

Test for **ELISA: Amplification of Biochemical Signals** Activity

Name: _____

Teacher: _____

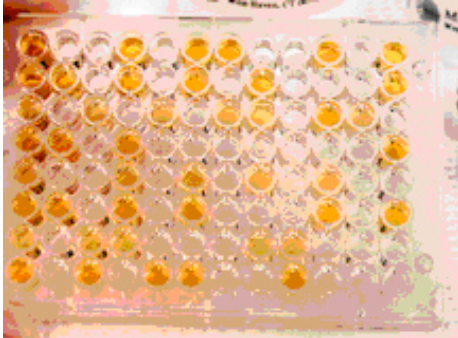
Date _____

Class _____

Check one:

Pretest

Posttest



1. The picture to your left shows the color of the solutions in different wells of an ELISA test.

What in ELISA gets colored and what does the presence of color tell you?

Explain in 2-3 sentences:

The presence of color indicates that in this test sample there are indeed specific substances for which the ELISA technique is testing. The color appears because the enzymes linked to secondary antibodies have changed the colorless substance into a colored product. The more color detected, the higher the level of specific antibodies in the sample.

Explanation Score

	Response includes...
Complete (3)	Includes a description of what gets colored, how color appears and what the presence of color indicates about level of specific antibody in the sample.
Mostly complete (2)	Includes a description of what gets colored, how color appears and what the presence of color indicates about level of specific antibody in the sample but doesn't include how level of color is related to amount of antibodies in sample.
Partial (1)	Includes either what gets colored how what the presence of color indicates but not both.
Incorrect (0)	Other

2. Which of the following is most correct:

A. ELISA measures the level of antigens in blood serum

B. ELISA measures the level of antibodies in blood serum

C. ELISA measures the level of specific antigen - antibody binding

Answers C	1
Other	0

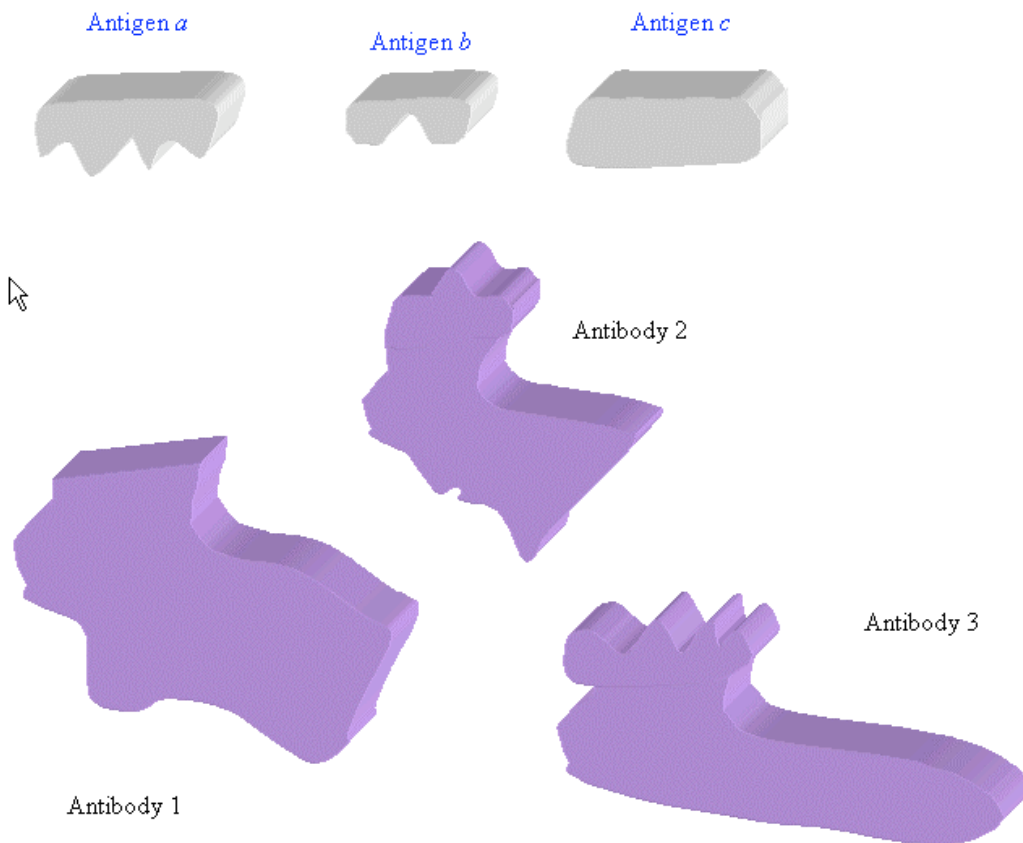
3: There are three antigens, *a*, *b* and *c* and three antibodies, 1, 2 and 3. For each antibody predict which antigen makes the strongest complex and which one makes the weakest. complex.

2a. Antibody 1: antigen c binds most strongly, antigen a binds weakly

2b. Antibody 2: antigen b binds most strongly, antigen c binds weakly

2c. Antibody 3: antigen a binds most strongly, antigen c binds weakly

Response includes...	
One point for each correct answer.	1
0 where it is incorrect.	0



4. Consider the different steps of the ELISA procedure. After adding antibodies, the protocol requires several thorough washings of the plates and careful removal of the washing fluid to assure that weakly bound antibodies are washed away. Why do you think there are strongly bound and weakly bound antibodies in the samples?

Response includes...	
Any two proteins have some kind of attraction to each other but only specific antigens and their complementary antibodies are strongly attracted to each other and therefore won't be washed away	2
Response indicates that you need to have complementary antibodies for strong attractions but doesn't address weak attractions.	1
Other	0

4b. What will happen with the test results if those weakly bound antibodies were not removed completely?

Answers the result will be a false positive	1
Other	0

5. When conducting the ELISA tests, a lab technician placed the plates with antigens and antibodies into a cold room (under 4°C (39°F)) for an hour as required by the protocol and went to the cafeteria. When she came back, she discovered that the air conditioning unit stopped working and the temperature in the previously cold room was about 27°C (80°F). Explain why she should expect a problem with the ELISA test as the result of the increased temperature during the incubation period in the cold room?

Too much heat energy may make the molecules move too much, causing them to overcome the attractive forces between antigens and their specific antibodies. In addition, temperature - induced denaturation may modify those unique shapes on an antigen that are responsible for antigen -antibody recognition, thus decreasing the specificity of the test. Finally, higher temperatures may convert some of the colorless substance into a colored product without the enzyme because the chemical reaction would be more likely at higher temperatures. All these can make the test-either false negative or false positive.

Explanation Score

	Response includes...
Complete (3)	Includes rise in temperature is responsible for 1) increase in molecular motion which may overcome attractions, 2) potential denaturation, or 3) might convert colorless substance to one with color causing a false result
Mostly complete	Includes 2 of the 3 reasons above.

(2)	
Partial (1)	Includes one possible problem from the list above.
Incorrect (0)	Other