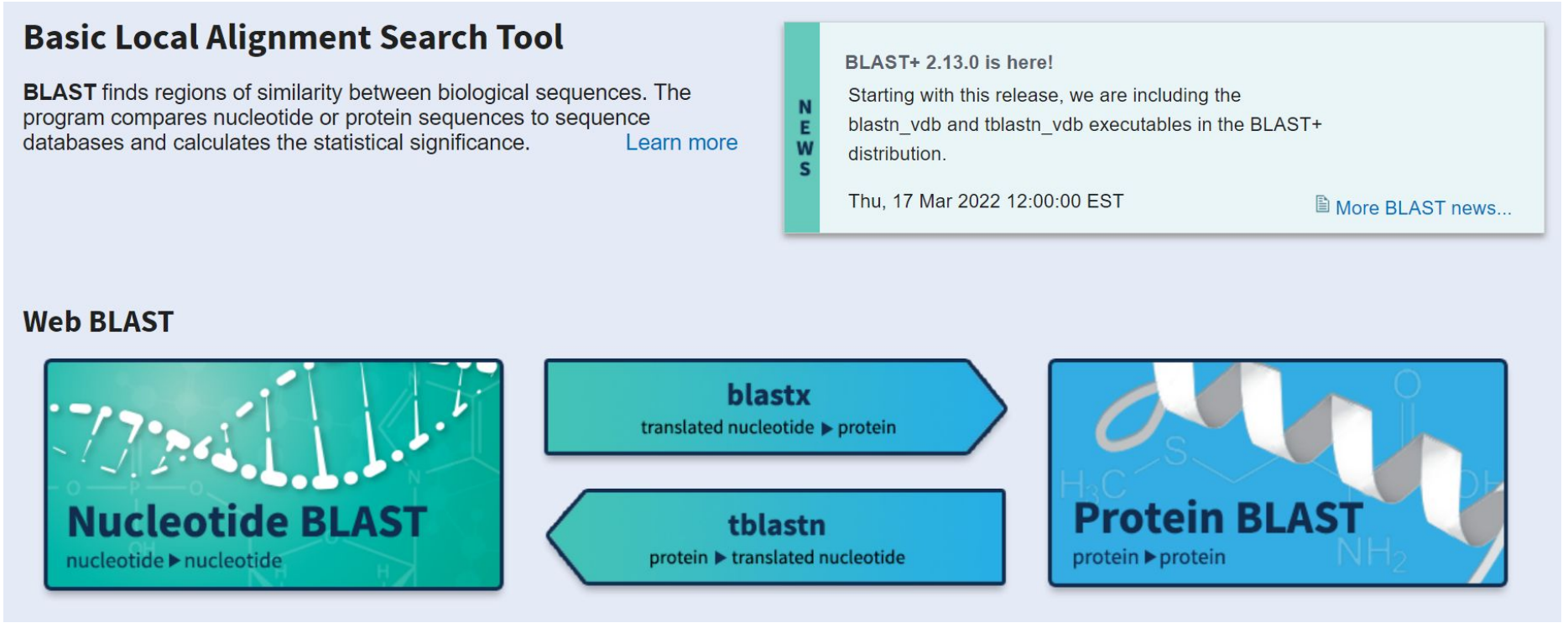
Science Presentation

By: Jacqueline Shum

What I learned:

- Nucleotide BLAST
- Using a Micropipette
- Plasmid Miniprep
- Protein Purification
- Series Dilution
- Protein Concentration Assay
- Protein Gel



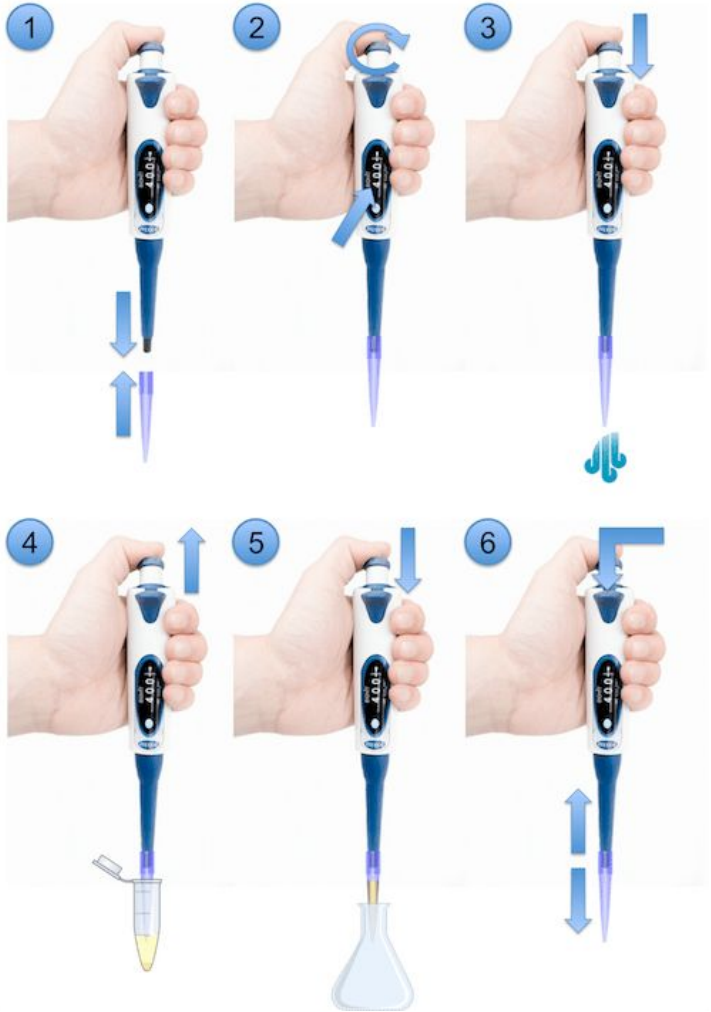
Nucleotide Blast

Using a Micropipette

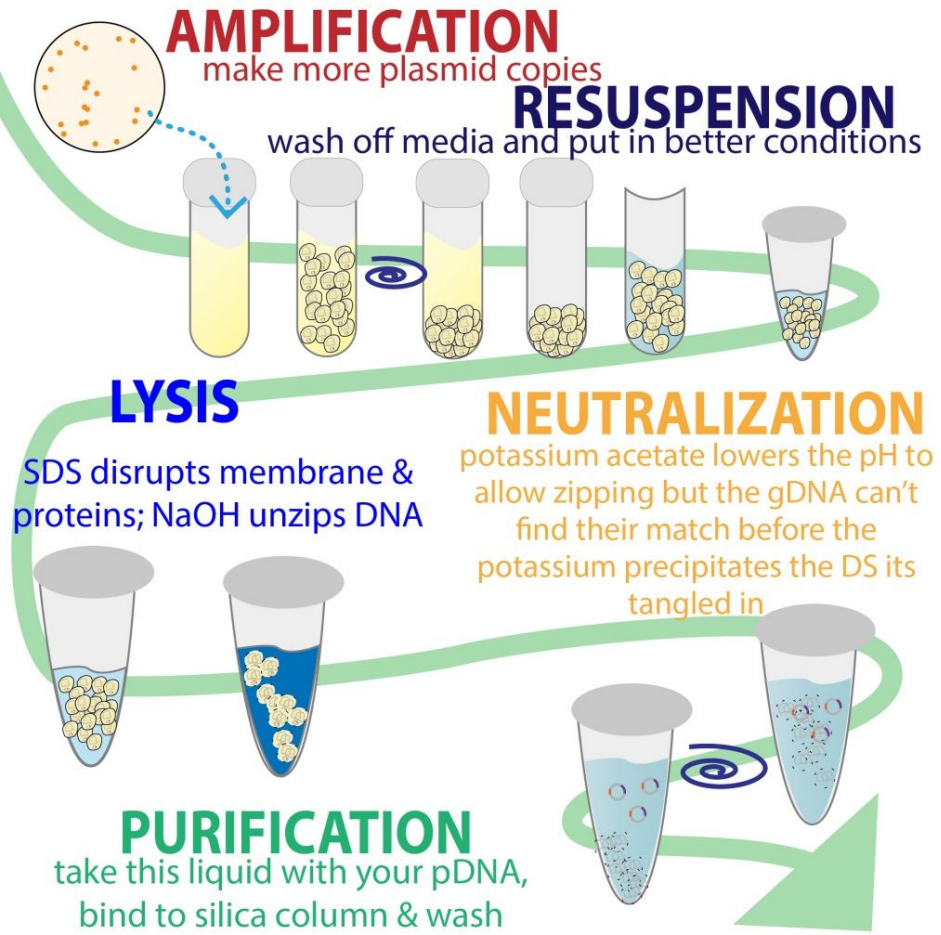
Purpose: To inject/collect an accurate amount of substance.

Things to remember:

- Never adjust the knob above the set amount that a pipette can hold
- Be careful with detergents, can create bubbles
- Never cross contaminate
- Always preset the measure that you want to use



Plasmid Miniprep



Purpose: To separate the plasmid from the cells.

This is important for procedures such as sequencing.

Things to keep in mind:

- Always make sure the centrifuge cap is screwed on properly before beginning
- After lysis, do not vortex to mix. Invert only.

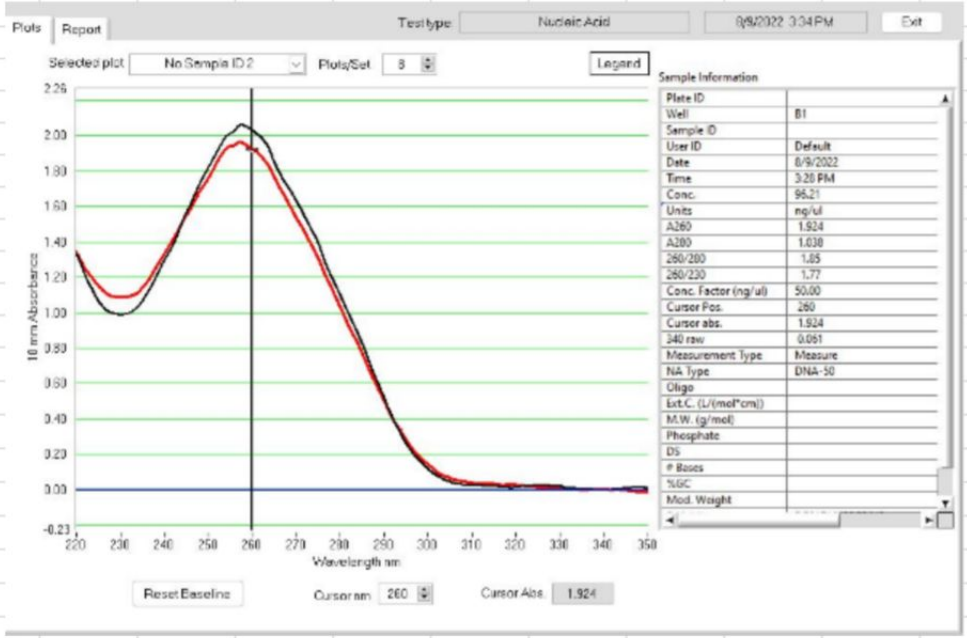
8/9/2022

Jacqueline Shum miniprep

plasmid grow over night in 4 ml LB (Kan), 1.5 ml used for miniprep

Zymo miniprep kit eluted in 25 ul

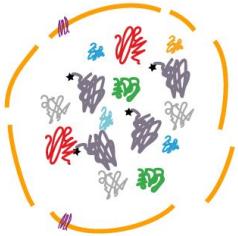
Plate ID	Well	Sample ID	User ID	Date	Time	Conc.	Units	A260	A280	260/280	260/230	Conc. Factor	Cursor Pos.	Cursor abs.	340 raw	NA Type
	A1		Default	8/9/2022	3:28 PM	101.5	ng/ul	2.031	1.108	1.83	2.06	50	260	2.031	-0.012	DNA-50
	B1		Default	8/9/2022	3:28 PM	96.21	ng/ul	1.924	1.038	1.85	1.77	50	260	1.924	0.061	DNA-50



protein purification

lysis

break cells open



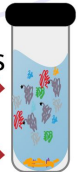
ultracentrifugation

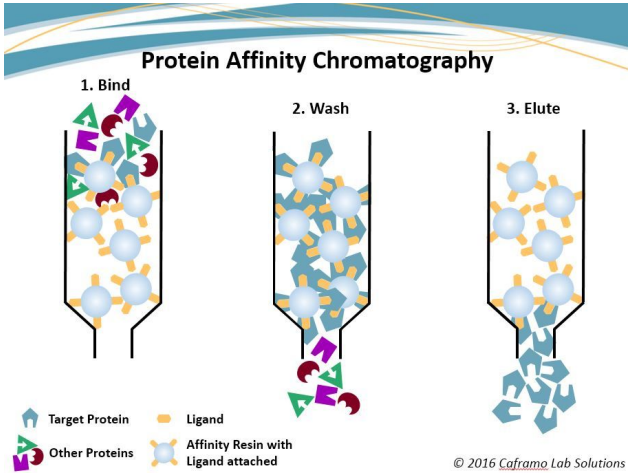
collect the soluble proteins

spin ***really*** fast to pellet out membrane bits

LOTS of water-soluble proteins
ONE of which you want....

membrane bits, etc.





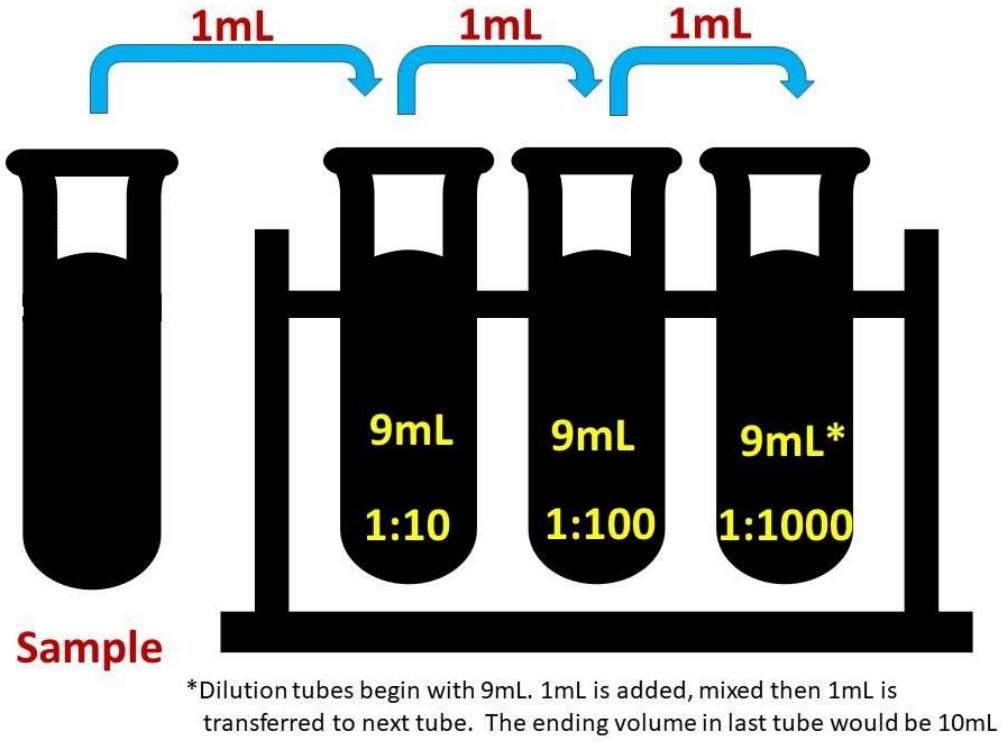
Purpose: To isolate a target protein.

This is important as it allows for us to study the protein by itself and get more accurate results.

Things to know:

- Lysis buffer contains protease inhibitor which is important to prevent denaturation of proteins
- Imidazole is used to elute out DNA

Series Dilution



*Dilution tubes begin with 9mL. 1mL is added, mixed then 1mL is transferred to next tube. The ending volume in last tube would be 10mL

Purpose: To produce samples of different concentrations.

This is important as it allows for us to determine the concentration of a sample.

Things to know:

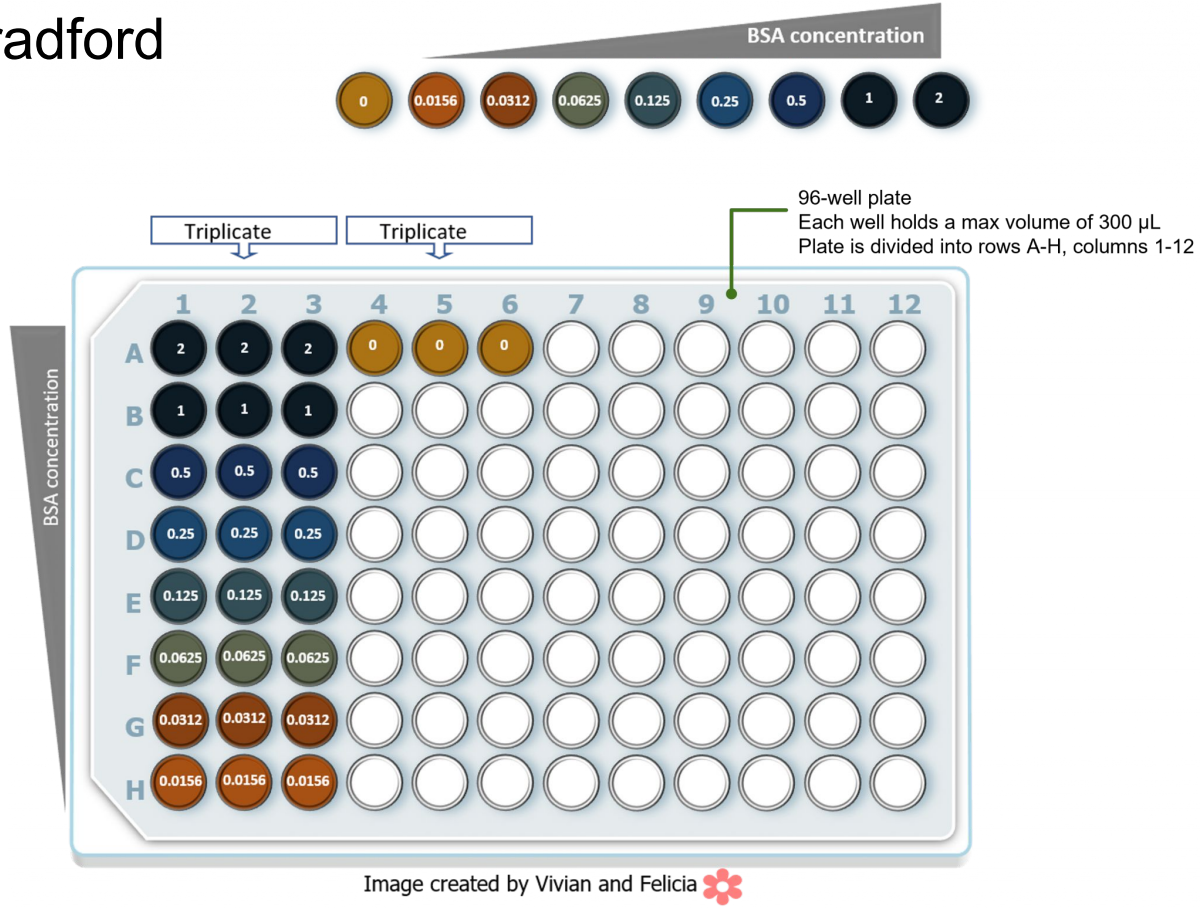
- Dilution Factor
  - Volume of Stock Transferred : Total Volume
- Produces fractions
- The first fraction that is produced is likely to have the highest concentration

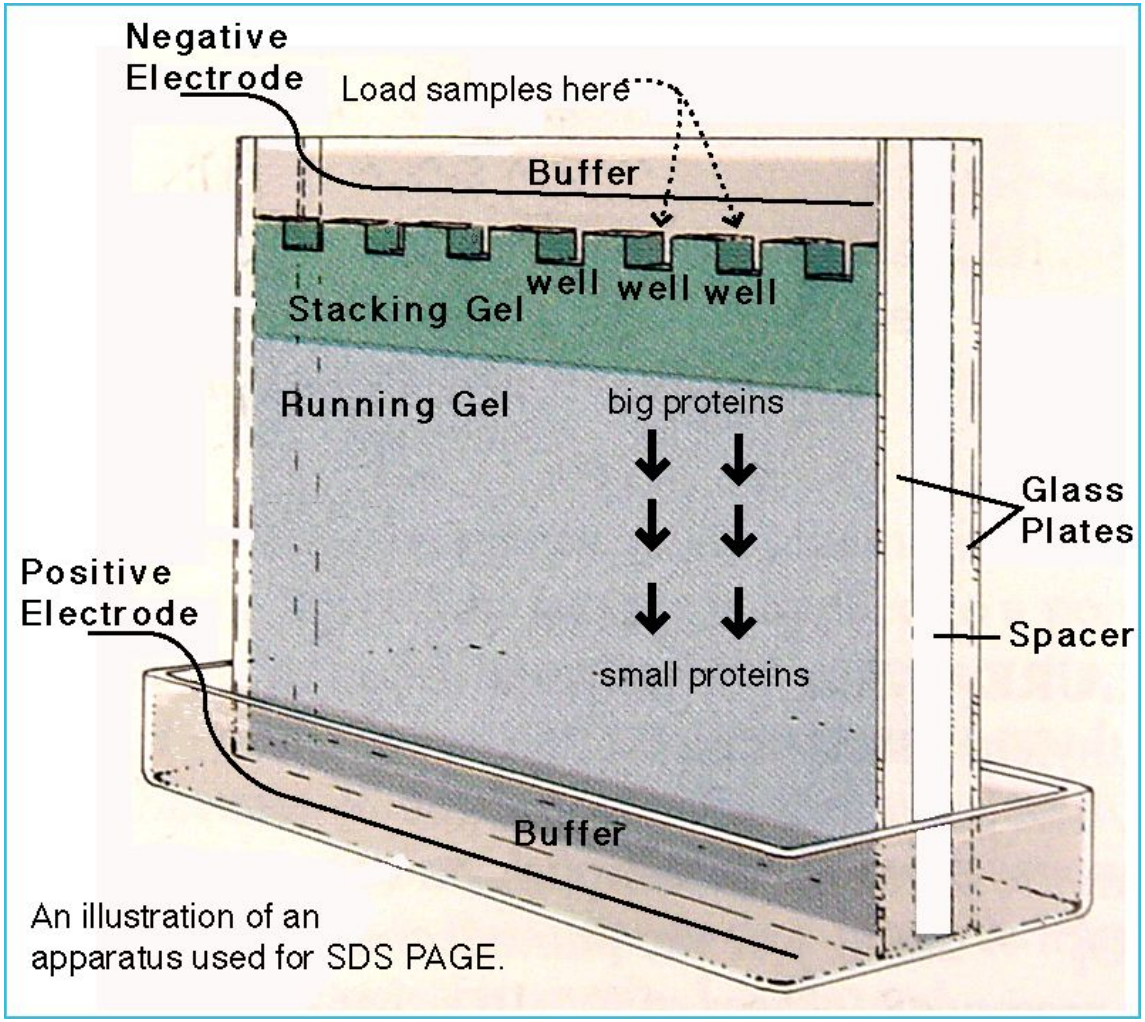
Protein Concentration/Bradford Assay

Purpose: Determine the concentration of a sample in a solution.

Things to keep in mind:

- Blue dye binds to protein to express blue color
- Triplicates are made to determine more accurate results





An illustration of an apparatus used for SDS PAGE.

Protein Gel

Purpose: To separate proteins by molecular weight.

This is important as it allows for us to determine the concentration of a sample.

Things to know:

- SDS in gel is used to eliminate movement of protein based off structure and charge
- Stacking Gel keeps protein together

Thanks for Listening!