PATHWAY: Biotechnology Research & Development

COURSE: Introduction to Biotechnology

UNIT 8: Biotechnology Protein Lab

INTRODUCTION

Annotation:

In this unit students will apply skills learned in lab procedures unit to make solutions of differing molarity, concentrations, assaying for enzyme activity, and using antibody staining techniques to identify a protein. The unit concludes with a culminating task -ELISA lab

Grade(s):

X	9th
X	10th
X	11th
X	12th

Time:

9 hours

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Students with Disabilities:

For students with disabilities, the instructor should refer to the student's IEP to be sure that the accommodations specified are being provided. Instructors should also familiarize themselves with the provisions of Behavior Intervention Plans that may be part of a student's IEP. Frequent consultation with a student's special education instructor will be beneficial in providing appropriate differentiation.

FOCUS STANDARDS

GPS Focus Standards:

HS-IBT-4 **Students will demonstrate how concepts of physical science connect to biochemical applications and techniques.**

a) Calculate and prepare buffers, stock solutions, and reagents.
b) Analyze and apply the concepts of homeostasis and molar relationships to biochemical reactions.
d) Analyze enzyme activity using assays for reactants and products.
f) Use antibody specificity for antigens to test for the presence of protein (*e.g.,* ELISA, Western Blot, antibody staining).

GPS Academic Standards:

SB1 Students will analyze the nature of the relationships between structures and functions in living cells.

b) Explain how enzymes function as catalysts.
c) Identify the function of the four major macromolecules (i.e., carbohydrates, proteins, lipids, nucleic acids).
d) Explain the impact of water on life processes (i.e., osmosis, diffusion).

SPS6 Students will investigate the properties of solutions.

a) Describe solutions in terms of, solute/solvent, conductivity, concentration.
b) Observe factors affecting the rate a solute dissolves in a specific solvent.
c) Demonstrate that solubility is related to temperature by constructing a solubility curve.

SC4 Students will characterize the properties that describe solutions and the nature of acids and bases.

a) Explain the process of dissolving in terms of solute/solvent interactions:
- Observe factors that effect the rate at which a solute dissolves in a specific solvent,
- Express concentrations as molarities,
- Prepare and properly label solutions of specified molar concentration

MM2P1 Students will solve problems (using appropriate technology).

b) Solve problems that arise in mathematics and in other contexts.
c) Apply and adapt a variety of appropriate strategies to solve problems.

UNDERSTANDINGS & GOALS

Enduring Understandings:

- Enzymes act as catalysts to help complex reactions occur.

Essential Questions:

- What is a buffer?
- How do you calculate a specified dilution from a concentrated stock solution?
- What is the role and function of enzymes in a living organism?

- How do you detect an enzyme's activity?
- How do antigens and antibodies interact?
- What are the types of staining techniques?

Knowledge from this Unit:

Students will be able to discuss:

- The importance of buffers.
- Enzymes, their activity, and their importance.
- Antibody staining and the applications of the technique.

Skills from this Unit:

Students will be able to:

- Make buffers
- Prepare solutions of varying concentrations of solute and solvent
- Make calculations of solution concentrations.
- Prepare dilutions of concentrated solutions
- Perform antibody staining techniques.
- Detect enzyme activity.

ASSESSMENTS

Assessment Method Type:

_____ Pre-test
X Objective assessment - multiple-choice, true- false, etc.
__ Quizzes/Tests
__ Unit test
X Group project
_____ Individual project
x Self-assessment - May include practice quizzes, games, simulations, checklists, etc.
__ Self-check rubrics
__ Self-check during writing/planning process
__ Journal reflections on concepts, personal experiences and impact on one's life
__ Reflect on evaluations of work from teachers, business partners, and competition judges
__ Academic prompts
__ Practice quizzes/tests
_____ Subjective assessment/Informal observations
__ Essay tests
__ Observe students working with partners
__ Observe students role playing
_____ Peer-assessment
__ Peer editing & commentary of products/projects/presentations using rubrics
__ Peer editing and/or critiquing
_____ Dialogue and Discussion
__ Student/teacher conferences
__ Partner and small group discussions
__ Whole group discussions
__ Interaction with/feedback from community members/speakers and business partners
X Constructed Responses
__ Chart good reading/writing/listening/speaking habits
__ Application of skills to real-life situations/scenarios
_____ Post-test

Assessment Attachments and / or Directions:

LESSON PLANS

Instructional planning: Before beginning the ELISA Lab, use the **ELISA General Tasks Teacher Handout** to be sure all supplies are available.

• LESSON 1: Introduction to Buffers

1. Identify the standards. Standards should be posted in the classroom.

   HS-IBT-4 **Students will demonstrate how concepts of physical science connect to biochemical applications and techniques.**
   a) Calculate and prepare buffers, stock solutions, and reagents.
   b) Analyze and apply the concepts of homeostasis and molar relationships to biochemical reactions.
   d) Analyze enzyme activity using assays for reactants and products.
   f) Use antibody specificity for antigens to test for the presence of protein (*e.g.,* ELISA, Western Blot, antibody staining).

2. Review Essential Question(s). Post Essential Questions in the classroom.

   - What is a buffer?
   - How do you calculate a specified dilution from a concentrated stock solution?
   - What is the role and function of enzymes in a living organism?
   - How do you detect an enzyme's activity?
   - How do antigens and antibodies interact?
   - What are the types of staining techniques?

3. Identify and review the unit vocabulary. Terms may be posted on word wall.

Solute	Solvent	Molarity
Enzymes	Catalysts	Substrates
Active Sites	Cofactors	Induced Fit Model
Lock and Key Model	Primary Antibody	Secondary Antibody
Monoclonal Antibody	Buffer	Homeostasis
Biochemical reactions	Biochemical reactions	

4. Interest approach – Mental set
   1. Ask students to stand on up and extend their arm out to the side, then have them try to stand on one foot. Ask them to describe how that felt. Then ask them what they think their body does inside when it is out of balance. Ask students to define homeostasis.
   2. Discuss Homeostasis using the information from a site such as http://coldwater.k12.mi.us/nicholsk/courses/chs/Phys/homeo.htm
   3. Discuss how we have buffers in our body to help maintain homeostasis.
   4. A "buffer" is something that resists change.
   5. A buffer helps maintain a near constant pH upon the addition of small amounts of H^+ and OH^- ions to a solution.

6. To prepare a buffer calculations must be performed first to determine the amount of each item to be used in making the buffer.
7. Review of how to make buffers and calculate molarity.
8. Have students calculate molarity .
9. Remind students they will be making buffers later in a few lessons.
10. Give homework to calculate buffers.

• LESSON 2: Buffers and Enzymes

1. Review Essential Questions. Post Essential Questions in the classroom.
   - What is a buffer?
   - How do you calculate a specified dilution from a concentrated stock solution?
   - What is the role and function of enzymes in a living organism?
   - How do you detect an enzyme's activity?
   - How do antigens and antibodies interact?
   - What are the types of staining techniques?

2. Ask students what they think miscalculation of a buffer and molarity can make the difference in an experiment. Ask them to recall how to calculate buffers and molarity-ask for a student volunteer to calculate on board and discuss results.
   3. State "Now we will look at another aspect of proteins". Ask students what are enzymes? Ask where can they be found? Discuss their responses and reemphasize the importance of enzymes.
3. Enzymes are very important and can be found everywhere. Enzymes are the biological substance (proteins) that act as **catalysts** and help complex reactions occur everywhere in life. They act like locks and keys and perform specific tasks. There are enzymes for various types of tasks-some are metabolic and some are digestive
4. Enzymes follow a four step process in working first the enzyme and substrate (the molecule an enzyme acts on) are located in the same area; second, the enzyme attaches to the active site of the substrate (which is a special site shaped like the enzyme); third the substrate changes through a process called catalysis and fourth, the enzyme detaches itself from the substrate and is ready to work again. The substrate is a different product when this happens
5. Activity: Ask students to name some enzymes. Ask if the know the name of the substrate that it acts on, for example Lipase, Amylase, maltase, lactase, If they have trouble recalling names give handout with a list of some digestive enzymes.. Ask the students to identify where in the body they are produced, what reactions do they catalyze, and analyze the consequences of the absence of those enzymes in the body-for example examine the Krebs cycle example found at http://incolor.inebraska.com/mcanaday/Krebs%20Phases.htm

   - **Amylase** from our salivary glands and pancreas digests starch to maltose in our mouth and small intestine.
   - **Lipase** from the pancreas digests lipids to fatty acids and glycerol in our small intestine.
   - **Pepsin** is a protease that begins digestion of proteins, breaking them into peptides and amino acids. Pepsinogen, is secreted by gastric glands of the stomach into the stomach. There, in the acid environment of the stomach, pepsinogen is converted into pepsin.
   - **Trypsin** is a protease secreted into the small intestine by the pancreas. As pepsin, trypsin digests proteins into peptides and amino acids and is made and secreted in an inactive form, trypsinogen. Although both pepsin and trypsin are proteases, they require quite different conditions of acidity and alkalinity for their action. The fifth enzyme, catalase, is found in cells of most tissues.

6. Have students perform the **Catalase Experiment** on the Enzyme handout and discuss results.

7. Ask students to identify the practical applications of antibody staining technique in medical, environmental, agriculture fields.

• LESSON 3: Virtual Enzyme Lab-Catalase

1. Review Essential Questions. Post Essential Questions in the classroom.
   - What is a buffer?
   - How do you calculate a specified dilution from a concentrated stock solution?
   - What is the role and function of enzymes in a living organism?
   - How do you detect an enzyme's activity?
   - How do antigens and antibodies interact?
   - What are the types of staining techniques?

2. Recap the function of enzymes and how they act of specific substrates that have openings that match the shape of the enzyme. The enzyme performs its work on the substrate then detaches itself from the substrate which has been altered in some way.
3. In the computer lab have each students go to this website http://www.phschool.com/science/biology_place/labbench/lab2/intro.html
4. Complete the virtual Enzyme Catalysis Lab and take the quiz at the end.

• LESSON 4: Enzymes, Catalase Lab.

1. Review Essential Questions. Post Essential Questions in the classroom.
   - What is a buffer?
   - How do you calculate a specified dilution from a concentrated stock solution?
   - What is the role and function of enzymes in a living organism?
   - How do you detect an enzyme's activity?
   - How do antigens and antibodies interact?
   - What are the types of staining techniques?

2. Discuss the outcome of the virtual enzyme lab and review the procedure performed.
3. Have students perform the Catalase Lab or Amylase Lab or other enzyme experiment

LESSON 5: Antigen and Antibodies

1. Review Essential Questions. Post Essential Questions in the classroom.
   - What is a buffer?
   - How do you calculate a specified dilution from a concentrated stock solution?
   - What is the role and function of enzymes in a living organism?
   - How do you detect an enzyme's activity?
   - How do antigens and antibodies interact?
   - What are the types of staining techniques?
2. Ask students what the think happens when someone is exposed to tuberculosis or some other common infectious disease.
3. Ask students what antigens and antibodies are.
4. Discuss how they react.
5. Discuss examples of antigen and antibody reactions.
6. Discuss how antibody staining is used to detect antibodies.

7. Have students complete the virtual immunology lab found at: http://www.hhmi.org/biointeractive/vlabs/immunology/index.html

LESSON 6: Buffer Solution Lab, Applications of ELISA.

1. Review Essential Questions. Post Essential Questions in the classroom.
   - What is a buffer?
   - How do you calculate a specified dilution from a concentrated stock solution?
   - What is the role and function of enzymes in a living organism?
   - How do you detect an enzyme's activity?
   - How do antigens and antibodies interact?
   - What are the types of staining techniques?

2. Discuss results of the virtual lab and recap the important points of staining and lab procedures in preparing buffers. Ask students to identify the practical applications of antibody staining technique in medical, environmental, agriculture fields. Talk about also first test to identify HIV, Pregnancy test and other uses.
3. Have students visit http://www.carolina.com/category/teacher+resources/instructions+and+buying+guides/chemistry+instruction+manuals/solution+preparation+manual.do to review the preparation of solutions.
4. Introduce the ELISA Lab using the **ELISA Scenario Handout**.
5. Have students make buffers and solutions for the ELISA Lab using pages 3-5 of the **ELISA Lab Handout.** To make sure you have all the necessary items, use the **ELISA General Tasks Teacher Handout**.

LESSON 7: Lab: Perform ELISA.

1. Review Essential Questions. Post Essential Questions in the classroom.
   - What is a buffer?
   - How do you calculate a specified dilution from a concentrated stock solution?
   - What is the role and function of enzymes in a living organism?
   - How do you detect an enzyme's activity?
   - How do antigens and antibodies interact?
   - What are the types of staining techniques?

2. Show students the **EPI DEMIC PowerPoint**
3. Students will follow the steps in the ELISA Lab and document results using the **ELISA Lab Handout** (starting from page 8).

LESSON 8: ELISA Results: What do they mean?

1. Review Essential Questions. Post Essential Questions in the classroom.
   - What is a buffer?
   - How do you calculate a specified dilution from a concentrated stock solution?
   - What is the role and function of enzymes in a living organism?
   - How do you detect an enzyme's activity?
   - How do antigens and antibodies interact?
   - What are the types of staining techniques?

2. Recap **ELISA Lab Handout**, discuss results, and discuss how lab error and other issues can case a false positive or negative results.
3. Ask students what they think will happen if buffers are not used properly.
4. Discuss the potential problems with the procedure results such as HIV-false positive and how to minimize procedure errors.

LESSON 9: Assessment

1. Review Essential Questions. Post Essential Questions in the classroom.
   - What is a buffer?
   - How do you calculate a specified dilution from a concentrated stock solution?
   - What is the role and function of enzymes in a living organism?
   - How do you detect an enzyme's activity?
   - How do antigens and antibodies interact?
   - What are the types of staining techniques?

2. Students will explain the steps in an ELISA and connect enzymes to substrates.
3. The **ELISA Class Activity** can also be used as a review activity

• ATTACHMENTS FOR LESSON PLANS

- **Catalase Experiment**
- **ELISA Scenario Handout**
- **ELISA Lab Handout**
- **ELISA General Tasks Teacher Handout**
- **EPI DEMIC PowerPoint**
- **ELISA Class Activity**

• NOTES & REFLECTION:

CULMINATING PERFORMANCE TASK

Culminating Unit Performance Task Title:

Disease Detection and Prevention

Culminating Unit Performance Task Description/Directions/Differentiated Instruction:

This task contains a multimedia presentation with the step by step procedure for the ELISA, rubrics and post lab questions

Attachments for Culminating Performance Task:

- These documents are found at https://www.georgiastandards.org/Frameworks/Pages/BrowseFrameworks/Science9-12.aspx and are also attached

- Disease Detection and Prevention (ELISA Lab Handout)
- ELISA Scenario Handout
- EPI DEMIC (ppt)

UNIT RESOURCES

Web Resources:

- This is a link to good information on Making Solutions http://www.scout.wisc.edu/biolink/SPT--AdvancedSearch.php?Q=Y&G27=436
- Specific Solutions http://www.bio-link.org/GMP/ReadMeFirst.pdf
- Resource for other enzyme labs - http://mdk12.org/instruction/curriculum/hsa/biology/enzyme_activity/enzyme.pdf
- Virtual Lab activity- http://www.phschool.com/science/biology_place/labbench/lab2/intro.html
- Several enzyme multimedia presentations- http://www.worldofteaching.com/biologypowerpoints.html

Materials & Equipment:

- Supplies for ELISA
- Enzyme Activity Supplies
- Items for each group:
  - Fresh pineapple - 1 (can be frozen and used later)
  - Canned pineapple- 1
  - Jello , 2 small boxes any flavor
  - Bowl- 1
  - Boiling & Cold water- 4 cups
  - Spoon -1
  - Paper cups
  - Knife - 1

21st Century Technology Used:

☐	Slide Show Software	☐	Graphing Software	☐	Audio File(s)
☐	Interactive Whiteboard	☐	Calculator	☐	Graphic Organizer
☐	Student Response System	☐	Desktop Publishing	☐	Image File(s)
☐	Web Design Software	☐	Blog	☐	Video
☐	Animation Software	☐	Wiki	☐	Electronic Game or Puzzle Maker
☐	Email	x	Website		