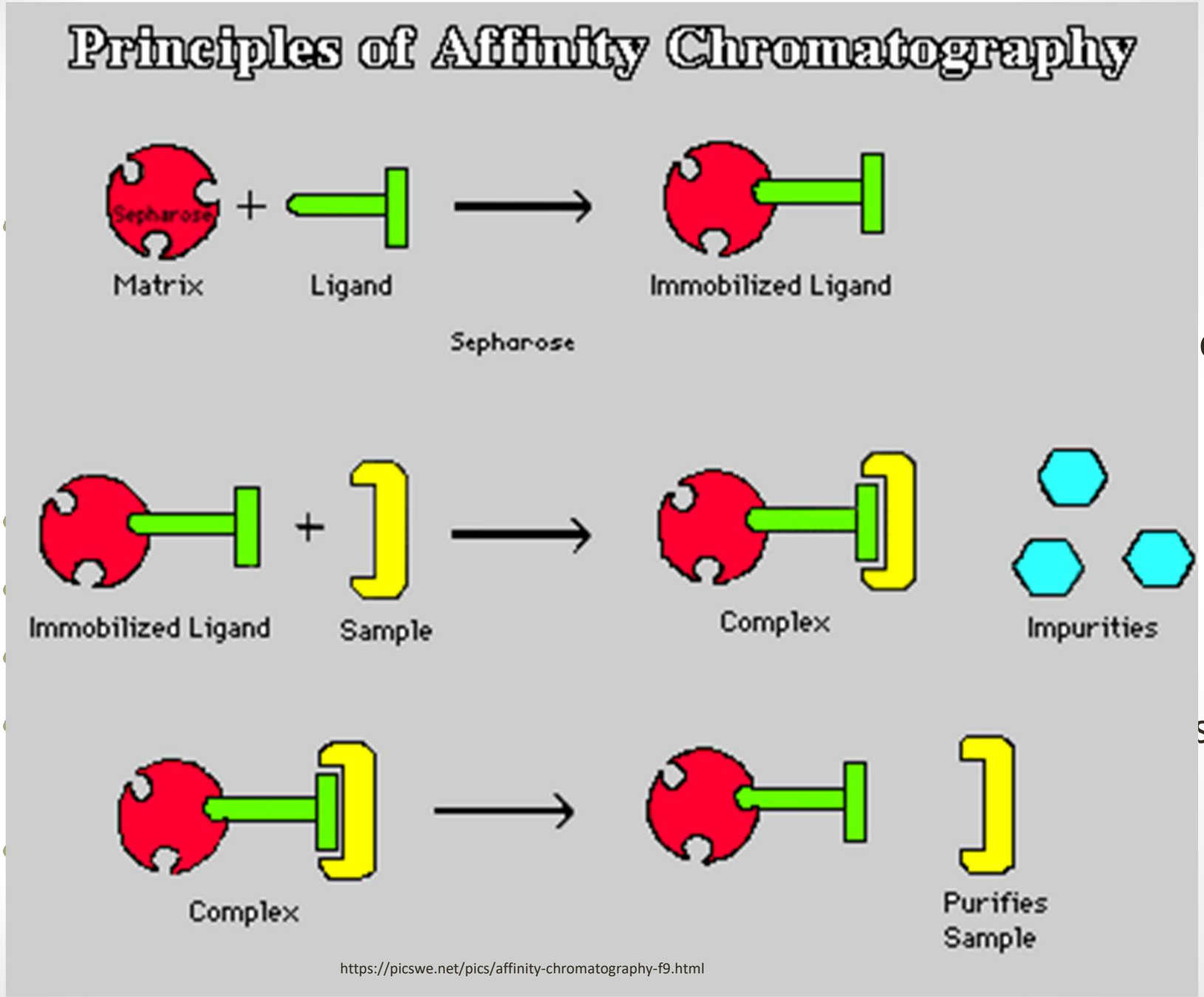
Affinity Chromatography

Loh Yu Min, Chatraporn Chatnithikul (Yam)



https://picswe.net/pics/affinity-chromatography-f9.html

e

s,

Basic requirements

- **Matrix:**

✓Chromatographic media

✓Usually in **bead form** to which a specific ligand is covalently bounded and presen on the surface of the bead

✓Resin = Affinity ligand + Matrix

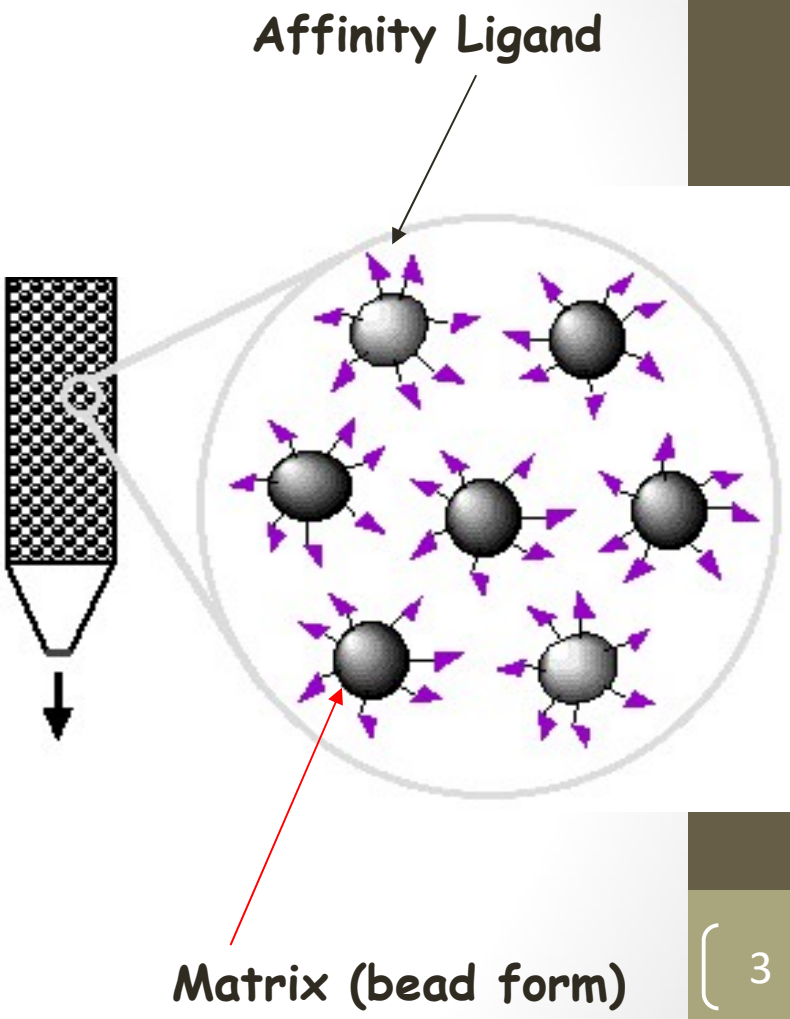
✓Chemically inert and stable (insoluble in solvents, cleansing agents and buffers)

✓easily coupled to a ligand or the spacer arm of the ligand

- **Immobilized Ligands:**

✓**Complementary shape** with the binding site on target molecule

✓Immobilized- **Covalently** bonded to the spacer arm attached on the matrix →Solid phase



http://affinity-chromatography.tripod.com/ac.html

Basic requirements

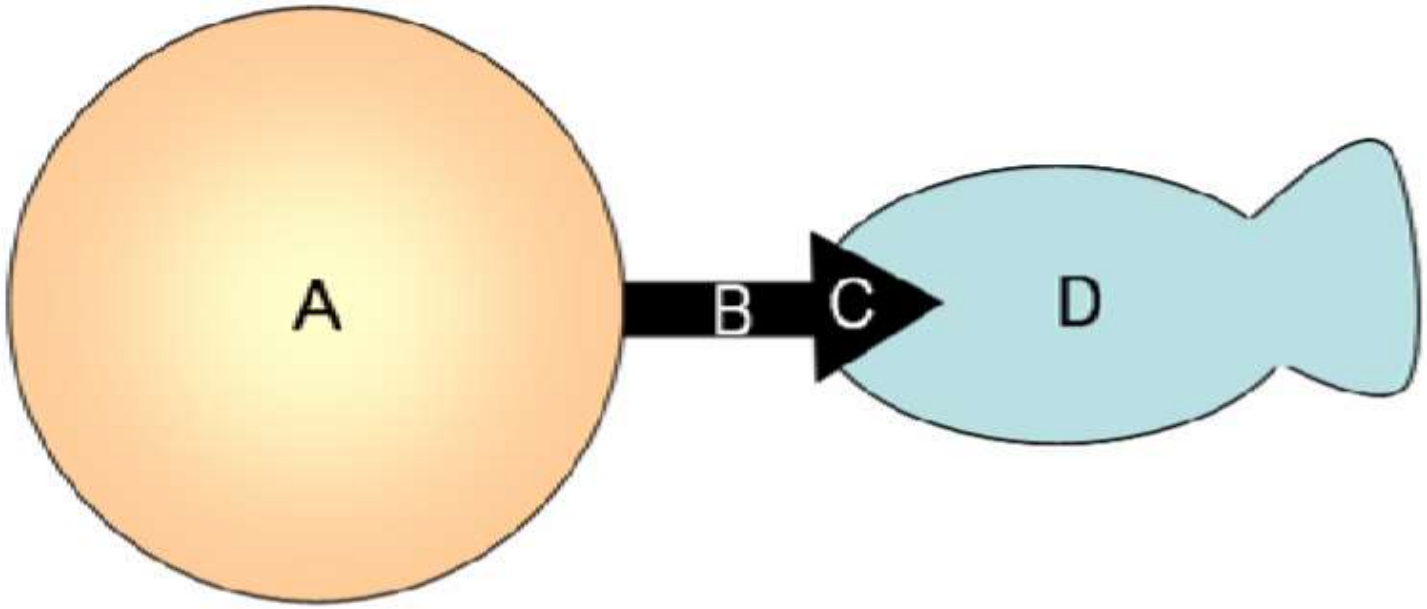
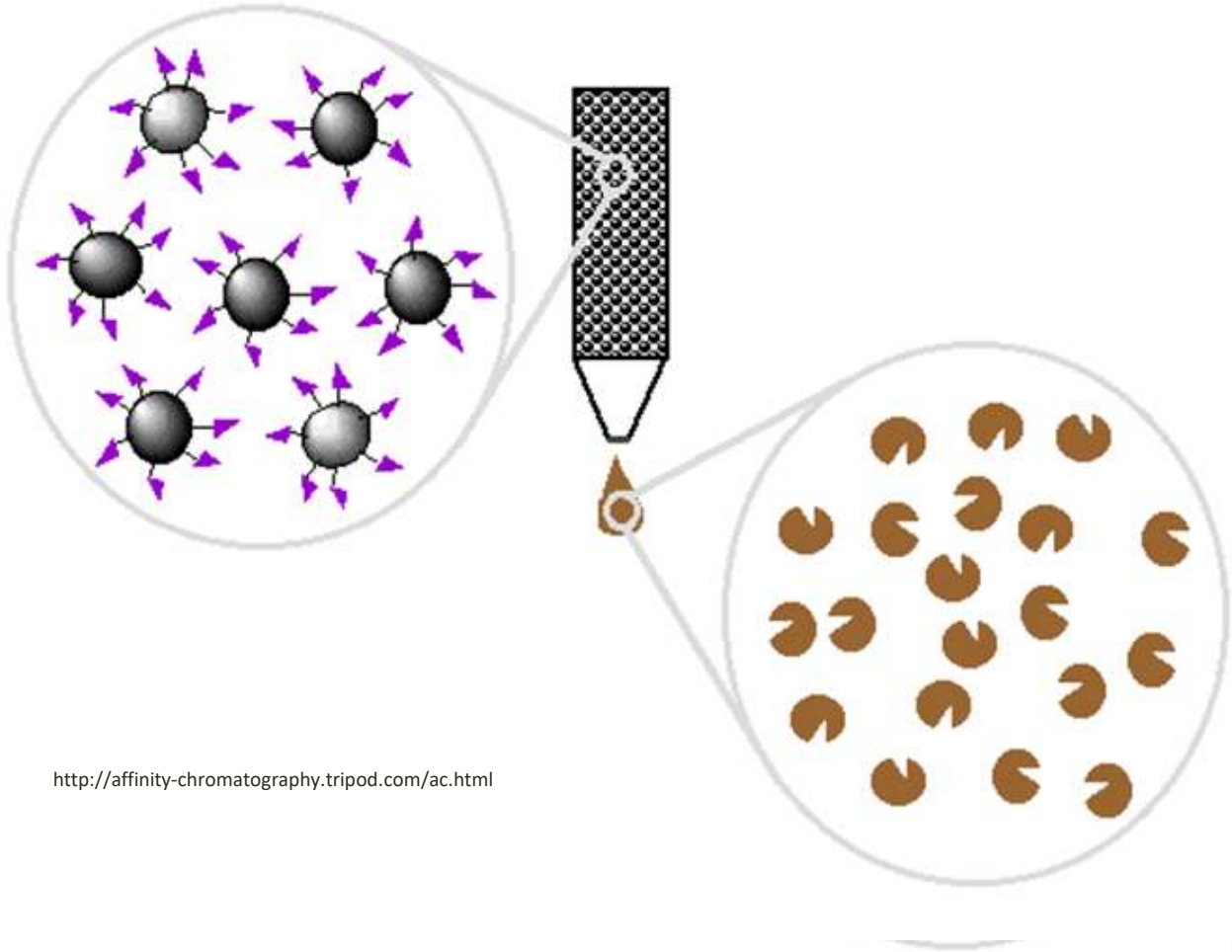


Figure 6. An affinity matrix binding to its target protein
A. The bead. **B.** The Spacer arm. **C.** The ligand. **D.** The target protein.

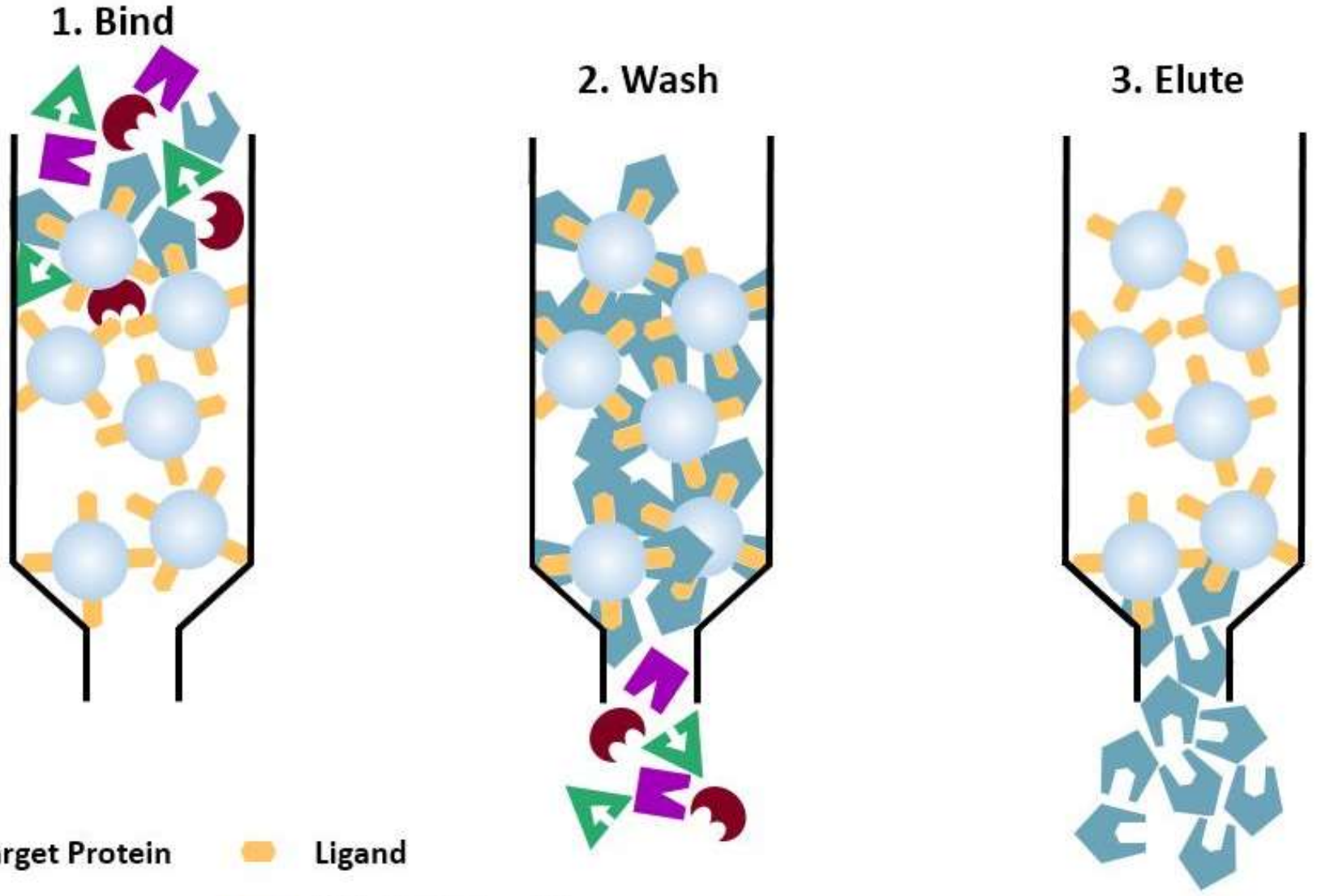
Experimental Procedures

Step 6. Elute proteins that bind tightly to ligand and collect purified protein of interest.



http://affinity-chromatography.tripod.com/ac.html

Protein Affinity Chromatography



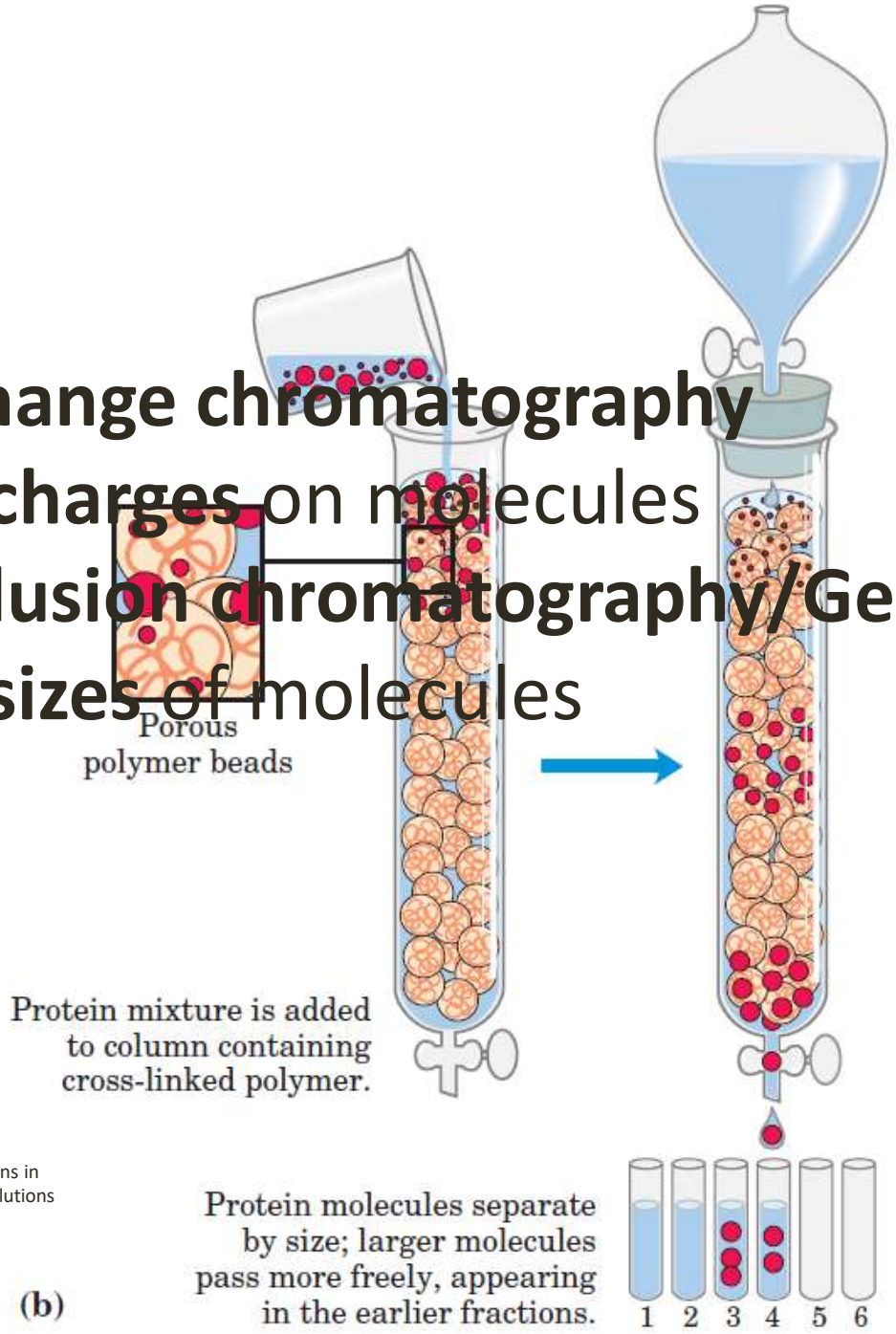
Other C ıethods

1. Ion-exchange chromatography

-based on **charges** on molecules

2. Size-exclusion chromatography/Gel filtration

-based on **sizes** of molecules



Marcell wolf (2015)「Effective interactions in liquid-liquid phase separated protein solutions induced by multivalent ions」