PEEBEDU ELISA Virtual Laboratory

Name: _____ **Period:** _____ **Date:** _____

Open **peebedu.com** and navigate to **ELISA Virtual Laboratory**. Click the **Put on gloves** button to begin. The simulation provides four assay types (Direct, Indirect, Sandwich, and Case Study) with a full set of reagents including antigens, antibodies, blocking buffer, and three substrate options. Use the **Molecular View** button to observe antibody-antigen interactions at the molecular level.

Free Response Questions

Question 1 — Conceptual Analysis

Simulation Task: *Select the* **Direct ELISA** *assay type and complete the full protocol: put on gloves, coat the well with antigen, add blocking buffer, add the primary antibody, wash the plate, add TMB substrate, and read the plate. Then repeat the protocol but* **skip the blocking step** *and observe how the results differ.*

(A) (1 pt) **Describe** the molecular interaction that allows an ELISA to detect a specific protein in a biological sample.

(B) (1 pt) **Explain** why an ELISA well that was not treated with blocking buffer produces a strong color signal even when the target protein is absent from the sample.

(C) (1 pt) **Predict** how ELISA results would differ between a patient whose immune system has produced a large quantity of antibodies against a pathogen and a patient with an immune deficiency that limits antibody production.

(D) (1 pt) **Justify** your prediction.

Question 2 — Analyze Model / Visual Representation

> **Simulation Task:** *Switch between all three assay types —* **Direct***,* **Indirect***, and* **Sandwich ELISA** *— and use the* **Molecular View** *to observe the differences in how antibodies and antigens are layered in each variant. Pay attention to how many antibody layers are present and where the enzyme label is attached in each assay type.*

(A) (1 pt) **Describe** how ELISA enables the detection of a specific protein that is the product of gene expression in a biological sample.

(B) (1 pt) **Explain** why the indirect ELISA produces a stronger signal than the direct ELISA when both are used to detect the same concentration of target protein.

(C) (1 pt) **Draw** In the box below, draw a labeled diagram comparing the molecular architecture of a **Direct ELISA** and an **Indirect ELISA** side by side. For each, show and label: the well surface, the coated antigen, the primary antibody, the secondary antibody (if applicable), the enzyme label, and the substrate. Use arrows to indicate where signal is generated.

Draw your diagram here.

(D) (1 pt) **Explain** how the introduction of a novel pathogen into a population with variation in immune-related protein structure could affect which individuals survive and reproduce.

6.8.A.1