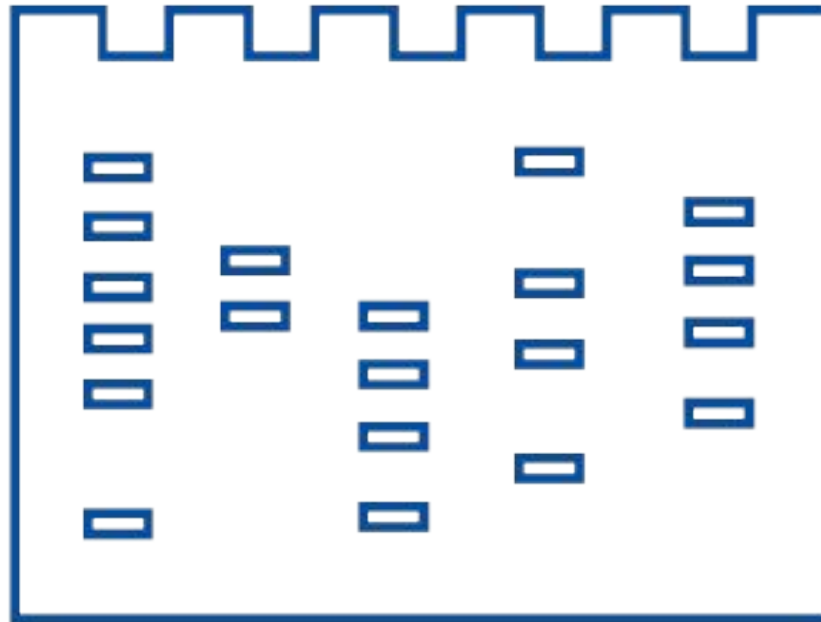


Preparative electrophoresis



Done by: Naizabayeva D.
Accepted by: Kenzhebayeva S.S.

Electrophoresis **by the purpose** of application is divided to analytical and preparative electrophoresis.

Analytical electrophoresis- for the determination of the sample composition and rarely for the obtaining little amount of separating substance (protein)

Preparative electrophoresis- targets the obtaining significant amount of purely separated substance (protein)

Preparative electrophoresis



Stages



Separation



Polyacrylamide gel electrophoresis



1. Gel preparation
2. Sample adding
3. Separation
4. Staining

Elution



Cutting of certain segment (band) of separated protein

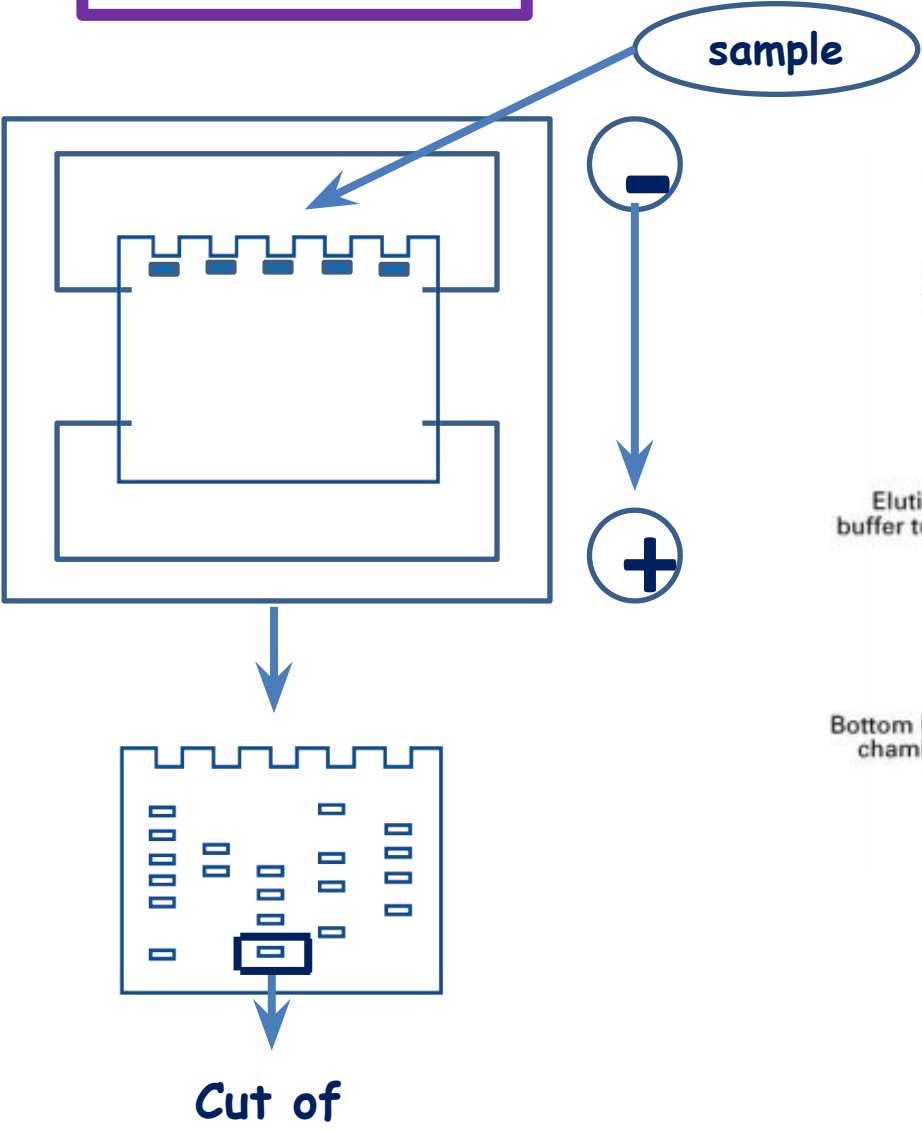


Elution by the electrophoresis

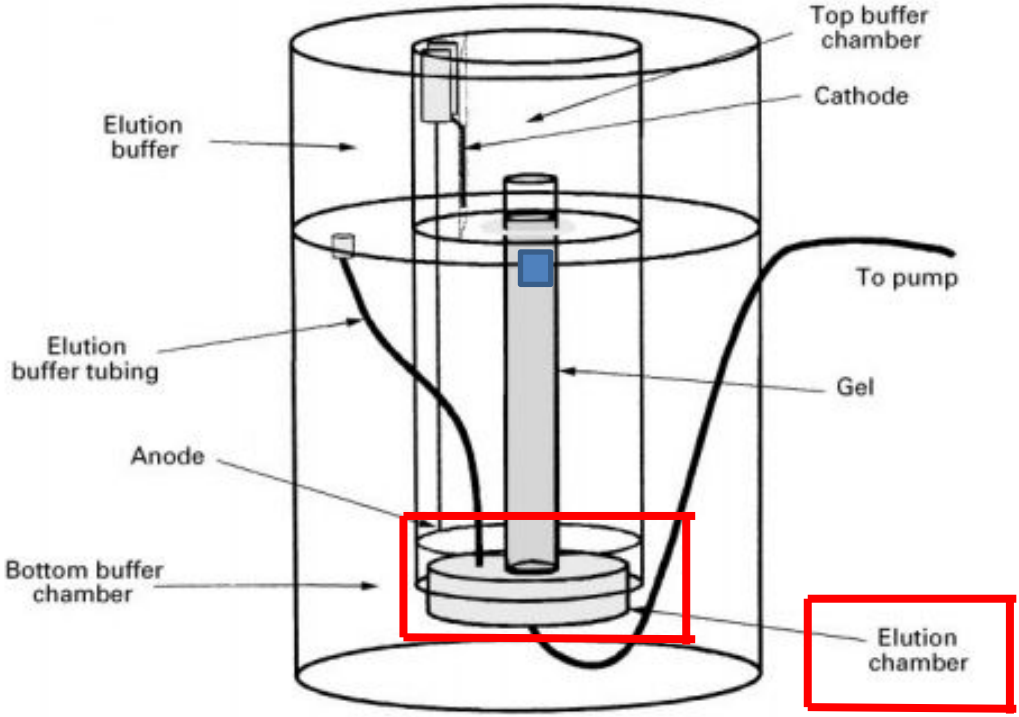


Column electrophoresis

Separation



Elution

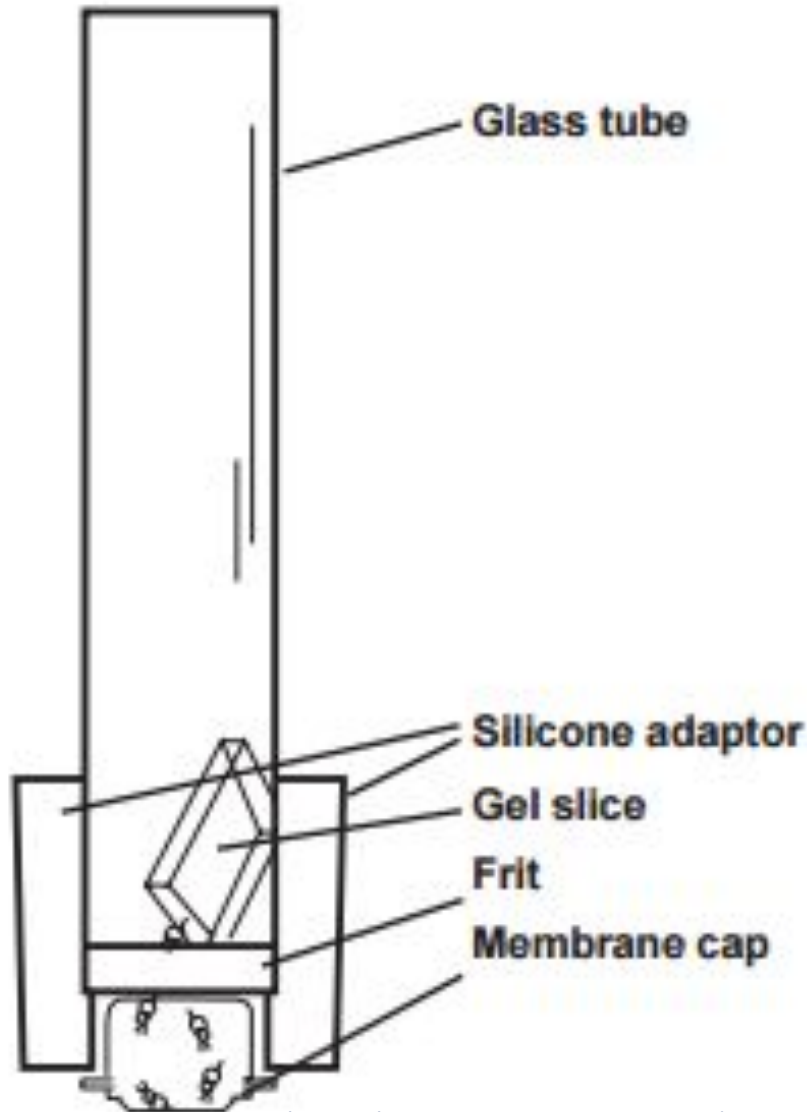


The Model 422 electro-eluter is an electroelution cell for preparative recovery of biomolecules from agarose and acrylamide gels. The eluter combines with the tank and lid of the Mini Trans-Blot[®] cell (or older Mini-PROTEAN[®] II or Mini-PROTEAN 3 cells) to elute macromolecules from single or multiple gel slices. Easy to assemble, the electro-eluter has six vertical glass tubes connecting the upper and lower buffer chambers.



A frit at the bottom of each tube retains the gel slice but permits macromolecules to migrate through when current is applied. When the macromolecules have passed through the frit, they are collected in the membrane cap for further analysis or testing.

Depending on the buffer system, the Model 422 electro-eluter can be used for elution or dialysis. Setup is quick and easy and the sample is collected in 400-600 μ l. The Model 422 electro-eluter can be used for one to six samples without increasing the run time (3-5 hr) or decreasing sample yield.



Membrane cap may be reused for at least five complete runs without decreasing the yield. Refrigerate the membrane cap in elution buffer with 0,05% sodium azide (NaN_3). It is not necessary to reheat or resoak the membrane caps after the first use.

Molecules are retained by dialysis membrane which is molded into a cap

1. Protein Elution Buffer

Tris base	3.0 g	25 mM
Glycine	14.4 g	192 mM
SDS	<u>1.0 g</u>	0.1%
	to 1 liter with dH ₂ O	

Store at 4 °C. Warm to 37 °C before use if precipitation occurs.

2. Volatile Buffer (for protein elution and concentration)

Ammonium bicarbonate (NH ₄ HCO ₃)	3.95 g	50 mM
SDS	<u>1.0 g</u>	0.1%
	to 1 liter with dH ₂ O	

Make up only 1 liter at a time because the buffer will volatilize. Store at 4° C.

***Note:** After elution volatile buffer is lyophilized in a spin-vacuum, leaving concentrated protein

Thanks
for
attention