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Chromatography: Modus Operandi of Paramount Significance in Analytical Chemistry

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Separation is a process in chemistry, whereby the different components of a mixture are isolated leading to various individual fractions enriched with a particular component.[1] The process can be used either for recovery of precious materials or for determination of solutes at low concentration levels making it very attractive at both lab and industrial scales. The differences in the physical (volatility, melting point, size, shape, mass, density) or chemical (ion exchange, complexation) properties of the components are exploited to achieve separation. Techniques like distillation, filtration etc that are dependent on physical properties are less selective than techniques like chromatography, ion exchange etc which are dependent on chemical properties of the solutes. Chromatography has a great bearing on analysis of a wide range of samples. This technique originated in the late nineteenth and early twentieth centuries from independent research studies of David T. Day (American geologist) and Mikhail Tsvet (Russian botanist) on petroleum fractionation using Fuller's earth, and leaf pigments separation on chalk column, respectively.[2] Tsvet extended his knowledge of extraction using filter paper to column method which was developed by Day. Tsvet demonstrated the separation of yellow, orange, and green plant pigments (identified later as xanthophylls, carotenes, and chlorophylls respectively) using a adsorption column containing calcium carbonate. It was Tsvet who coined the word chromatography using the Greek words of "*chroma*" and "*graphy*" meaning *colour* and *recording* respectively. It is also known in literature that his surname in Russian meant colour indicating maybe this was added reason for the term to be coined. Irrespective of the reason, Tsvet not only coined the term but also explained the concepts leading to him being rightly called as the Father of Chromatography. However, it was later in 1940s that chromatography became popular. This was due to the development of partition chromatography by Archer Martin and Richard Laurence Millington

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Synge. This technique was an amalgamation of chromatography and counter-current solvent extraction and could separate solutes with very small differences in their partition coefficients between two liquid solvents. Martin and Synge also used filter paper as SP leading to discovery of paper chromatography. Martin, in alliance with Anthony T. James developed gas chromatography in 1949. The extensive use of ion exchangers was seen in the Manhattan project during World War II but the original concept of separation using ion exchangers was conceived by Sir Thompson and J T Way. It was around 1975 that ion chromatography became commercial due to the extensive work of William Bauman, Hamish Small and Tim Stevens. Today, chromatography refers to diverse separation techniques based on a series of partitioning or distribution of a sample between two phases, one of which is moving (Mobile phase MP) and other is immobile (stationary phase SP). MP can be gas, liquid or a supercritical fluid (SFC) while SP is a solid or liquid within a solid support. There are two types of chromatography, namely adsorption and partition. Both these types involve the separation due to differences in solubility between the SP and MP. Adsorption chromatography involves the direct binding of solute to sites on the surface of SP and the difference in binding leads to separation. MP can be liquid, gas or supercritical fluid. In partition chromatography the solutes are separated between the MP (liquid only) and the liquid within the solid support. Though the scope of expansion on adsorption chromatography has stalled, partition chromatography became the basis for expansion of chromatographic techniques. The chromatographic techniques can also be categorized based on the SP geometry into planar and column. Both the techniques follow the same principle of separation. Techniques like paper and thin-layer chromatography (TLC) that use paper / plate respectively are examples of planar chromatography as the SP has planar geometry. MP used in these studies are polar solvents and are run against gravity. The advantages of planar chromatography are speed, low solvent inventory, very small sample volume and is found suitable for analytical determinations. Techniques which involve the use of SP packed inside the volume of a column or on the walls of column (HPLC, GC) are termed as column chromatography. MP can be polar or non polar based on the SP used. Non polar chromatography involves a non-polar MP and a polar SP and the reversed combination is used in reversed phase liquid chromatography. Column chromatography can be used for both analytical as well as for preparative studies but the analysis time, solvent/sample inventory are more than that of planar chromatography. Reversed phase chromatography is carried out using columns filled with very small size particles of SP and therefore the MP is pumped under the pressure through it. Therefore it is also known as reversed phase high pressure liquid chromatography(RPHPLC). It can be used for analysis of organic and inorganic molecules.[3-5] It is seen that even metal ions can be separated using a non polar SP by as its organic complex.[3] Ion chromatography (IC) is column chromatography used exclusively for inorganic ions. In this the SP is an ion exchanger while the MP is aqueous solution of appropriate eluent. IC finds extensive applications in analysis of samples pertaining different industries including nuclear industry[4,5] due to its ease of analysis, excellent detection selectivity and sensitivity, wide dynamic range.

Chromatography is a simple and exceedingly flexible technique which will expand and lead to new variations for wide range of applications.

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