

Supplemental Material

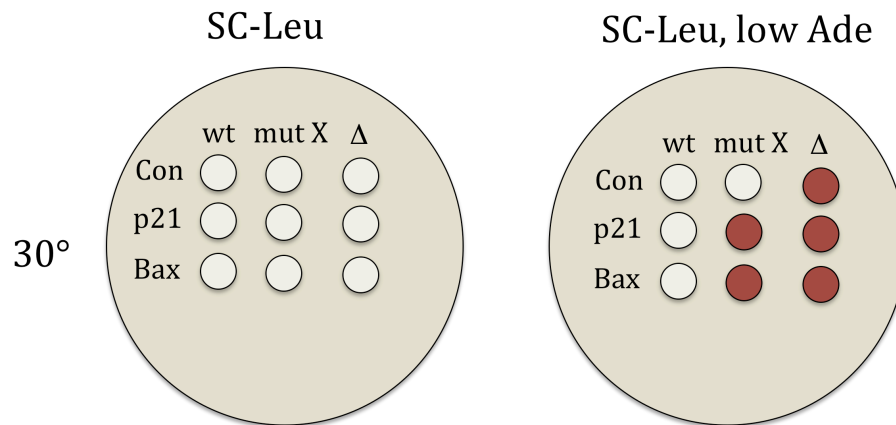
CBE—Life Sciences Education

Brownell et al.

Supplemental Material

Sample questions from Exam 3 that are specific to the course-based undergraduate research experience (CURE)

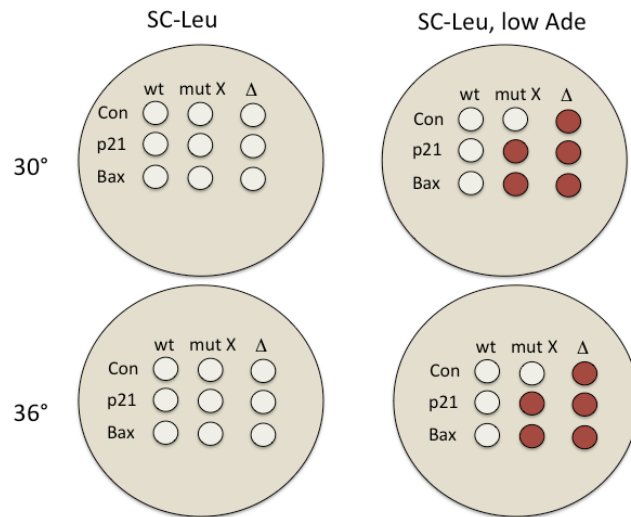
Part I. Suppose that your friend signs up for Bio44X next quarter and will be working on a new p53 mutant, p53-mutX. Because your friend knows that you took the course this quarter, she decides to ask you some questions about how to analyze her data for p53-mutX. The first experiment she conducts is a spot assay at 30 °C using yeast strains with Con, p21, and Bax p53 response elements driving the *ADE2* and *lacZ* reporter genes. She spots these yeast strains onto SC-Leu and SC-Leu, low Ade plates, and obtains the following results after 2 days at 30 °C.



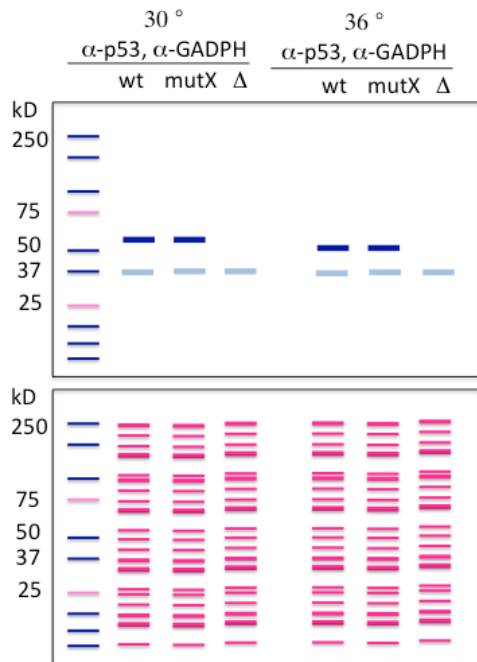
1. Based on this data, what is wrong with p53-mutX in 1-2 sentences? (5 pts)
2. Your friend suggests using a different response element to further probe the function of Mutant X using the spot assay. You recommend using the p53-RE from R2, a gene whose product is involved in **DNA repair**. Given what you know about the relative strengths of p53-REs, what would you predict for the results? Please circle one. (5 pts)
 - a) p53-mutX would show a more severe defect in this assay with the R2 p53-RE than with the Bax one.
 - b) p53-mutX would show a more severe defect in this assay with the R2 p53-RE than with the Con one.
 - c) p53-mutX would show a less severe defect in this assay with the R2 p53-RE than with the Con one.

Your friend continues working on p53-mutX and performs the same series of experiments as you conducted in Bio44X with the appropriate yeast strains expressing p53-mutX using the exact same protocols. The following are data are representative of her mutant group conclusions for each assay.

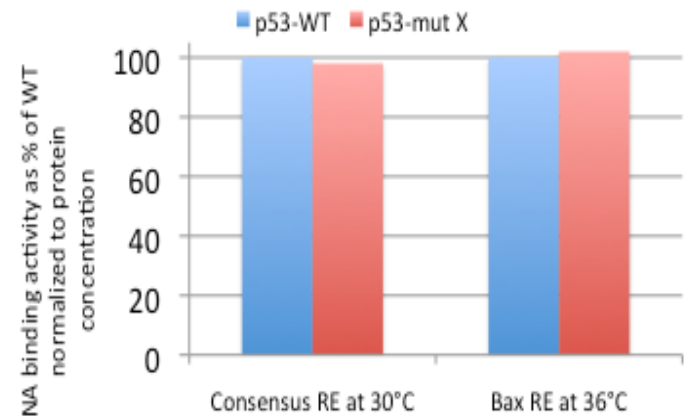
A) ADE2 spot assay



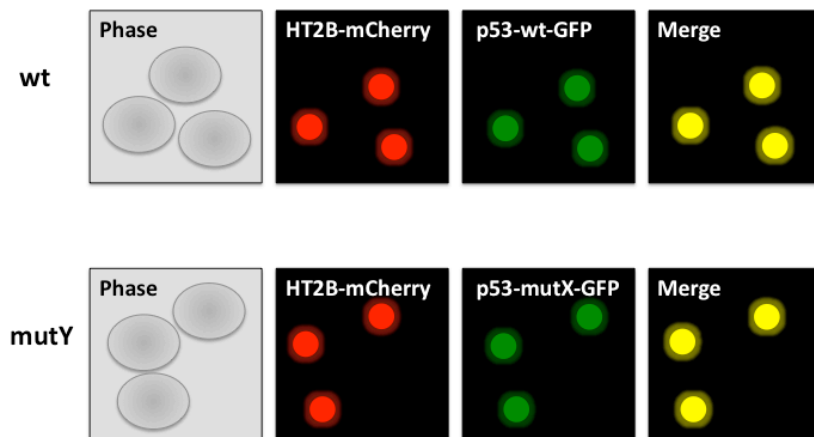
B) Western blot and Ponceau-S



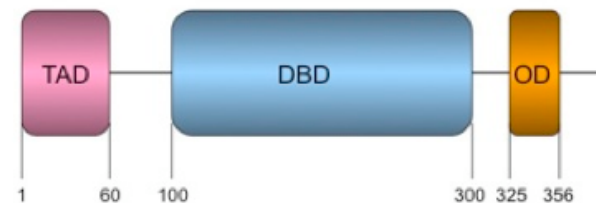
C) DNA binding assay



D) Microscopy



E) DNA sequencing reveals that Mutant X contains the change L43R.



3. Based on these data, which of the following statements are **true** (*not* speculative!) about the functional defect of p53-mutX? Please circle all that apply. (20 pts, 2 pts each)

- a) p53-mutX is being degraded more than p53-wt
- b) p53-mutX is binding to DNA as well as p53-wt
- c) p53-mutX *cannot* form tetramers as well as p53-wt
- d) p53-mutX *does not* localize to the nucleus as well as p53-wt
- e) p53-mutX *does not* recruit RNA polymerase as well as p53-wt
- f) p53-mutX has a mutation in the oligomerization domain
- g) p53-mutX *does not* have a temperature sensitive defect
- h) p53-mutX *does not* have a mutation in the DNA binding domain
- i) p53-mutX is *not* being produced at the same levels as p53-wt in the cell
- j) The p53-mutX extract has the same protein concentration as p53-wt protein extract

4. In 1-2 sentences, please state what you think is the most likely reason for the defect you see in p53-mutX. (5 pts)