QUICK GUIDE: Protein Electrophoresis

WHAT IS SDS-PAGE?

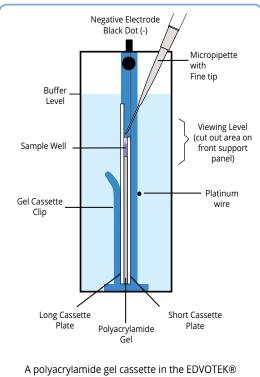
SDS polyacrylamide gel-electrophoresis, or SDS-PAGE, is a technique that is used to separate proteins according to their molecular weight.

HOW DOES SDS-PAGE SEPARATE PROTEINS?

Proteins produce a unique challenge for electrophoresis because they have complex shapes and different charges, which affect how they migrate through the gel. In order to accurately separate proteins by molecular weight and not by shape or charge, the secondary structure of the protein is unfolded using the anionic detergent sodium dodecyl sulfate (SDS) and a reducing agent. The reducing agent breaks covalent bonds that link protein subunits.

After denaturation, the mixture of proteins is added into depressions (or "wells") within a gel, and then an electrical current is passed through the gel. Because the SDS-protein complex has a strong negative charge, the current drives the proteins through the gel towards the positive electrode. At first glance, a polyacrylamide gel appears to be solid. On the molecular level, the gel contains channels through which the proteins can pass. Small proteins move through these holes easily, but large proteins have a more difficult time squeezing through the tunnels. Because molecules of different sizes travel at different speeds, they separate into discrete "bands" within the gel. After the current is stopped, the bands are visualized using a stain that sticks to proteins.

WHAT DO I NEED FOR PROTEIN ELECTROPHORESIS?



Vertical Electrophoresis Apparatus, Cat. #581.

To complete protein electrophoresis, you'll need some equipment! You'll use a vertical electrophoresis apparatus like the MV10 (Cat. #581), a power supply like the QuadraSource (Cat. #5010-Q), variable micropipettes and tips, and polyacryl-amide gels (Cat. #651). Of course, you'll also need some protein samples and some Tris-Glycine SDS Buffer (Cat. #655)!



HOW DO I VISUALIZE MY PROTEINS AFTER ELECTROPHORESIS?

Unlike how we can visualize DNA electrophoresis with fluorescent stains like SYBR® Safe, protein gels have to be stained after the electrophoresis using a stain that will bind to the proteins. We recently designed a new stain, FlashBlue™ Protein Stain, which can be used to stain and visualize your gel in under an hour!

