Ion Chromatography (IC)

Purpose: This module provides an introduction to ***Ion Chromatography (IC).*** In this module the basic theory and applications of IC will be presented at a level that assumes a basic general chemistry background.

Learning Objectives: At the end of this content, students will be able to:

1. Explain the basic principles, operation and application of IC.
2. Explain the chemical basis for stationary phase effects and mobile phase effects.
3. Differentiate between stationary phases used in anion exchange and cation exchange.
4. Explain the basis for the common IC detection methods.
5. Describe the general process of analyzing a sample by IC.

Ion Chromatography (IC) is a useful tool for determining the presence and concentration of ions in samples and is utilized in numerous settings including environmental analyses such as the determination of anions (PO_4^{3-}, Cl^-, NO_3^-, etc) in surface waters. Current IC methods are often used to quantify concentrations in the low ppm level, depending upon the specific instrumentation used.

IC is a subset of liquid chromatography methods: *ion exchange*, *ion exclusion*, *ion pair* chromatography. IC methods were first reported around 1850 when H.Thomson and J.T. Way used various clays as an ion exchange and extracted labile calcium, magnesium, and ammonium ions. In 1927, the first zeolite column was used to remove Mg^{2+} and Ca^{2+} from water. Cation exchange using a sulfonated polystyrene/divinylbenzene column was developed in the 1940s as part of the Manhattan project. Very large columns were used to concentrate and purify the radioactive nucleotides required for the atom bomb. In the late 1940s anion exchange was performed with the attachment of a quaternary ammonia on the polystyrene/divinylbenzene support. (For additional description of the history see Small, H *J Chem Ed*, 2004, **81**(9), 1277-1284.)

Basic Principles:

Ion-exchange is the basic principle behind the removal of cations and anions from drinking water using most commercial, such as Brita®, water filters. Ion-exchange is also a natural process that occurs with clay substrates, resulting in the mobility of cations in soils.

Ion exchange columns used in chromatographic applications have a fixed ion that is covalently bound to a solid support. This solid support is packed inside the column, producing the stationary phase. The mobile phase that is pumped through the column in ion exchange chromatography uses water as the solvent. The fixed ion must have an oppositely charged counterion to balance the change. The figure below represents an ion exchange resin that would be used for the analysis of anions. The two ions are held in place by electrostatic forces. Central to the functioning of ion-exchange resins is that the counterion is exchangeable. In the figure below, the anion currently associating with the resin can be displaced by another anion that is in the aqueous mobile phase. The ion in the mobile phase that is used to displace anions from the resin is referred to as the eluent ion.

The basic process of chromatography using ion exchange can be represented in 5 steps: eluent loading, sample injection, separation of sample, elution of analyte A^-, and elution of analyte B^-, shown and explained below. Elution is the process where the ion of interest is moved through the column. Elution happens because the eluent ion is constantly pumped through the column. The chemical reactions below are for an anion exchange process.

Step 1: Run the eluent anion through the column to displace any other anions bonded to the resin. This loading step saturates the resin surface with only the eluent anion. Note, for reproducible chromatographic results, it is essential that all the ionic sites on the column are occupied by eluent ions before starting the analysis.

(key: Eluent ion = ▲, Ion A= ▫, Ion B = ◎)

The following reaction can be used to represent the eluent ion (E^-) displacing any bound anion (represented by X^-) that were initially associated with the resin:

$$Resin^+\text{-}X^- + E^- <=> Resin^+\text{-}E^- + X^-$$

Step 2: A small sample (often less than 100 µL) containing A^- and B^- is injected onto the column. This sample could contain many different ions, but for simplicity this example uses just two different ions.

Step 3: After the sample has been injected, the continued addition of eluent anion in the mobile phase causes A^- and B^- to move through the column by an ion-exchange process.

Q1. *Write the chemical reaction for the association of A^- and B^- with the ion exchange resin.*

Q2. *Write the chemical reaction for the elution of A^- and B^- from the ion exchange resin.*

Although not rigorously correct, a helpful way to examine the separation of A^- and B^- is to think of the reactions you wrote to answer Q2 as equilibrium processes.

Q3. *Write the equilibrium expression constant for A^- and B^-. In chromatographic separations, this term (Kc) is referred to as the distribution coefficient.*

Q4. *Do you think the magnitude of the distribution coefficients are the same for A^- and B^-? Why or why not?*

Q5. *If the distribution coefficient of A^- (▫) is smaller than the distribution coefficient of B^- (◎), draw a sketch of the elution process that is similar to the figure in Step 1 but at a point where A^- and B^- are partway through the column.?*

Step 4: As the eluent continues to be added, A^- eventually moves through the column in a band and ultimately elutes from the column.

Q6. *Suppose B^- had a very strong affinity for the resin, what would happen to its elution time?*

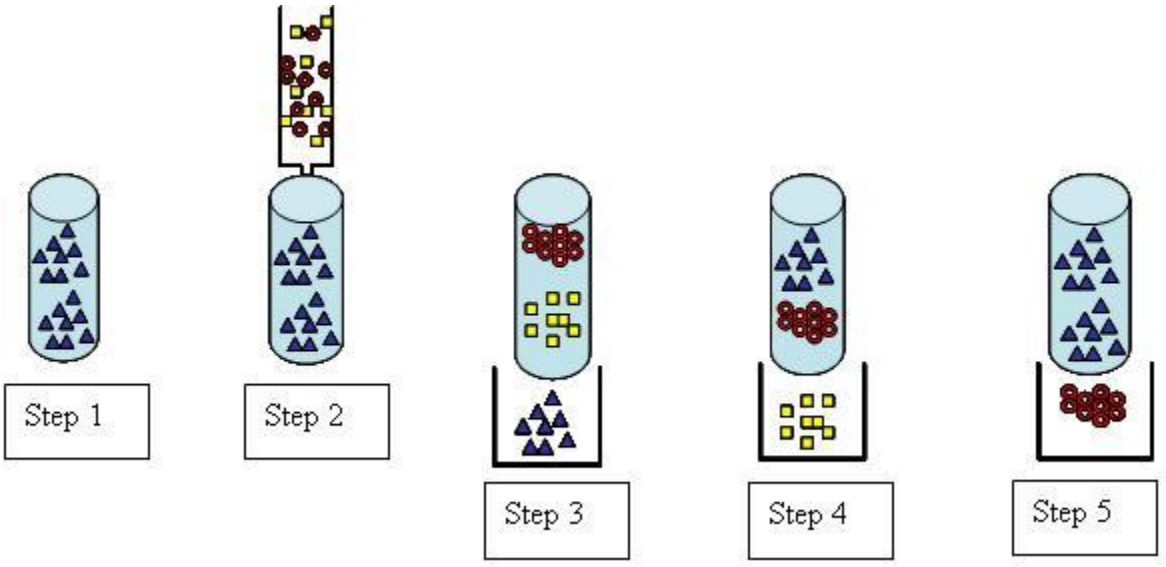
Q7. *Is this a desirable or undesirable situation if you were trying to analyze A^- and B^- in a mixture?*

Q8. *Is there a situation you can think of when it might be desirable for B^- to have a very strong affinity for the resin?*

Step 5: The eluent eventually causes the elution of B^- off the column.

$$Resin^+\text{-}B^- + E^- \Longleftrightarrow Resin^+\text{-}E^- + B^-$$

The overall 5 step process can be represented pictorially:



Stationary phase (or resin) composition

There are a number of different resins or stationary phases that have been developed for use in IC.

Q9. *If you want to separate cations, what would be different about the stationary and mobile phases?*

All of the resins used in ion exchange columns consist of very fine particles.

Q10 *Would water flow easily through a column containing very fine particles?*

Q11*. If not, how could you get the water through the column?*

Q12. *Considering that the column is packed with very fine particles, what must be done to surface water samples before injecting them onto the column?*

Detection Methods In order for an ion exchange separation to be useful information, you need detect the ions as they elute from the column.

Q13. *Can you think of a way to detect the presence of ionic substances in water?*

Q14. *Would this detection method distinguish between Na^+ and Ca^{2+}?*

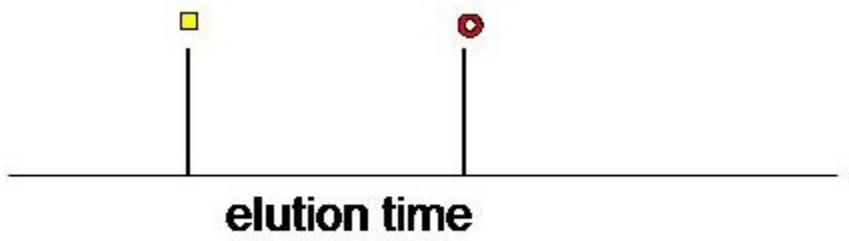
Q15. *If you utilize this detection method with the chromatographic separation, how important is the selective response of the detector?*

It is important to remember that the Na^+ and Ca^{2+} only move through the column because of the continuous presence of the eluent ion in the mobile phase.

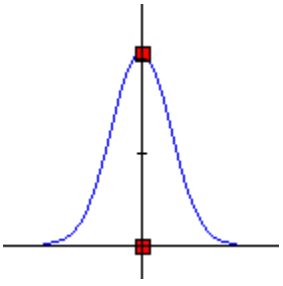
Q16. *Will the eluent ion respond to the detection method you thought of above to measure the presence of Na^+ and Ca^{2+}.*

We will not go into the details of how the process works, but modern ion chromatographic systems are designed in such a way that the eluent ion either undergoes a chemical reaction or is removed from the system after the column such that the only ionic substance left in solution is the analyte ion. This suppression of the signal from the eluent ion allows for the selective detection of only the analyte ions as they elute from the column.

The results from the detection give rise to a chromatogram, which shows the detectors response as a function of elution time. If all of the analyte came out at the same time the chromatogram would look like:



In reality, the peaks are actually broadened due to factors we will not discuss. Thus the peaks will look more like:



Q17. *How will the chromatogram change as you increase the concentration of A^- and B^- injected into the column? Make sure to label the axes of your chromatogram.*

Q18. *Since neither axis in the chromatogram you drew above is concentration, how can we calibrate the detector response to determine the concentration of A^- and B^-?*

You now have an outline of the basic ion chromatography process. The exact way the sample is loaded onto the column varies with the instrument. This process is often either a direct injection or using an inline automated sampling system. The sample is eluted off of the column, through the detector. The signal from the detector is then converted into the chromatograph. The peak areas then can be converted to a concentration using a calibration curve.