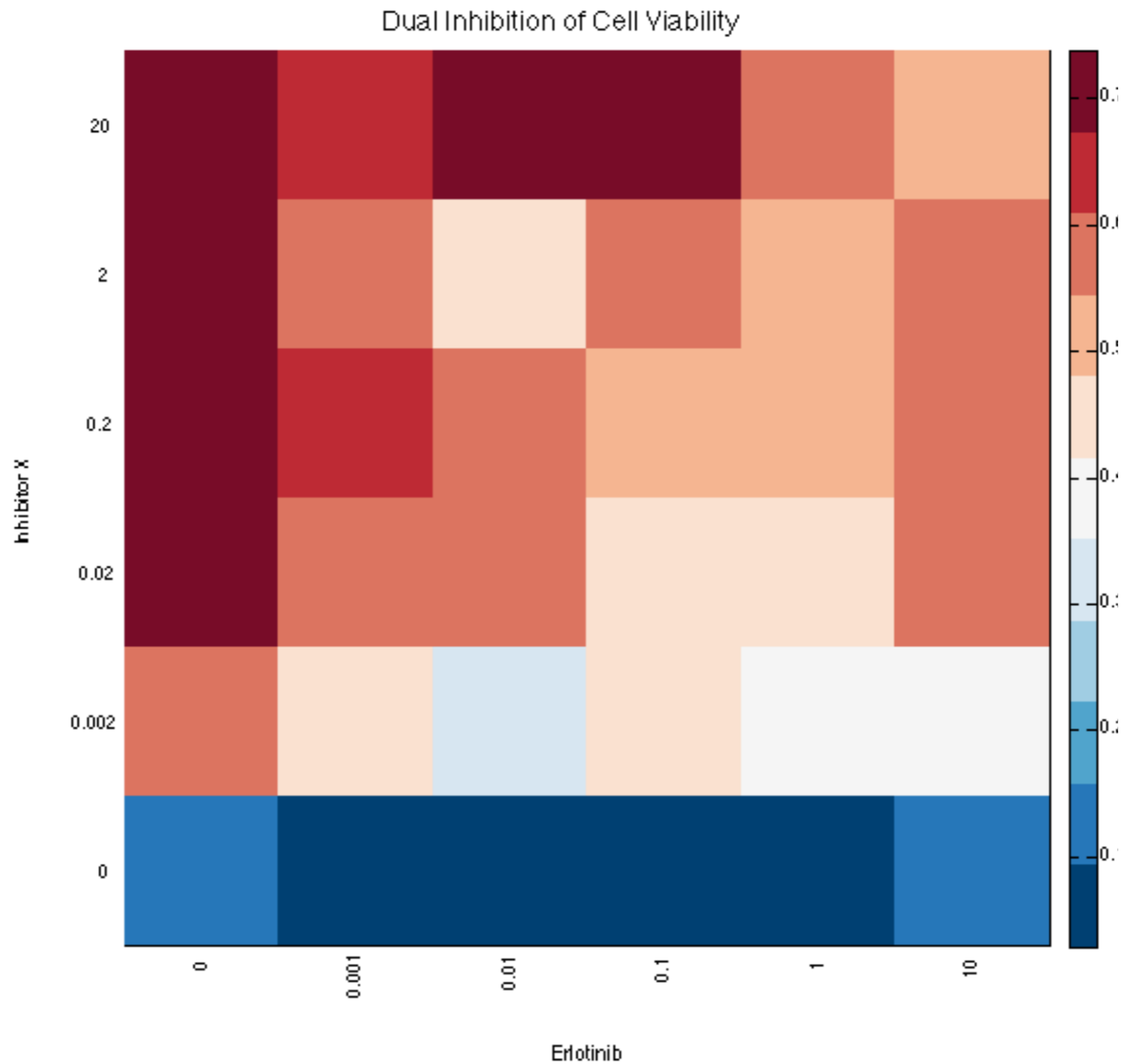
The dual inhibition study can be analyzed in a similar way.

Recommend starting with:

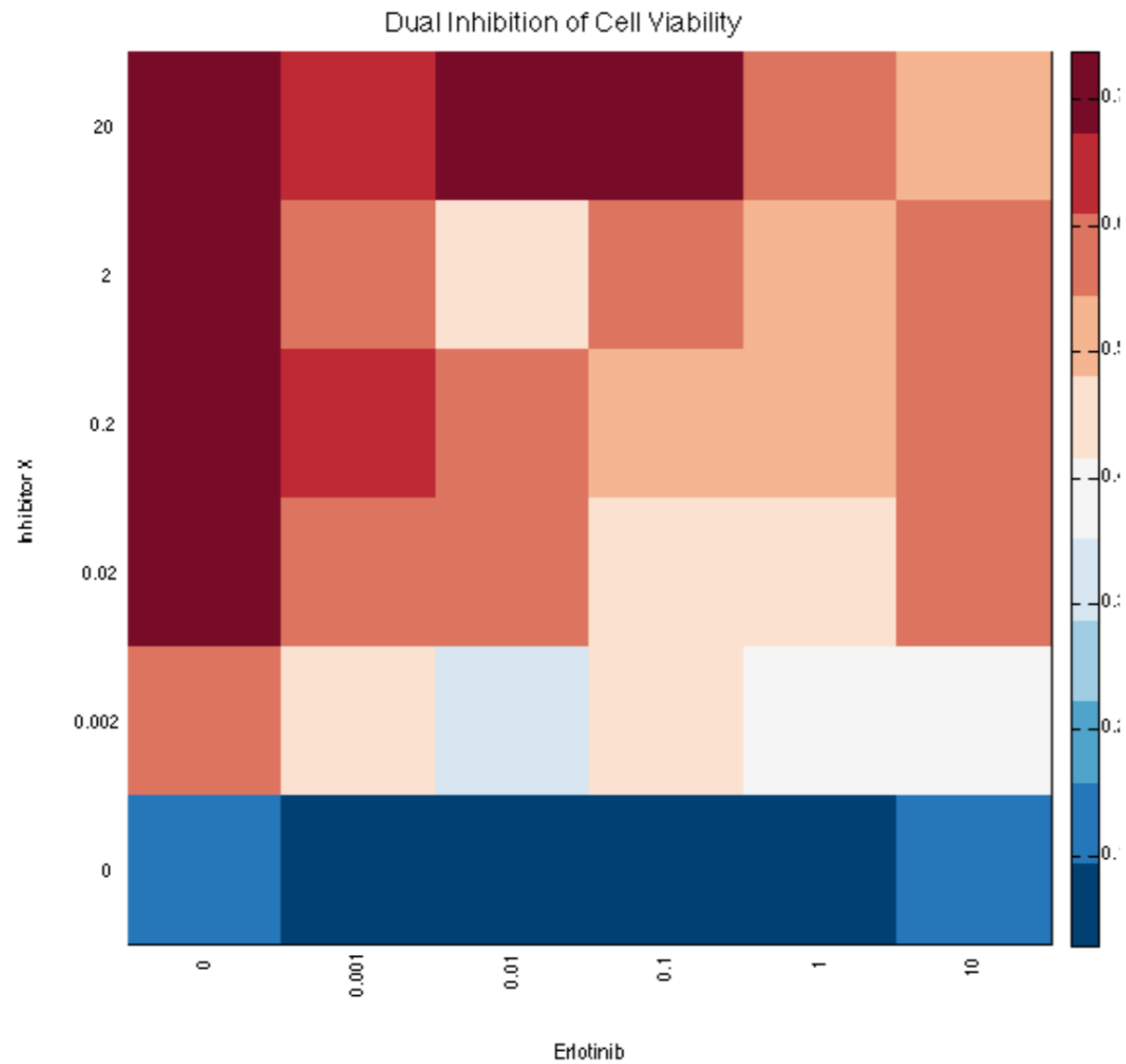
★ Does Inhibitor X effect
the IC_{50} of EVL to inh?



How can you visualize the data?



How can you visualize the data?



How can you visualize the data?

Some representation of error is required (somewhere).

What are the interesting comparisons?

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