**Lec.10: *Bioseparation Technology***

***Gel Filtration Chromatography Media (Resins)*:**

**The choice of media depends on the properties of the components to be separated and other experimental factors. The following are general considerations when determining the choice of gel filtration chromatography media:**

* **Fractionation range**
* **Size exclusion limit**
* **Operating pressure**
* **Flow rate**
* **Sample viscosity**
* **pH range**
* **Operating temperature**
* ***Gel Filtration Resins:***
* **The gels currently in use are three types, Dextran, Agarose and polyacrylamide.**
* ***Dextran (e.g. Sephadex):***
* **- is a polysaccharide composed of glucose residues.**
* **- prepared with various degree of cross-linking controlled to yield a series of gels having different pore size.**
* **- supplied in the form of dry beads that swell when water is added.**
* **- it is mainly used for separation of small peptides and globular proteins.**
* **# Sephadex G-100 will separate molecules with molecular weight from 4,000 to 150,000 Da, so, those molecules which are with molecular weight larger than 150,000Da will be excluded from the beads, because of their size thy cannot get throw the pores of the beads, and elute first. Caution must be taken:**
* **It is important that the gel should be, homogenous, free from bubbles, free from cracks, free from spaces between the walls and it should covered by the liquid mobile all the time. The Gel Filtration is not recommended for separating protein with only a small difference in molecular weight. Good separation usually requires long columns and slow flow rate of mobile phase**

***Advantage of Gel Filtration*:**

**It is the best method for separation of molecules differing in molecular weight because:**

**1- It doesn't depend on temperature,pH, ionic strength and buffer composition, so, separation can be carried out under any conditions.**

**2- There is less zonal spreading than in other techniques.**

**3- The elution volume is related to the molecular weight.**

**4- Important method in protein purification.**

***Disadvantage of Gel Filtration:***

**1- Limited sample volume.**

**2- Poor selectivity compared with SDS-PAGE.**