

2023



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# AP<sup>®</sup> Biology

## Sample Student Responses and Scoring Commentary

### **Inside:**

#### **Free-Response Question 1**

- Scoring Guidelines**
- Student Samples**
- Scoring Commentary**

## Question 1: Interpreting and Evaluating Experimental Results with Experimental Design

9 points

In eukaryotic microorganisms, the PHO signaling pathway regulates the expression of certain genes. These genes, *Pho* target genes, encode proteins involved in regulating phosphate homeostasis. When the level of extracellular inorganic phosphate (Pi) is high, a transcriptional activator Pho4 is phosphorylated by a complex of two proteins, Pho80–Pho85. As a result, the *Pho* target genes are not expressed. When the level of extracellular Pi is low, the activity of the Pho80–Pho85 complex is inhibited by another protein, Pho81, enabling Pho4 to induce the expression of these target genes. A simplified model of this pathway is shown in Figure 1.

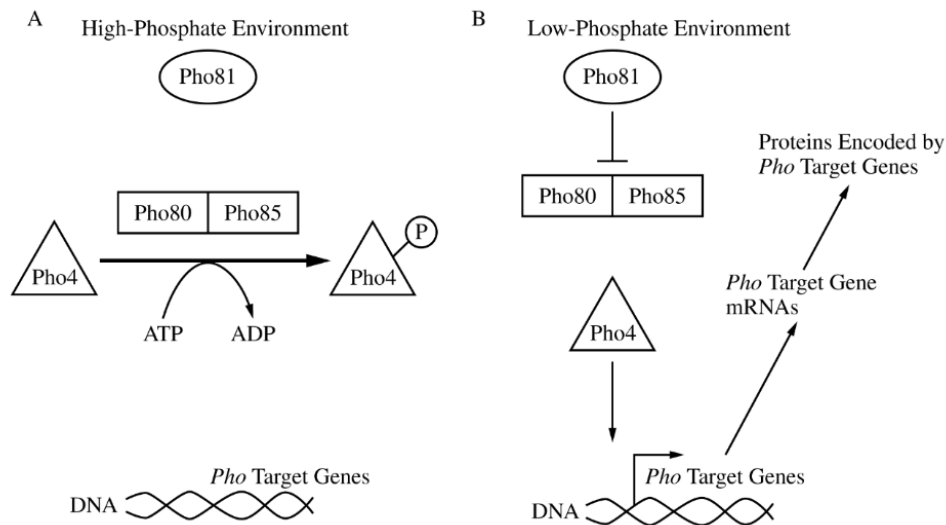


Figure 1. A simplified model of the regulation of expression of *Pho* target genes in (A) a high-phosphate (high-Pi) environment and (B) a low-phosphate (low-Pi) environment

To study the role of the different proteins in the PHO pathway, researchers used a wild-type strain of yeast to create a strain with a mutant form of Pho81 (*pho81mt*) and a strain with a mutated form of Pho4 (*pho4mt*). In each of these mutant strains, researchers measured the activity of a particular enzyme, APase, which removes phosphates from its substrates and is encoded by *PHO1*, a *Pho* target gene (Table 1). They then determined the level of *PHO1* mRNA relative to that of the wild-type yeast strain, which was set to 10.

TABLE 1. APase ACTIVITY AND RELATIVE AMOUNTS OF *PHO1* mRNA IN WILD-TYPE AND MUTANT STRAINS OF YEAST IN HIGH- AND LOW-PHOSPHATE ENVIRONMENTS

Yeast Strain	Mutation	APase Activity in High- Pi Environment (mU/mL/OD <sub>600</sub> ) ±2SE <sub><math>\bar{x}</math></sub>	APase Activity in Low- Pi Environment (mU/mL/OD <sub>600</sub> ) ±2SE <sub><math>\bar{x}</math></sub>	Relative Amounts of <i>PHO1</i> mRNA in High- Pi Environment ±2SE <sub><math>\bar{x}</math></sub>	Relative Amounts of <i>PHO1</i> mRNA in Low- Pi Environment ±2SE <sub><math>\bar{x}</math></sub>
Wild-type	None	0.5 ± 0.1	17.3 ± 0.9	0.1 ± 0.0	10 ± 2.0
<i>pho81mt</i>	Nonfunctional Pho81	0.4 ± 0.1	0.6 ± 0.1	0.7 ± 0.2	0.9 ± 0.8
<i>pho4mt</i>	Nonfunctional Pho4	0.5 ± 0.0	0.8 ± 0.2	0.6 ± 0.4	0.3 ± 0.1

**(a)** Describe the effect the addition of a charged phosphate group can have on a protein that would cause the protein to become inactive. **1 point**

- It changes the structure/shape of the protein.

**Explain** how a signal can be amplified during signal transduction in a pathway such as the PHO signaling pathway. **1 point**

- Each enzyme (in a signal transduction pathway) can act on many copies of a protein.

**Total for part (a) 2 points**

**(b)** Based on Table 1, **identify** a dependent variable in the researchers' experiment. **1 point**  
Accept one of the following:

- APase activity
- (Relative) amount of *PHO1* (mRNA)

**Justify** the researchers' using the wild-type strain for the creation of the mutant strains. **1 point**  
Accept one of the following:

- It ensures that any observed differences (in experimental results) between the strains are due to the introduced mutations (and not to other genetic differences between the yeast strains).
- It ensures that the strains are genetically identical except for the introduced mutations.

**Justify** the researchers' using mutant strains in which only a single component of the pathway was mutated in each strain. **1 point**

Accept one of the following:

- It allows them to test the effect of each mutation separately.
- It allows them to (better) determine which component is responsible for any observed differences.

**Total for part (b) 3 points**

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<b>(c)</b>	Based on the data in <u>Table 1</u> , <b>identify</b> the yeast strain and growth conditions that lead to the highest relative amount of <i>PHO1</i> mRNA. <ul style="list-style-type: none"><li>• Wild-type yeast in a low-Pi environment</li></ul>	<b>1 point</b>
	<b>Calculate</b> the percent change in APase activity in wild-type yeast cells in a high-Pi environment compared with that of wild-type cells in a low-Pi environment. Accept one of the following: <ul style="list-style-type: none"><li>• 3,360% <math>[(17.3-0.5)/0.5 \times 100\%]</math></li><li>• -97% <math>[(0.5-17.3)/17.3 \times 100\%]</math></li></ul>	<b>1 point</b>
<b>Total for part (c)</b>		<b>2 points</b>
<b>(d)</b>	In a follow-up experiment, researchers created a strain of yeast with a mutation that resulted in a nonfunctional Pho85 protein. Based on <u>Figure 1</u> , <b>predict</b> the effects of this mutation on <i>PHO1</i> expression in the mutant strain in a high-Pi environment. <ul style="list-style-type: none"><li>• <u>It/PHO1/Target genes</u> will be expressed.</li></ul> Provide reasoning to <b>justify</b> your prediction.	<b>1 point</b>
	<ul style="list-style-type: none"><li>• (In a high-Pi environment) a nonfunctional Pho85 will be unable to <u>phosphorylate/inhibit</u> Pho4.</li></ul>	<b>1 point</b>
<b>Total for part (d)</b>		<b>2 points</b>
<b>Total for question 1</b>		<b>9 points</b>

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## BEGIN Question 1

Begin your response to QUESTION 1 on this page. Do not skip lines.

- a. The addition of a charged phosphate group likely changes the tertiary folding structure of a protein, and since structure determines function, a change in its folding due to the phosphate group could cause the protein to become inactive. A signal is amplified when there is a chain reaction, likely a phosphorylation cascade. One reaction leads to another, often increasing the size and impacts of the reactions as they continue or pass along.
- b. one dependent variable is the activity of APase. In using a wild-type strain to create the mutant strain, the researchers can be sure that the only mutation is the one they induced, ~~and~~ and all other genes are consistent with the wild type. Further, by ~~using~~ using mutant strains that had only a single component mutated, they could know that any significant ~~and~~ differences in enzyme activity were for sure caused by

## Additional page for answering Question 1

Continue your response to **QUESTION 1** on this page. Do not skip lines.

that one mutation since everything else was the same.

c. The wild-type yeast in the Low-Pi environment had the highest relative amount of PHO1 mRNA.

$$\text{Percent change: } \frac{17.3 - .5}{.5} \times 100 = 3360\%$$

d. A mutation resulting in nonfunctional Pho85 would allow transcription of the Pho target gene despite a High Pi environment. This is because Pho85 would not be able to form the Pho80-Pho85 complex and phosphorylate Pho4. Therefore, Pho4 will transcribe the Pho target genes even when it should not, leading to increased PHO1 expression.

## BEGIN Question 1

Begin your response to QUESTION 1 on this page. Do not skip lines.

- a) A charged phosphate group can inhibit the protein's receptor. A signal can be amplified during signal transduction <sup>pathway</sup> through the electron transport chain.
- b) A dependent variable is the APase Activity of the strains of yeast. It was important for the researchers to use the wild-type strain to create the mutant strain to ensure no other mutations were present in the strain. It was important for the researchers to only mutate a single component of the pathway so that they can accurately determine whether or not it is causing differences in APase activity/amount of PHO1 mRNA. If multiple components were mutated, the researchers would not know which component is causing the difference.
- c) wild-type, low-pi environment.

$$\frac{1.5 - 17.3}{17.3} \times 100 = \boxed{97.11\%}$$

- d) This would decrease PHO1 expression b/c it will stop the conversion of ~~ADP~~ ATP  $\rightarrow$  ADP, which is necessary for PHO1 formation.

## BEGIN Question 1

Begin your response to QUESTION 1 on this page. Do not skip lines.

- a) the addition of a charged phosphate group would cause the protein to become inactive due to the disruption of homeostasis, when the level of phosphate is high, more protein is being used to phosphorylate  $\text{Pto4}$ . A signal can be amplified during signal transduction by balancing phosphate amounts.
- b) A dependent variable in the experiment would be the APase activity in both high-pi and low-pi environments. The researchers used the wild-type strain in order to have a control group to compare the data to. The researchers mutated only one component in each strain to see a difference when certain independent variables are added.
- c) The highest relative amount of  $\text{Pto1}$  mRNA resulted from the wild type yeast strain in a low-pi environment. The percent change in APase activity in wild type yeast cells in a high-pi environment compared to a low-pi environment is 31.6%.
- d) After adding the strain of yeast with a mutation that results in nonfunctional phosph protein, the relative amounts of  $\text{Pto1}$  would decrease due to the lack of signal amplification.



## Question 1

**Note:** Student samples are quoted verbatim and may contain spelling and grammatical errors.

### Overview

Question 1 described the PHO signaling pathway, which regulates phosphate homeostasis in yeast. The question stimulus presented a simplified model of the signal transduction pathway and a data table from an experiment designed to study the roles of Pho81 and Pho4, two proteins in the PHO pathway.

In part (a) students were expected to describe the effect of adding a charged phosphate group to a protein (Skill 1.A; Learning Objective [LO] SYI-1.C from the AP Biology Course and Exam Description [CED]). Students were also expected to explain how signals can be amplified in a signal transduction pathway (Skill 1.C; LO IST-3.D).

In part (b) students were expected to demonstrate understanding of experimental design by identifying a dependent variable, justifying the researchers' using a wild-type strain of yeast as the background for creating mutant strains, and justifying the use of mutant strains that each contained a mutation to a single component of the PHO pathway (Skill 3.C).

In part (c) students were expected to describe data from the table by identifying the yeast strain and growth conditions that led to the highest relative amount of *PHO1* mRNA (Skill 4.B). Students were then asked to calculate the percent change in APase activity in wild-type cells exposed to a high extracellular inorganic phosphate (high-Pi) environment compared with those exposed to a low-Pi environment (Skill 5.A).

In part (d) students were expected to predict the results of a follow-up experiment that tests the effects of a loss-of-function mutation to Pho85, another protein in the PHO pathway (Skill 3.B; LO IST-3.G). Students were then expected to justify their predictions (Skill 6.C).

### Sample: 1A

#### Score: 8

The response earned 1 point in part (a) for describing that “The addition of a charged phosphate group likely changes the tertiary folding structure of a protein.” The response did not earn a point in part (a) because it does not explain that each enzyme can act on many copies of a protein. The response earned 1 point in part (b) for identifying a dependent variable as APase activity. The response earned 1 point in part (b) for justification by stating that the only genetic difference between the two strains is the induced mutation. The response earned 1 point in part (b) for justifying that “by using mutant strains that had only a single component mutated, they could know that any significant differences in enzyme activity were for sure caused by that one mutation.” The response earned 1 point in part (c) for identifying that the wild-type yeast strain in a low-Pi environment leads to the highest relative amount of *PHO1* mRNA. The response earned 1 point in part (c) for calculating the percent change as 3,360%. The response earned 1 point in part (d) for predicting a nonfunctional Pho85 would allow for the transcription of “the Pho target gene.” The response earned 1 point in part (d) for justifying that the mutant Pho85 would not phosphorylate Pho4, thus allowing the latter to promote transcription of *PHO1*.

### Sample: 1B

#### Score: 5

The response did not earn a point in part (a) because it does not correctly describe how the addition of a phosphate group affects protein structure. The response did not earn a point in part (a) because it does not correctly explain how a signal can be amplified during signal transduction. The response earned 1 point in part (b) for identifying APase activity. The response earned 1 point in part (b) for justifying the use of the wild-type strain to create mutant strains to “ensure no other mutations were present in the strain.” The response earned 1 point in part (b) for justifying the

**Question 1 (continued)**

use of mutant strains in which only a single component of the pathway was mutated “so that they can accurately determine whether or not it is causing differences in APase activity ... If multiple components were mutated, the researchers would not know which component is causing the difference.” The response earned 1 point in part (c) for identifying “wild-type, low-pi environment.” The response earned 1 point in part (c) for correctly calculating the percent change as 97.11%. The response did not earn a point in part (d) because it does not correctly predict the effects of a mutation of Pho85 on PHO1 expression in a high-Pi environment. The response did not earn a point in part (d) because it does not justify that Pho4 will not be inhibited.

**Sample: 1C****Score: 2**

The response did not earn a point in part (a) because it does not correctly describe how the addition of a phosphate group affects protein structure. The response did not earn a point in part (a) because it does not correctly explain how a signal can be amplified during signal transduction. The response earned 1 point in part (b) for identifying a dependent variable as APase activity. The response did not earn a point in part (b) because it does not correctly justify the use of the wild-type strain to create mutant strains. The response did not earn a point in part (b) because it does not correctly justify the use of mutant strains in which only a single component of the pathway was mutated. The response earned 1 point in part (c) for identifying that the wild-type yeast strain in a low-Pi environment leads to the highest relative amount of PHO1 mRNA. The response did not earn a point in part (c) because it incorrectly calculates the percent change in APase activity in wild-type yeast cells in a high-Pi environment compared with wild-type yeast cells in a low-Pi environment. The response did not earn a point in part (d) because it does not correctly predict the effects of a mutation of Pho85 on PHO1 expression in a high-Pi environment. The response did not earn a point in part (d) because it does not justify that nonfunctional Pho85 will be unable to inhibit Pho4.