ION-EXCHANGE CHROMATOGRAPHY

M.PRASAD NAIDU
Msc Medical Biochemistry,
Ph.D Research scholar.

introduction

- The process by which a mixture of similar charged ions can be separated by using an ion exchange resin
- Ion exchange resin exchanges ions according to their relative affinities.
- There is a reversible exchange or similar charged ions
- Mostly similar charged ions like cations or anions can be separated by this technique

Principle

- Reversible exchange of ions b/n the ions present in the solution and those present in the ion exchange resin.
- Cation exchange:
- Separation of cations
- Solid-H^+ + M^+ (solution) → Solid-M^+ + H^+ (solution)

The cations retained by the solid matrix of ion exchange resin can be eluted by using buffers of diff strength and hence separation of cations can be effected.

Anion exchange

- Separation of anions using
- Anion exchange resin
- Solid-OH^- + A^- → Solid-A^- + OH^-

  (solution) (solution)
- The anions retained by the solid matrix of ion exchange resin can be eluted by using buffers of diff strength

Classification of resins

- According to the chemical nature
- 1. Strong cation exchange resin 2. Weak cation exchange resin
- 3. Strong anion exchange resin 4. Weak anion exchange resin
- According to the source
- Natural: cation→ Zeolytes, clay etc
- anion → Dolomite
- Synthetic: inorganic & organic resins
- Organic resins are the most widely used
- Org ion exchange resins are polymeric resin matrix containing exchange sites.
- The resin is composed of polystyrene & Divinyl benzene, polystyrene contains sites for exchangeable functional groups
- Divinyl benzene acts as a cross linking agent & offers adequate strength i.e, mechanical stability

Functional groups present in different ion exchange resins

- Strong cation exchange resin → SO_3H
- Weak cation exchange resin→ COOH, OH, SH, PO_3H_2
- Strong anion exchange resin → N^+R_3, NR_2
- Weak anion exchange resin → NHR, NH_2

Common ion exchange resins

Class of resin	Nature	pH range	applications
Cation-strong	Sulfonated polystyrene	1-14	Fractionation of cations, inorganic separions, peptides, aminoacids, B vits
Cation weak	Carboxylic methacrylate	5-14	Fractionation of cations, biochemical separations, org bases, antibiotics

Common ion exchange resins

Class or resin	Nature	pH range	Applications
Anion – strong	Quaternary ammonium polystyrene	0-12	Fractionation of anions Alkaloids, vitamins, fattyacids
Anion-weak	Polyamine polystyrene or phenol H-CHO	0-9	Fractionation of anionic complexes, anions of diff valency vitamins, aminoacids

Structural types of ion exchange resins

- 1. Pellicular type with ion exchange film
- 2. porous resin coated with exchange beads
- 3. macroreticular resin bead
- 4. surface sulfonated and bonded electrostatically with anion exchanger

Structural types of ion exchange resins

- 1. Pellicular type with ion exchange film:
- The particles have a size of 30-40μ with 1-2μ film thickness.
- These have very low exchange capacity
- Ion exchange efficiency: 0.01 – 0.1 meq/g of ion exchange resin.
- 2. Porous resin coated with exchanger beads: size 5-10μ -
- They are totally porous & highly efficient
- Exchange capacities 0.5-2 meq/g or ion exchange resin

Structural types of ion exchange resins

- 3. macroreticular resin bead: A reticular network of the resin is seen superficially on the resin beads
- They are not highly efficient & have very low exchange capacities
- 4. surface sulfonated and bonded electrostatically with anion exchanger:
- The particles are sulfonated, & they are bonded electrostatically with anion exchanger resin.
- They are less efficient & have low exchange capacity
- Exchange capacity is 0.02meq/g of exchange resin.

Physical properties or resins

- 1. Particle size: 50-200 mesh fine powder,
- should allow free flow of mobile phase, should contain more exchangeable functional groups
- 2. Cross linking & swelling:
- When more cross linking agent is present, they are more rigid, but swells less
- When swelling is less, separation of ions of diff sizes is difficult as they can’t pass through the pores present.

Physical properties or resins

- When less cross linking agent is present, they are less rigid, but swell more
- Separation will not be efficient as exchange of functional groups does not take place due to wide pore
- Hence an optimum quantity of cross linking agent should be added to the polymeric ion exchange resin for the separation to be effective.

Practical requirements

- 1. column material & dimensions:
- Glass, stainless steel or polymers which are resistant to strong acids & alkalies
- Length: diameter ratio 20:1 to 100:1

Selection of ion exchange resin

- Depends upon
- 1. type of the ions to be separated – cations or anions
- 2. nature of the ions to be separated- strong or weak
- 3. efficiency of the resin: measured by ion exchange capacity
- Ion exchange capacity is the total ion exchange capacity in terms of the exchangeable functional groups expressed as meq/g of the ion exchange resin
- m.eq/g = 1000/eq.wt
- 4. particle size of the resin: 50-100 mesh or 100-200
- 5. structural type of the resin: porous, pellicular etc
- 6. Amount of cross linking agent present: which decides swelling of the resin.

Packing of the column

- Wet packing
- The resin is mixed with the mobile phase & packed in the column uniformly
- The sample to be separated is dissolved in the mobile phase and introduced all at once into the column.

Mobile phase

- Organic solvents are less useful & they are not used at all.
- Only diff strengths of acids, alkalies & buffers are used as eluting solvents
- Eg: 0.1N HCl, 1N NaOH, phosphate buffer acetate buffer, borate buffer, phthalate buffer .etc.,

Development of the chromatogram & elution

- 1. isocratic elution technique
- 2. gradient elution technique

Analysis of the elute

- Spectrophotometric method
- Polarographic method
- Conductometric method
- Amperometric method
- Flame photometric method
- Radiochemical methods (GM counter, ionization chamber method)

Regeneration of the ion exchange resin

- The ion exchange resin after separation may not be useful for next separation as exchangeable functional groups are lost
- But due to cost of the ion exchange resins they cannot be disposed off
- Hence reactivation, regeneration
- Regeneration makes the used ion exchange resin to be as efficient as a virgin resin.
- Regeneration: replacement of the exchangeable cations or anions present in the original resin
- Hence regeneration of the cation exchange resin is done by the charging the column with strong acid like HCl
- Vice versa

Factors affecting ion exchange separations

- 1. Nature & properties of ion exchange resins:- Cross linking & swelling
- 2. Nature of exchanging ions:
- A) Valency of ions: at low conc & ordinary temp , extent of exchange increases with increase in valency

$$Na^{+} < Ca^{2+} < Al^{3+} < Th^{4+}$$

Factors affecting ion exchange separations

- B) Size of ions: for similar charged ions, exchange increases with decrease in the size of hydrated ion.
- $Li < H^{+} < Na^{+} < NH^{4+} < K^{+} < Rb^{+} < Cs^{+}$
- C) Polarizability: exchange is preferred for greater polarizable ion
- Eg: $I^{-} < Br^{-} < Cl^{-} < F^{-}$
- D) conc of solution: in dilute solutions, polyvalent anions are generally adsorbed preferentially
- E) conc and charge of ions: if resin has higher +ve charge and solution has lower +ve charge , exchange is favoured at higher conc.
- If the resin has lower +ve charge and solution has high +ve charge , then exchange is favoured at low conc.

applications

- Softening of water
- Demineralisation or deionisation of water
- Purification of some solutions to be free from ionic impurities
- Separation of inorganic ions
- Organic separations: mixture of pharmaceutical compounds ca be separated
- Biochemical separations like isolation of drugs or metabolites from blood, urine etc
- Conc of ionic solutions

Thank you