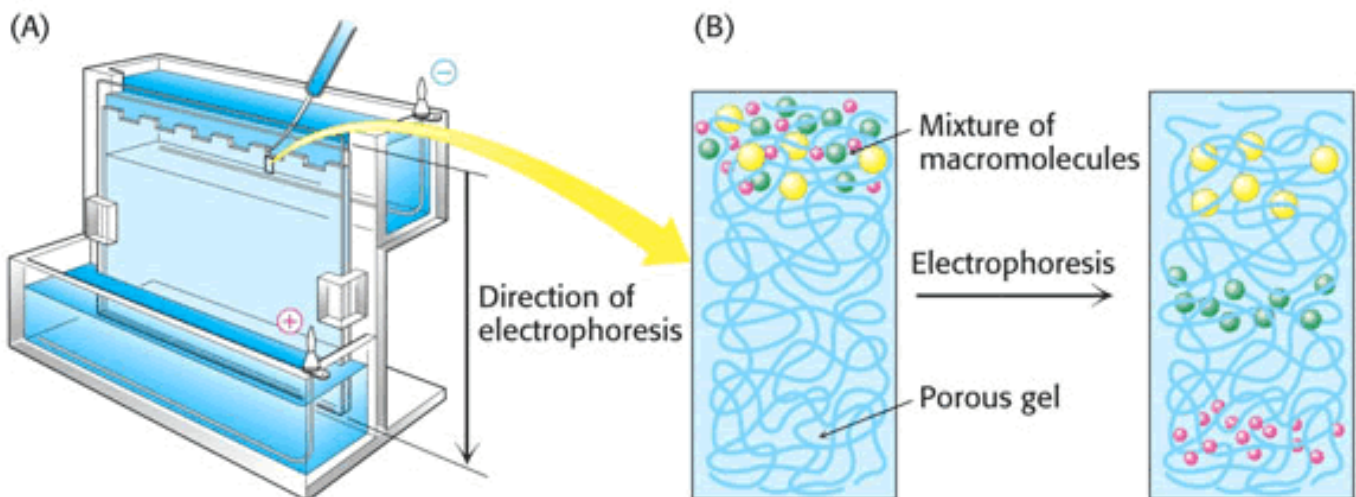
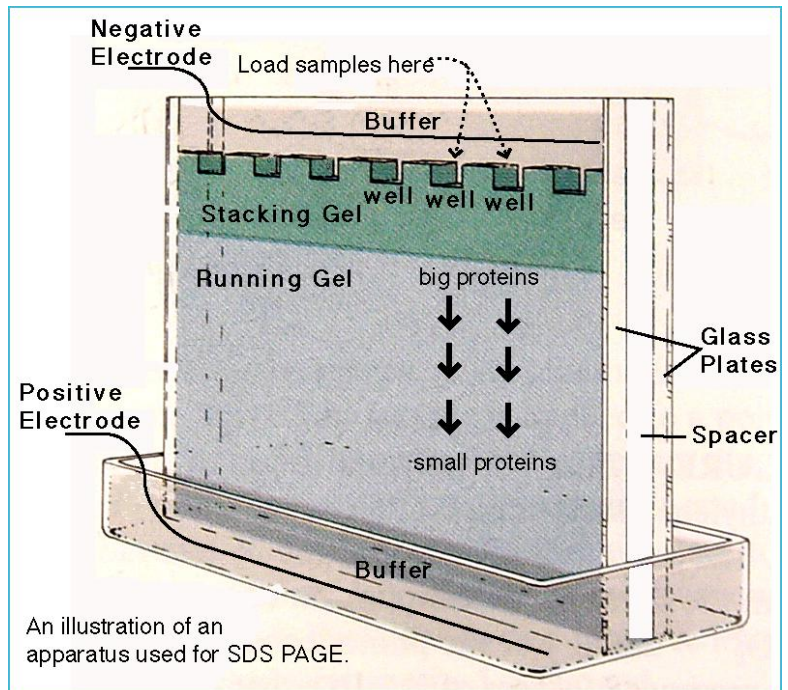


SDS-PAGE

Sodium Dodecyl Sulfate-Polyacrylamide gel Electrophoresis

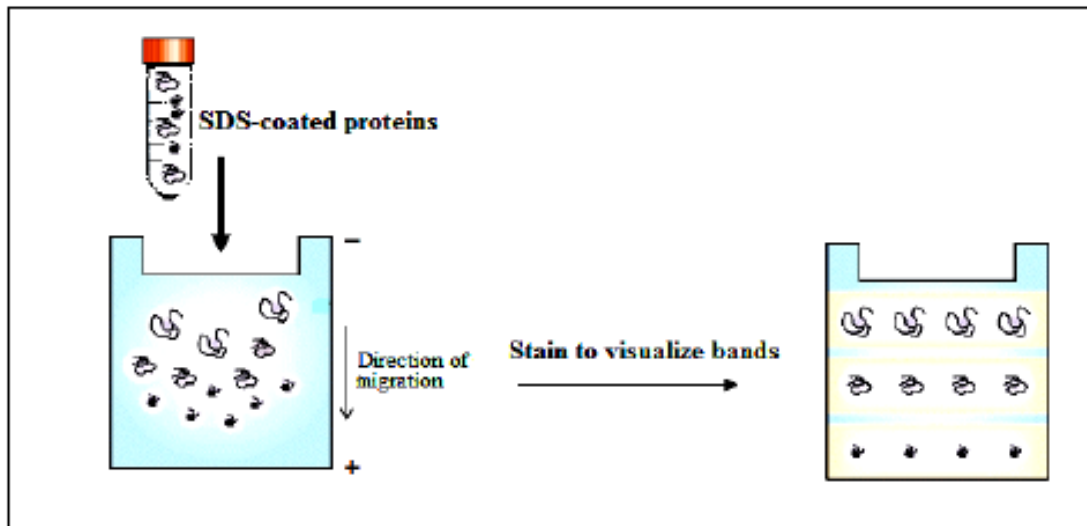
An electric field is applied across the gel, causing the negatively-charged proteins to migrate across the gel towards the positive (+) electrode (anode). Depending on their size, each protein will move differently through the gel matrix: short proteins will more easily fit through the pores in the gel; while larger ones will have more difficulty (they encounter more resistance). After a set amount of time (usually a few hours, though this depends on the voltage applied across the gel; protein migration occurs more quickly at higher voltages, but these results are typically less accurate than at those at lower voltages) the proteins will have differentially migrated based on their size; smaller proteins will have traveled farther down the gel, while larger ones will have remained closer to the point of origin. Proteins may therefore be separated roughly according to size (and thus molecular weight).

Following electrophoresis, the gel may be stained (most commonly with Coomassie Brilliant Blue R-250, allowing visualization of the separated proteins, or processed further (e.g. Western blot). After staining, different proteins will appear as distinct bands within the gel. It is common to run molecular weight size markers of known molecular weight in a separate lane in the gel, in order to calibrate the gel and determine the approximate molecular mass of unknown proteins by comparing the distance travelled relative to the marker.

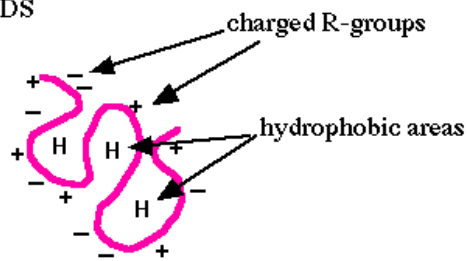


Sample preparation:

The sample to be analyzed is mixed with SDS, an anionic detergent which denatures secondary and non-disulfide-linked tertiary structures, and applies a negative charge to each protein in proportion to its mass. Heating the samples to at least 60°C further promotes protein denaturation, helping SDS to bind. Treatment with (dithiothreitol, β -mercaptoethanol) is also denatured agent by reducing all disulfide bond present in the sample.



BEFORE SDS



AFTER SDS

