

Principles of Gel Electrophoresis

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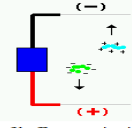
Electrophoresis is a technique used to separate and sometimes purify macromolecules - especially proteins and nucleic acids - that differ in size, charge or conformation. As such, it is one of the most widely-used techniques in biochemistry and molecular biology.

When charged molecules are placed in an electric field, they migrate toward either the positive or negative pole according to their charge. In contrast to proteins, which can have either a net positive or net negative charge, nucleic acids have a consistent negative charge imparted by their phosphate backbone, and migrate toward the anode.

Proteins and nucleic acids are electrophoresed within a matrix or "gel". Most commonly, the gel is cast in the shape of a thin slab, with wells for loading the sample. The gel is immersed within an electrophoresis buffer that provides ions to carry a current and some type of buffer to maintain the pH at a relatively constant value.

The gel itself is composed of either agarose or polyacrylamide, each of which have attributes suitable to particular tasks:

Agarose is a polysaccharide extracted from seaweed. It is typically used at concentrations of 0.5 to 2%. The higher the agarose concentration the "stiffer" the gel. Agarose gels are extremely easy to prepare: you simply mix agarose powder with buffer solution, melt it by heating, and pour the gel. It is also non-toxic.



1. What is the purpose of using a Gel Electrophoresis?

2. In what direction do charged molecules move with electricity?

3. What is the charge on DNA?

4. In what direction do DNA fragments move across the gel?

5. We use agarose when we analyze DNA. What are the characteristics of agarose?