

Molecular Biology " Buffers "

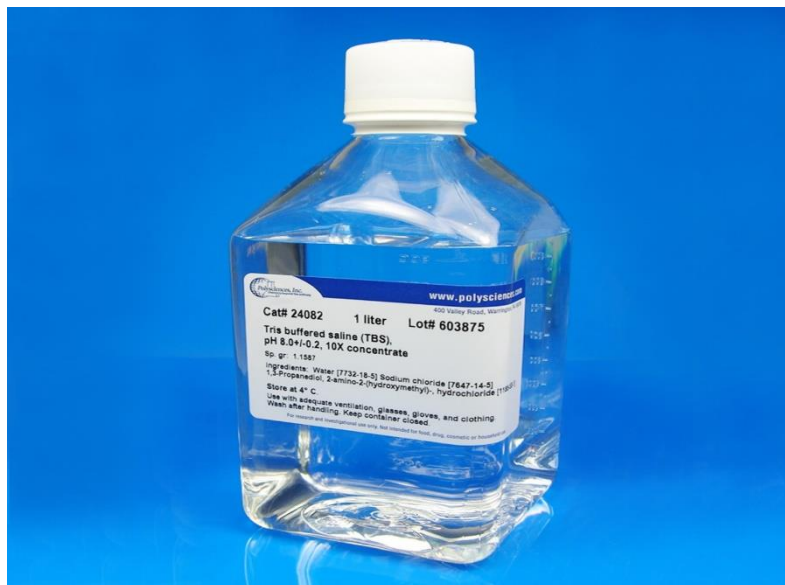
Buffers

Buffer: is a solution containing either weak acid and its salt or weak base and its salt which resist changing in pH.

Buffer: also defined as (solution resist changing in pH in dilution or in addition of acid or base. Examples (Phosphate buffer, Tris-buffer).

Buffer capacity: means the ability of buffer to resist changing in pH.

Buffer agent: is a weak acid or weak base in buffer.



Buffers Properties

- 1- It has known pH.
- 2- It has pH value don't change even in long time or adding of strong acid or base.

Types of Buffers

- 1- TBE (Tris-borate-EDTA) buffer.
- 2- TSE (Tris-sucrose-EDTA) buffer.
- 3- STET (Sucrose-Tris-EDTA-TritonX- 100) buffer.

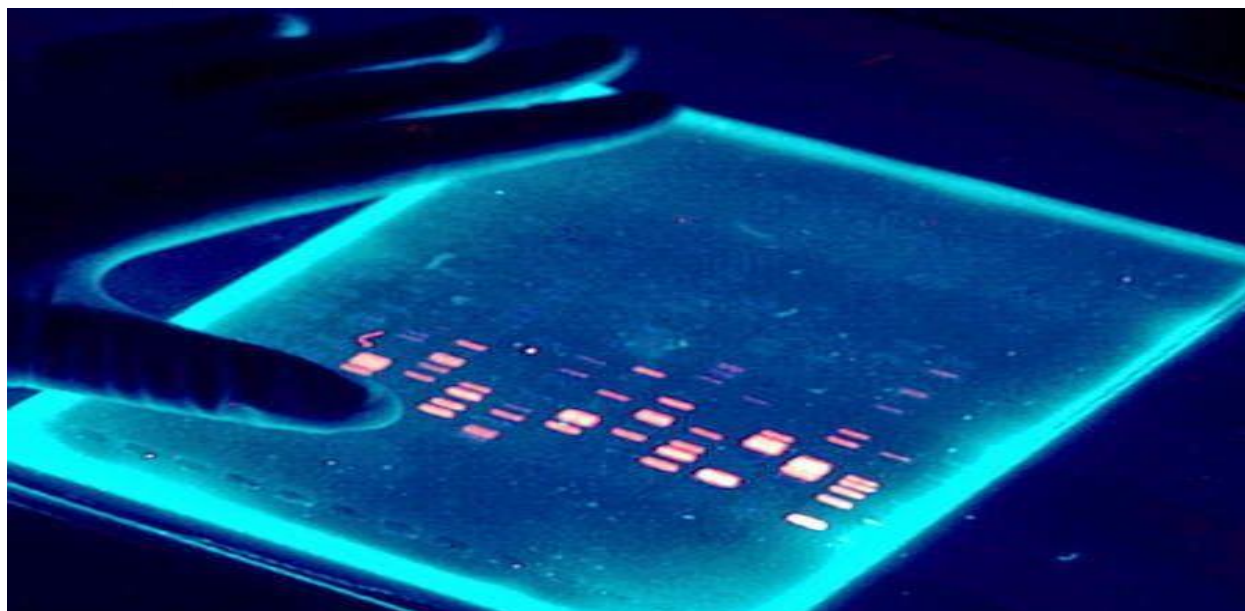
Tris buffers are solution effective in basic conditions which keeps the DNA deprotonated and soluble in water.

EDTA is a chelator agent divalent cations, for example Mg^{+2} that act as co-factor for many enzymes including nuclease, therefore the role of EDTA is to protect the nucleic acid from enzymatic degradation.

Mg⁺² is also act as co-factor for many useful DNA modifying enzymes such as restriction enzymes and DNA polymerase

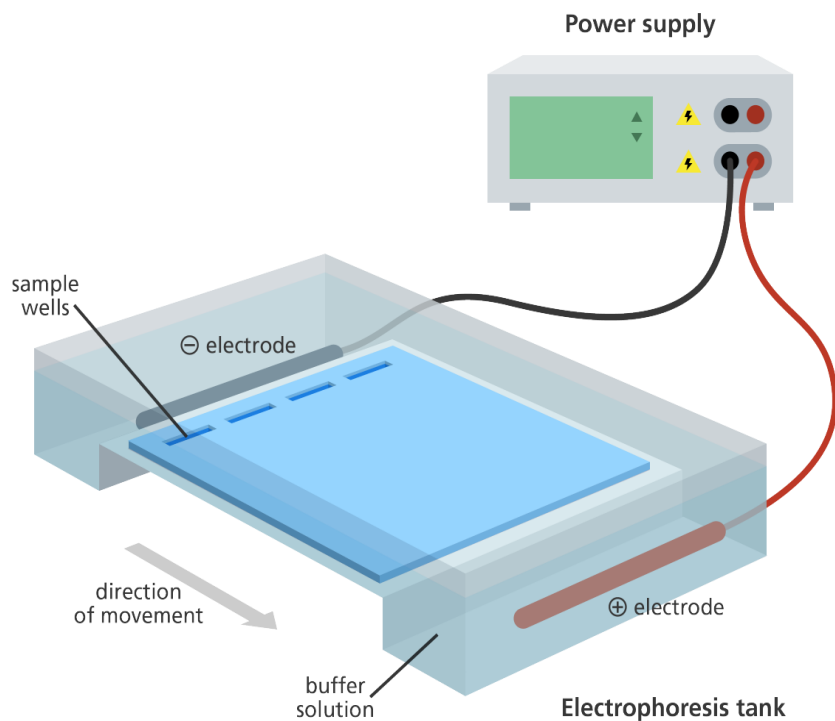
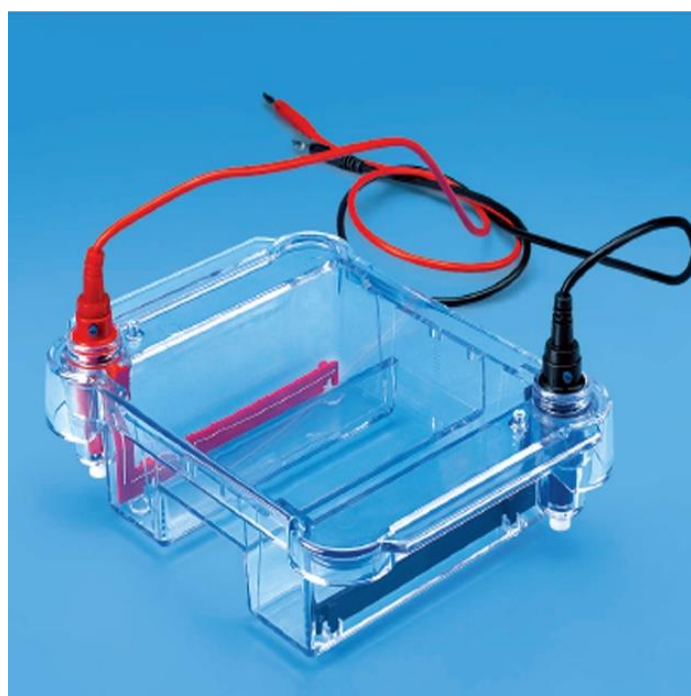
These buffers are used in procedures of nucleic acid such as in electrophoresis.

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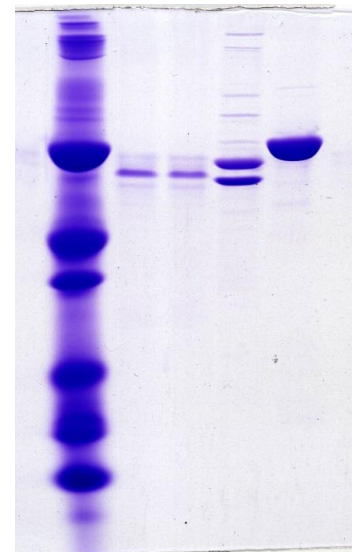
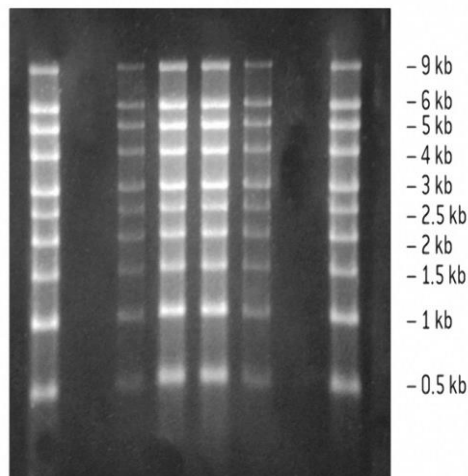
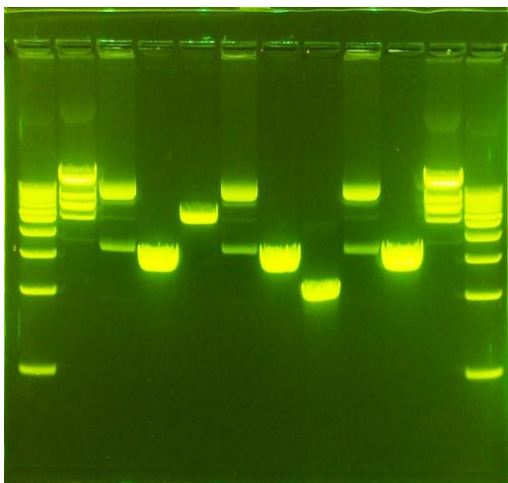
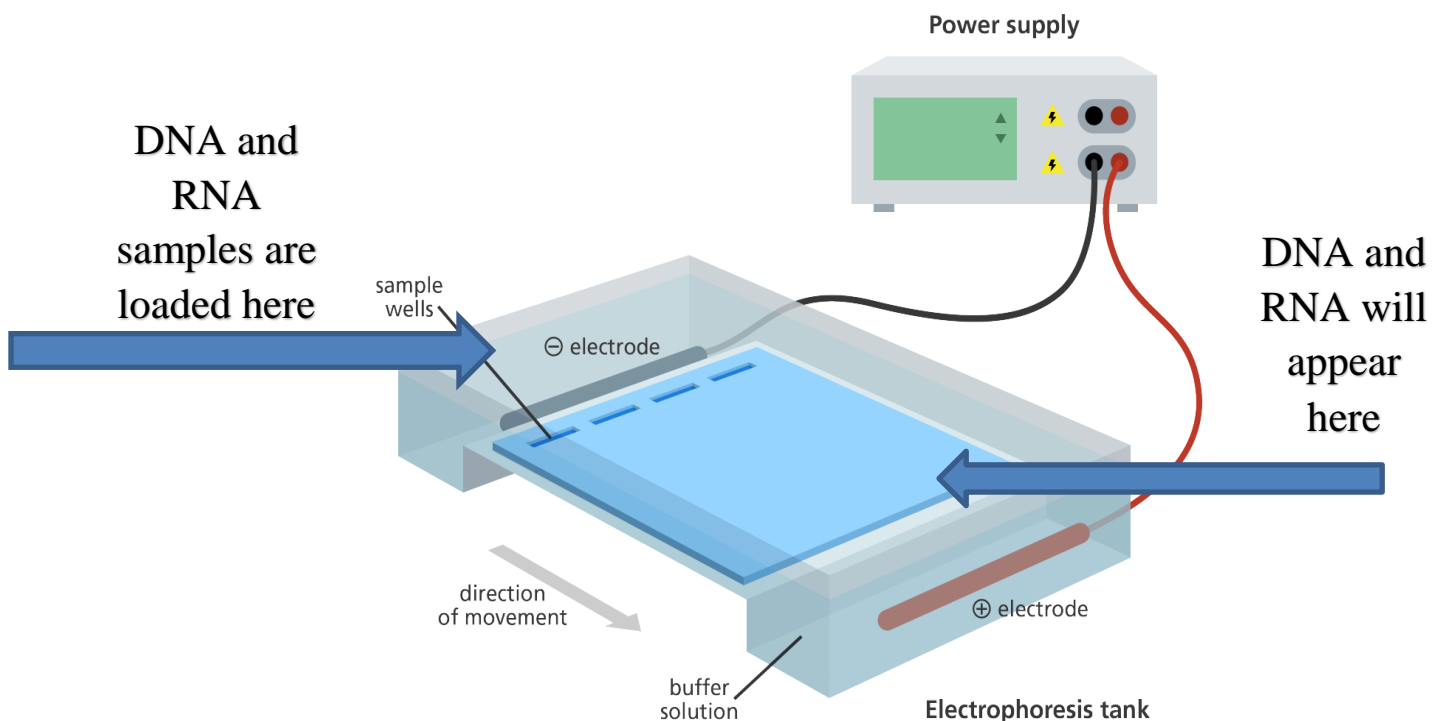
Electrophoresis

Electrophoresis is a laboratory technique used to separate DNA, RNA, or protein molecules based on their size and electrical charge. An electric current is used to move molecules to be separated through a gel.



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- To separate DNA using **agarose gel electrophoresis**, the DNA is loaded into pre-cast wells in the **gel** and a current applied. The phosphate backbone of the DNA (and RNA) molecule is negatively charged, therefore when placed in an electric field, DNA fragments will migrate to the positively charged anode.



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Three purposes using a buffered solution in gel electrophoresis:

- 1- It provides the necessary ion to conduct electricity,
- 2- Helps maintain a stable pH and a stable temperature.
- 3- A buffer also keeps the gel from melting.