

Brianna Samuels

Dr. Urbinati

Cell Biology + Lab

January 22, 2018

SDS Page/Western Blot Assignment

Protein electrophoresis is a way for us to separate and visualize proteins. This is useful because we can determine how many proteins and what types of proteins are present in a sample fairly quick. Something that is important to note is that proteins are able to be separated into one dimensional or two dimensional polyacrylamide gel electrophoresis. One-dimensional PAGE separates by size whereas two-dimensional PAGE separates by size and charge. Polyacrylamide is the best type of material when specifically visualizing proteins due to the better resolution quality. In electrophoresis, SDS protein complexes travel through the polyacrylamide gel. The smaller the molecule the faster it travels because it is more easily able to fit through the small pores of the gel. The technique most commonly used to complete this process is the western blot technique. The way to go about it is to first separate the protein with the polyacrylamide gel. Depending on how far they move, this will tell us the logarithmic size of the protein. The next step of this process is to blot the proteins by electrophoretic transfer to a membrane. Normally you want to do this in the same pattern seen on the gel. After the blotting is complete, a labeled antibody is used to identify the target protein. Once it binds, a wash is required to ensure that there aren't extra antibodies present. Then, the membrane is incubated with another antibody linked to an enzyme. This causes the two antibodies present to combine. Finally a chromogenic

substrate is added so that the enzyme can catalyze and start a reaction that will mark the protein of interest. The step by step process is depicted below in Figure 1.¹

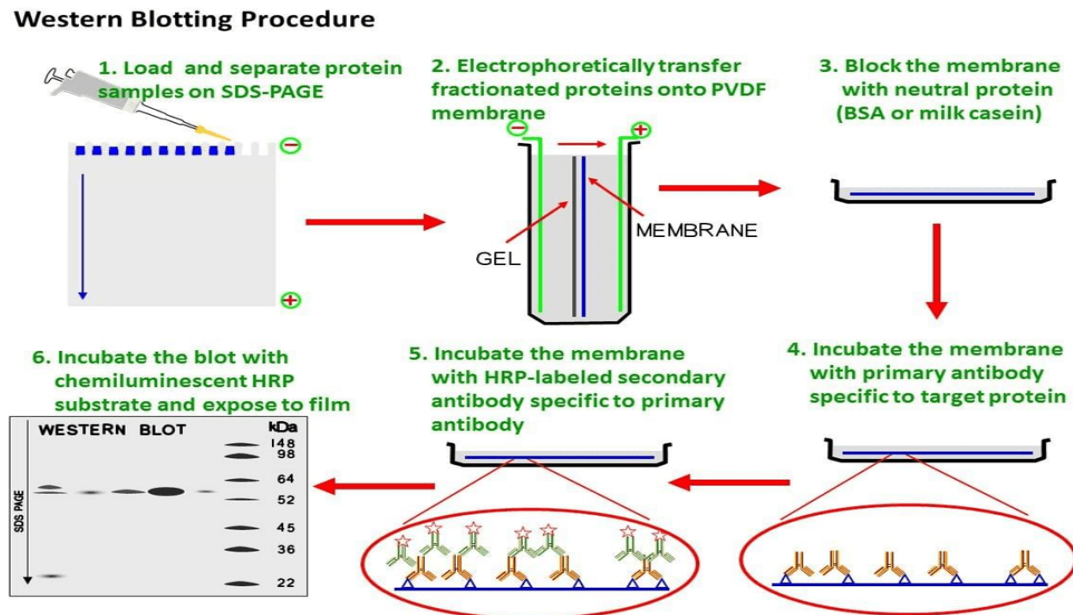


Figure 1: This figure shows the steps of what a typical Western Blot procedure looks like.

¹ “Western Blot Technique: Principle, Procedures and Uses -.” *Home* -, 13 May 2017, microbeonline.com/western-blot-technique-principle-procedures-advantages-and-disadvantages/.