

Biology 210 – Genetics – Exam 3 (Final) – Fall 2007

Name (see below): _____

Instructions:

- 1) Print your name (first name first, middle initial and last name) on the line above. Please use the form of your name that matches university records (e.g., no nicknames, or recently changed names).
- 2) Read each question carefully before you begin work on it. Many questions can be answered more easily than you might expect!
- 3) Be sure to put your final answers in the boxes or on the lines provided. You will receive full credit only for answers that are in the correct places.

Section A. Fact based questions: The answers are in the paper. (7 points each)

1. What does MDC1 bind to at sites of DNA damage? (Your answer should refer to a specific part of a particular protein)

The phosphorylated tail of H2AX, or γ H2AX

2. What site (amino acid at a specific position) in RAP80 is phosphorylated by ATM?

S101 or serine 101

3. Which of the twenty amino acids (e.g. leucine) links ubiquitin molecules into polymers? (please write out the full name of the amino acid)

lysine

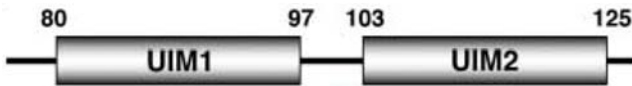
4. Which form of polyubiquitin is preferentially catalyzed by the E3 polyubiquitin ligase activity of BRCA1?

K6

5. What drug was used to enrich for cells in the M phase of the cell cycle?

nocodazole

6. The diagram below was used to represent a part of a specific protein. What protein is it?



RAP80

7. What is the protein that BRCA1 is usually associated with as a heterodimer?

BARD1

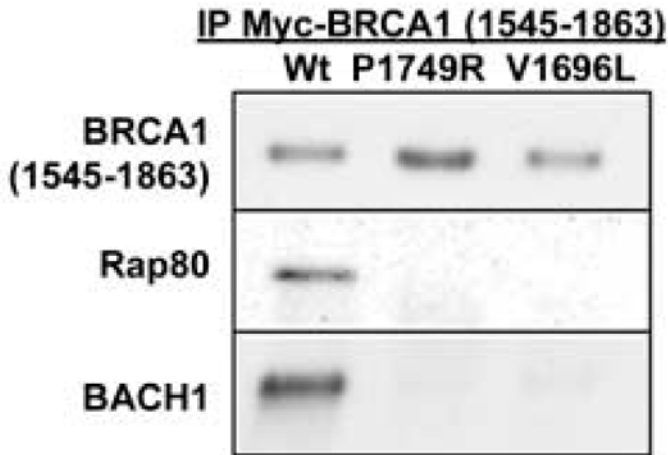
8. The JAMM family is a group of evolutionarily related (homologous) deubiquinating enzymes. Which of the proteins from this study appears to belong to this family?

BRCC36

9. What epitope (you can use its abbreviation) allowed proteins to be purified on glutathione-conjugated sepharose beads?

GST or glutathione-S-transferase

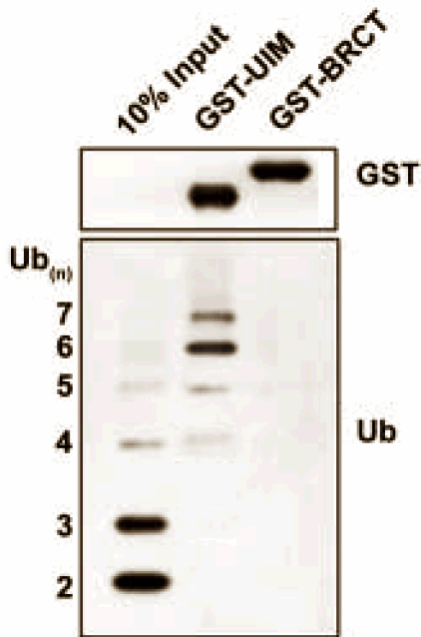
10. In the figure below, two of the columns are for BRCA1 missense mutations that have been clinically shown to cause an increased risk of breast cancer. The amino acid positions of these mutations are 1749 and amino acid 1696. What domain of the BRCA1 are these mutations located in?



BRCT

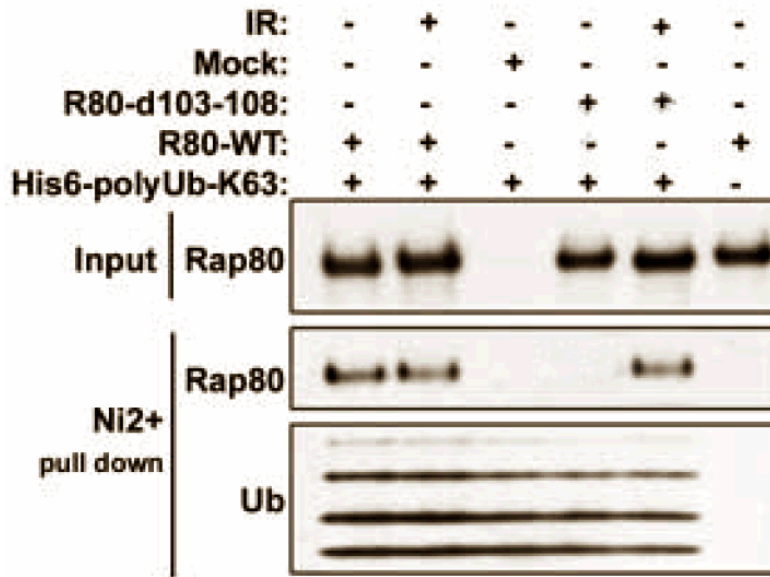
Section B. Questions that require a basic understanding of the paper. (5 points each)

1. The figure below is an altered version of Fig 2C. What would you have concluded from this figure?



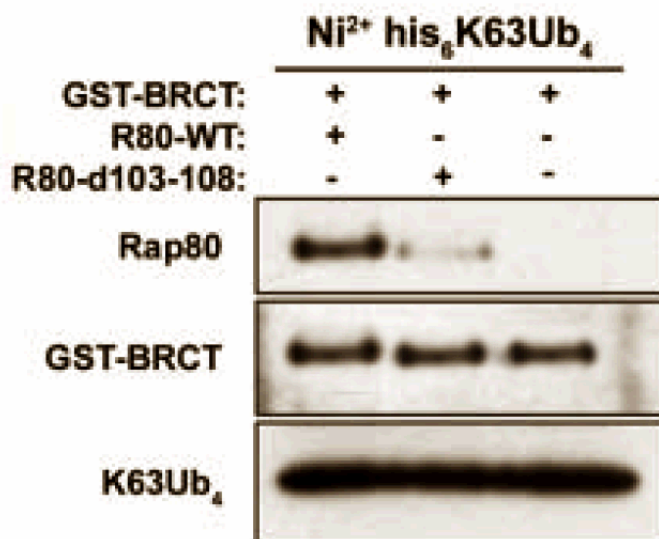
RAP80 has a preference for ubiquitin polymers with at least six ubiquitin molecules

2. The figure below is an altered version of Fig 2E. It has been altered in multiple ways. What would you have concluded from this figure?



Wild-type RAP80 binds to polyubiquitin whether it is from a cell that has been irradiated or not, but RAP80 with a portion of UIM2 deleted only binds to polyubiquitin when it is from an irradiated cell.

3. The figure below is an altered version of Fig. 3B. What would you have concluded from this figure?



The BRCT domain of BRCA1 does not require RAP80 to interact/bind with ubiquitin.

4. If you purified epitope-tagged BARD1 from human cells by immunoprecipitation, what five other proteins would you expect to find with BARD1?

BRCA1, RAP80, BACH1, BRCC36, BRCC45

**Section C. Questions that require an advanced understanding of the paper and lectures
(5 points each)**

1. BRCA1 is phosphorylated by ATM in cells exposed to ionizing radiation. Does this require RAP80? Support your answer by referring to the results shown in one of the figures in this paper.

Probably no, but the results are inconclusive. Figure 1C shows that RAP80-P is detected after IR with either WT RAP80 or S101A RAP80. However, it is possible that even if it cannot be phosphorylated itself by ATM (S101A) RAP80 is still required for BRCA1 get phosphorylated.

2. BRCA1 and RAP80 both bind to polyubiquitin, which suggests the possibility that RAP80 does not bind directly to BRCA1, but instead that both bind to polyubiquitin. However, there is evidence in the paper that suggests the UIM2 domain of RAP80 is not needed for the interaction between BRCA1 and Rap80. What is that evidence?

A mutant form of RAP80 with a deletion in UIM2 coimmunoprecipitated with BRCA1 like RAP80

Have a Great Holiday !