“What Did You Say?”
How Cells Talk to Each Other

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Define the roles of B cells and T cells
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Distinguish between the various types of B cells and T cells

a particular antigen. A second exposure to an antigen results in a more rapid and enhanced immune response.

LO 2.29 - The student can create representations and model

7. Protein gel loading dye, 2X
   Amazon, P-18100-5.0, $38.99
8. Equipment for protein electrophoresis is also needed for this lab

*School districts that have a purchasing contract with Fisher Science may get a discount

Thermo if your district has an account with any of the Fisher companies.

*BioRad offers an educator (EDU) discount and will apply this discount even if the catalog does not show it as an option on that product. The antibody lists for considerably more than $168.00, but they gladly offered me the discount when I contacted them.

*Aside from the antibody, the rest of these lab components will last for more than 1 school year.

Adaptive Immunity Activity

1.
2. Sixlets
3. Large puffballs (or cotton balls)
4. Googly eyes (to put on the puffballs)
5. Smaller sparkly puffballs
6. Poker chips
7. pieces
8. Ziploc sandwich bags

For each group of students (3-4 per group), put the following into a bag:

4 Hershey kisses
1 sleeve of Sixlets
2 large puffballs with eyes
2 blue poker chips
4 sparkly small puffballs
2 red poker chips
Roughly 10-12 pipe cleaner pieces

4. **1 mg/ml anti-albumin solution (prey) -** the anti-albumin comes from Bio Rad as 5 mg/ml. Add 80 µL of anti-albumin antibody to 320 µL Tris buffer to make your working stock. Keep frozen until use.

General Lab Prep Suggestions

1. Aliquot solutions so that you have enough for each lab group to have their own supply of materials.
2. Make sure all materials for day one are kept on ice.
3. If you are crunched for time, it is probably best for you to put the gels in the protein rigs yourself rather than teaching the kids to do it.
4. Students should have a working knowledge of how to pipette, balance a centrifuge, and load a gel.

Analysis Question Answers

1. Explain what is meant by the terms bait protein and prey protein.
   A bait protein is a protein used to attract another protein. It is added to the column first in a pull down assay. A prey protein is a protein that should bind to the bait protein when it is added to the column.
2. Explain the interactions between the beads, the bait protein, and the prey protein.
   There is a substance coated on the beads in the column that will bind the bait protein to the beads. The prey protein will then bind to the bait protein as it is run

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Student Lab Protocol

Fishing for Protein Interactions

interact with each other. A

protein and ends with running a protein gel to see if the two proteins leave the column separately (no interaction) or together (interaction). In a nutshell, it goes something like this:

A buffer is run through the bead column

through the column next. The membrane (or beads) is designed to be attractive to the bait protein so it will stick to them.

time to give the two proteins time to interact (if they will).

A solution is run through the column lastly to remove the bait protein (and hopefully the prey too) from the membrane.

A sample of what comes off the column is taken each time something is run through. These samples are then run out on a protein gel. If the bait and prey proteins show up in the same sample, then they interacted and came off the column together. If they show up in separate samples only, then there was no interaction between them.

Materials

Ice bucket
Micro pipettes and tips
1% albumin solution (bait)
1 mg/ml anti-albumin solution (prey)
Tris buffer solution
Pierce strong anion exchange column
6 microtubes
4-20% tris glycine protein gel
1x tris glycine SDS protein buffer
Loading dye
Protein reference ladder
Protein electrophoresis equipment
Centrifuge
90 hot bath/block
Float rack
Sharpie