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Paper Chromatography

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Abstract: *A Real estate private prediction model leveraging machine learning to estimate property values. It utilizes historical data and various features to build and train predictive models, including linear regression and tree-based algorithms. real estate price prediction is the process of estimating or forecasting the future prices of real estate properties, such as houses, apartments, or commercial buildings. The goal is to provide accurate property rates to buyers, sellers, investors, and real estate professionals to make informed decisions about real estate transactions.*

Keywords: Geo-Spatial Analysis, Machine Learning, Deep Learning, Building Age, Property Type, Number of Rooms, Python

I. INTRODUCTION

The technique of paper chromatography was first discovered by Synge and Martin in 1943. Paper Chromatography is specific type of technique that operates on a specific piece of paper. It is type of planar chromatography, in which separation of compounds is performed using a filter paper made up of cellulose which acts as a stationary phase. The method is comparatively cheap and helps to separate dissolved chemical substance by their different rates of migration through paper sheets. The method requires very minute quantity of sample for analysis.Paper chromatography definition explains that is an inexpensive and powerful analytical technique, which requires a piece of paper or strips serving as an adsorbent in the stationary phase across which a particular solution is allowed material For the separation of dissolved chemical substances and lipid samples (in particular), paper chromatography is found to be very trustable. This analytical tool employs very few quantities of material. A paper chromatography method, two dimensional chromatography involves using two solvents and spinning 90 ° of paper in between. This is useful for separating complex mixture of compounds with similar polarity. Paper chromatography is a useful technique because it is relatively simple and requires only a small amount of material. Separations in paper chromatography are based on the same concepts as those in thin-layer chromatography, as is the form of thin-layer chromatography.

The method consists of applying the test solution or sample as a spot near one corner of a sheet of filter paper. The paper is initially impregnated with some suitable solvent to create a stationary liquid Phase. An edge of the paper close to the test spot is then Immersed in another solvent in which the components of the mixture are soluble in varying degrees. The solvent penetrates the paper by capillary action and, in passing over the sample spot, carries along with it the various components of the sample. The components move with the flowing solvent at velocities that are dependent on their solubilities in the stationary and flowing solvents. Separation of the components is brought about if there are differences in their relative solubilities in the two solvents. Before the flowing solvent reaches the farther edge of the paper, both solvents are evaporated, and the location of the separated components is identified, usually by application of reagents that form coloured compounds with the separated substances. The separated components appear as individual spots on the path of the solvent. If the solvent flowing in one direction is not able to separate all the components.

Principle of Paper chromatography

Paper chromatography is a form of liquid chromatography where the basic principle involved can be either partition chromatography or adsorption chromatography. In paper chromatography separation of component is distributed between phases of liquid. Here, one phase of liquid is water that is held amidst the pores of filter paper and the other liquid is the mobile phase that travels along with the filter paper. Separation of the mixture is the result that is obtained from the differences in the affinities towards the water and mobile phase when travelling under capillary action between the pores of the paper Though in a majority of paper chromatography applications, the principle is based on partition

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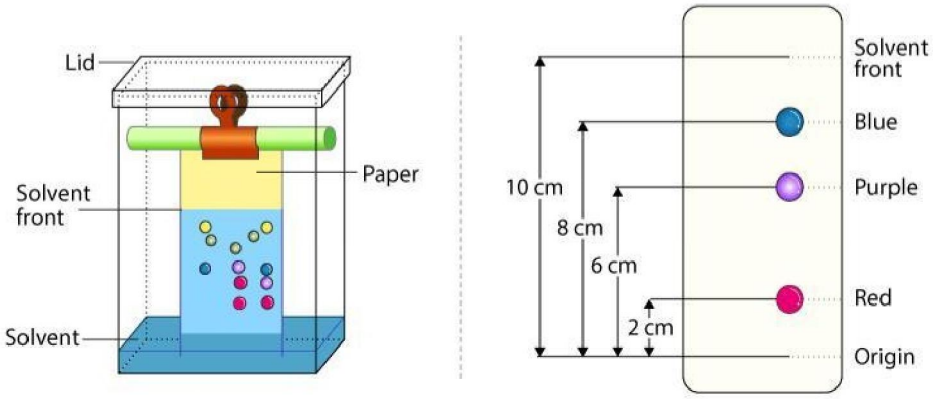
chromatography but sometimes, adsorption chromatography can take place where the stationary phase is the solid surface of the paper and the mobile phase is the liquid phase.



• This technique is a type of partition chromatography in which the substances are distributed between two liquids, i.e., one is the stationary liquid (usually water) which is held in the fibers of the paper and called the stationary phase, the other is the moving liquid is the moving liquid or developing solvent and called the moving phase.
• Cellulose filter paper is often used as the stationary phase in paper chromatography.
• Since it is hydrophilic, it is usually covered with thin film of water.
• The procedure is often regarded as liquid-liquid chromatography.
• The components of the separated migrate at different rates and appear as spots at different points on the paper.
• In this technique, a drop of the test solution is applied as a small spot on a filter paper and the spot is dried.
• The paper is kept in a close chamber and the edge of the filter paper is dipped into a solvent called developing solvent.
• As soon as the filter paper gets the liquid through its capillary axis and when it reaches the spot of the test solution (a mixture of two or more substances), the various substances are moved by solvent system at various speeds.
• When the solvent has moved these substances to a suitable height (15-18 cm) the paper is dried and various spots are visualized by suitable reagent called visualizing agent.
• The movements of substances relative to the solvent are expressed in terms of Rf values (retardation factor or retention factor).
• Rf: The R is related to the migration of the solute front relative to the solvent front as:
• R is a function of the partition coefficient. For a given substance it is constant provided the conditions of chromatographic system are kept constant with respect to temperature, type pf paper, duration and direction of development, nature and the shape and the size of the wick used (i.e., radial chromatography), the amount of liquid in the reservoir humidity, etc.
• The Rf defines the movement of the substances relative to the solvent front in a given chromatographic system.
• The Rf value of a substance depends upon a number of factors which are:
o The solvent employed
o The medium used for separation, i.e., the quality of the paper in case of paper chromatography
o The nature of the mixture
o The temperature
o The size of the vessel in which the operation is carried out.

Stationary phase:
o It is liquid that is, the water trapped in the molecular structure of the paper and is invisible.
o Supporting material for the stationary phases is the matrix of cellulose fibers of chromatography paper.
o Chromatography papers are available in three running characteristics: slow, medium, and fast.
o Most frequently used chromatographic paper is Whatmann No.1 or its equivalent.

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Mobile phase:

o It is generally a liquid e.g. for a pure paper system, the eluent may be a single solvent or a mixture of solvents that can move through the paper.

o Among mixed solvent systems, a water-organic mixture is frequently used, e.g. n- butanol, acetic acid: water (4:1:5, top layer) for flavonoid, glycosides, acetic acid: conc.HCl: water (30:3:10) (Forrestal system for flavonoid aglycones), toluene:acetic acid: water (4:1:5, upper phase for flavonoid aglycones).

o The solvents used are selected from eluotropic series which is a list of solvents arranged in order of increasing polarity.

o Petroleum ether < n-hexane< carbon tetrachloride< toluene < benzene < chloroform < dichloromethane < diethyl ether < n-butanol < isopropanol < acetone < ethanol < methanol < water

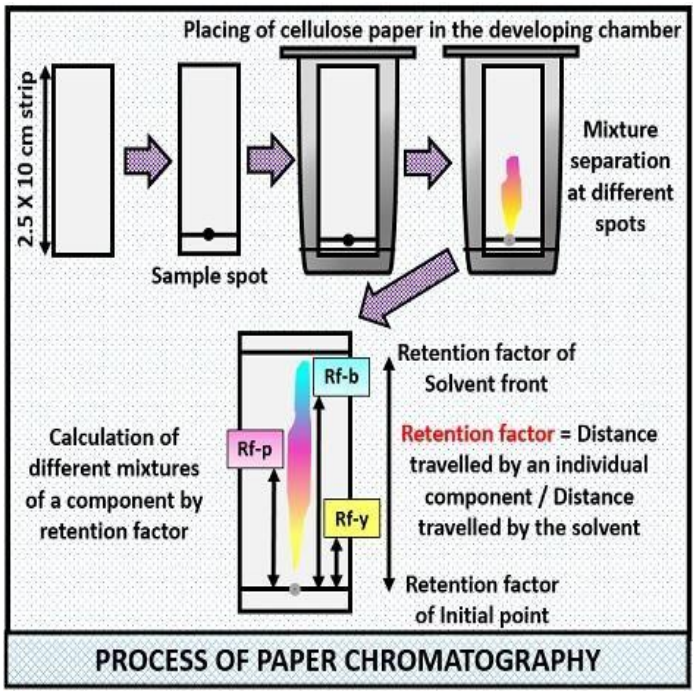
o Generally, higher the solubility of a solute in a solvent, the greater the solute mobility in that solvent.

o If a solute dissolve more readily in the mobile phase, then it will travel with the solvent, hence the partition occurs between two phases.

o Different solutes travel at different rates up the paper, it is a result of their different solubility in two phases.

Methodology of Paper Chromatography

- Selection of the ideal type of development
- Selection of filter paper
- Sample preparation
- Sample loading or spotting on the paper
- Chromatogram papar
- Drying of papar detection of the compound



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1. Selection of the ideal type of development

Based on factors such as the complexity of the solvent, mixture, paper, etc. the development type is chosen. Mostly either Radial or Ascending type of paper chromatography is employed because of the easiness they offer while handling and performing which ultimately leads to obtaining the chromatogram faster within a shorter duration of time.

2. Selection of filter paper

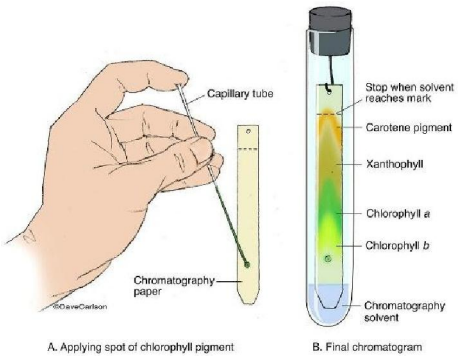
As per the pores' size and the sample quality.

3. Sample preparation

This involves the dissolution of the sample in an ideal solvent that is being utilized in developing the mobile phase.

4. Sample loading or spotting on the paper

With the help of a capillary tube, micropipette, the sample is spotted on the paper at an accurate position. This promotes the interpretation of the chromatogram more quickly and easily.



A. Applying spot of chlorophyll pigment B. Final chromatogram

5. Chromatogram paper

This is carried by the paper immersion in the mobile phase. The mobile phase crosses over the sample on the paper because of the capillary action of the paper.

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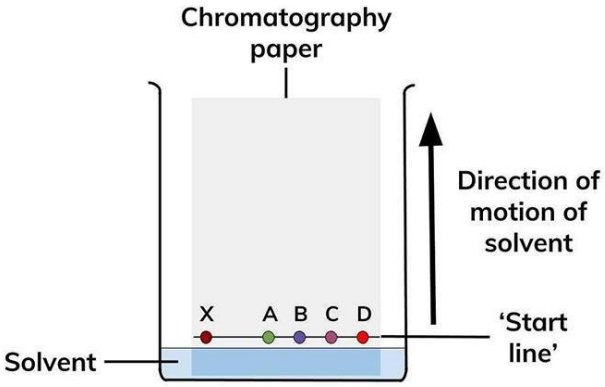


6. Drying of paper detection of the compound

With the aid of air drier, the paper is dried as soon as the chromatogram is developed. On the chromatogram developed paper, the detecting solution is sprayed and dried thoroughly for the identification of the sample chromatogram spots.

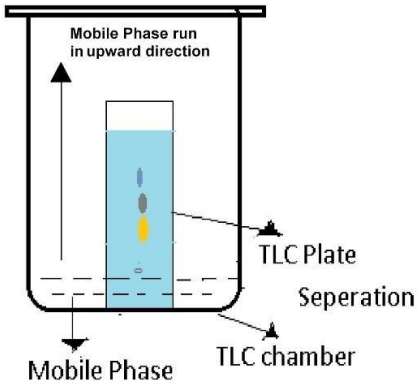
Types of Paper Chromatography

- Ascending Paper Chromatography
- Descending Paper Chromatography
- Ascending – Descending Paper Chromatography
- Horizontal or circular Paper Chromatography
- Two Dimensional Paper Chromatography

Ascending Paper Chromatography

- As per the name, the developing solvent is found to be moving in an upward direction. Here, a sufficient quantity of mobile phase is poured into the development chamber.
- Sample and reference are spotted on the line drawn a few centimetres from the bottom edge of the paper suspended from a hook or clip at the top.When the development of the paper is done by allowing the solvent to travel up the paper, it is known as ascending technique.
- Both ascending and descending techniques have been employed for separation of organic and inorganic substances.
- But the ascending technique is preferred if the Rf values of various constituents are almost same.

Ascending Chromatography



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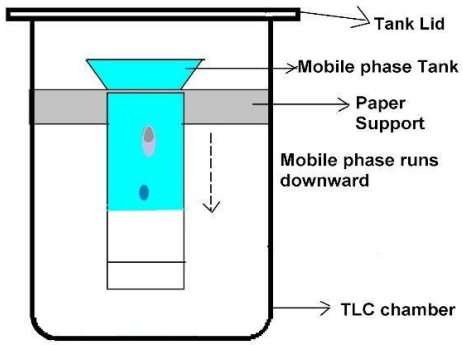
Descending Paper Chromatography

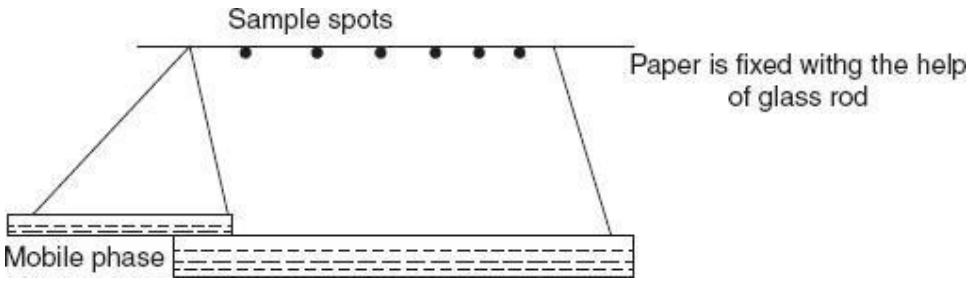
• Here, the solvent front travels down the length of paper suspended from the top inside the developing chamber.
• The mobile phase is kept in a trough in the upper chamber.
• The paper with spotting on the line drawn a few centimetres from the top is clamped to the top.
• Before elution, the jar is covered and equilibrated with the mobile phase vapour.when the development of the paper is done by allowing the solvent to travel down the paper, it is known as descending technique.

Descending Chromatograpphy



Ascending – Descending Paper Chromatography

• It is a mixed type of chromatography where the solvent first travels upwards on the paper that is folded over a rod and after crossing the rod it moves downwards.
• It is the hybrid of the above two techniques.
• In this technique, the upper part of the ascending chromatography can be folded over a glass rod allowing the ascending development to change over into the descending after crossing the glass rod.



Horizontal or circular Paper Chromatography

• This is also known as circular paper chromatography.
• This makes use of radial development.
• In this technique a circular filter paper is employed.
• Then the various materials to be analyzed are placed at the center.
• After drying the spot the paper is fixed horizontally on the petri-dish possessing the solvent so that the tongue or the wick.
• When solvent front has moved through a sufficient large distance, the components get separated in the form of concentric circular zones.
• This allows the separation of sample components in the form of concentric circular zones through the radial movement of the liquid phase.

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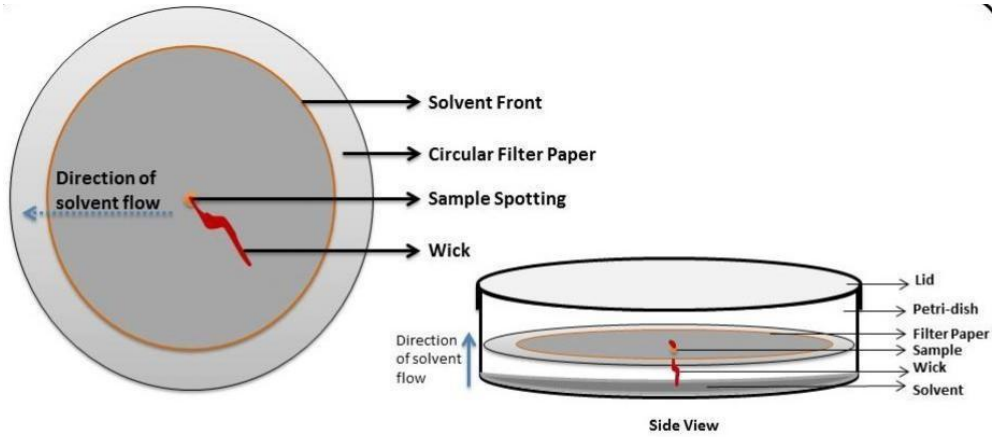
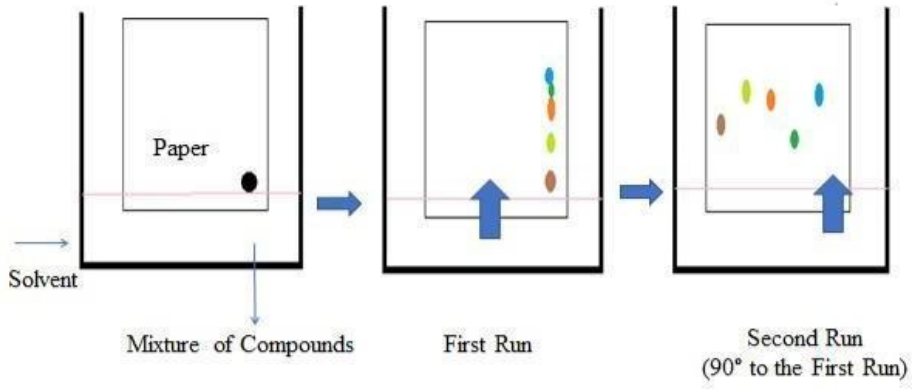


Two Dimensional Paper Chromatography

- In this, a square or rectangular paper is used.
- The sample is applied to one of the corners.
- The second development is performed at right angle to the direction of the first run.
- This type of chromatography can be carried out with identical solvent systems in both the directions or by two solvent systems.
- This helps in resolving substances that have similar Rf values.
- Where, Retardation factor (Rf) = The distance travelled by the solute/ distance travelled by the solvent front



Advantages of Paper Chromatography

- Itrequires fever quantitative material.
- Separation of compounds in a short time.
- Analysis requires a low amount of sample.
- Compare to other chromatography methods paper chromatography is a cheap technique.
- Organic as well as inorganic compounds can be identified by paper chromatography method.
- The setup of paper chromatography occupies less space than the other chromatographic method.
- Easy to handle and setup.
- The less sample quantity required for the analysis.
- Cost-effective method.

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Disadvantages of Paper Chromatography

- Volatile substances cannot be separated using paper chromatography techniques.
- Paper chromatography cannot be compatible with large amounts of sample.
- Quantitative analysis is not useful in paper chromatography.
- Paper chromatography cannot be separated complex mixture.
- As compared to the HPLC, TLC or HPTLC, paper chromatography has less accuracy.
- Data cannot be saved for long periods .

Applications of Paper Chromatography

1. Qualitative Analysis

- Involves the identification of compounds present in the mixture.
- Identification involves the use of Rf value based on Rf of standard compound.

2. Quantitative Analysis

- It is done in the paper or after the removal of the component from the paper.
- The latter is generally preferred – the component is cut from the paper, extracted by a suitable solvent, measured by colorimeter or UV-Vis spectrophotometer.
- Alternatively, the extracted solution is evaporated in the vacuum to remove solvent,Thus, obtained residue is weighed.

3. Preparative Paper Chromatography

- Operates with large amount (gram quantity) of substances to yield substances enough for further work in the laboratory.
- Practically, it is done in Whatman No.3 paper.
- The sample is streaked.
- The separated bands are cut, extracted with suitable solvent and filtered.
- The filtrate is evaporated off in vacuum to yield the residue of the component.

Typical Uses of Paper Chromatography

Seperating Coloured Pigments

- An effective technique used for separating colored pigments from a mixture. How does it work?
- A few drops of the colored pigments mixture are placed on the filter paper and then it is slowly submerged into a jar of solvent. Depending on their polarity, it dissolves the molecules present in the mixture, as the solvent rises up the paper. As the solvent continues to rise up the filter paper, molecules of each pigment leaves the solution at different places because of different polarity. Hence, every pigment rises up to a particular level on the chromatography paper and gets seperated in the process. Useful in the seperation of plant pigments.

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Reaction Monitoring

• Over a period of time, the concentration of reactants decreases, whereas the concentration of production increases in a chemical reaction. One can get a fair idea on the progress of reaction by spotting the reactants and developing the chromatogram over different time intervals. The availability of densitometers made quantitative estimations possible, but traditionally the technique was used for qualitative monitoring. However, as a reaction monitoring option, the rapid methods using spectroscopic techniques are limiting the paper chromatography application.

Isolation and Purification

• For components of mixture, paper chromatography has been put to use as a purification and isolation technique. Using spectro-photometric methods, the separated components on the paper are cut, dissolved in suitable solvents and their absorption is characterized at specific wavelengths.

Pathology and Forensic Science

• For investigation of crimes, paper chromatography is useful in the field of forensic science, as this process can be successfully carried out with even very small quantities of material. Using this technique, samples from crime scenes are collected to be analysed and identified.Used in DNA and RNA fingerprinting. Moreover, to detect the presence of alcohol or chemicals in blood, pathological laboratories use paper chromatography.

Foods

• Both natural and synthetic food colors are added to foods to improve their acceptability and to make them more popular. Paper chromatography has been primarily used for analysis of food colors in ice creams, sweets, drinks and beverages, jams and jellies. To ensure that no non-permitted coloring agents are added to the foods, only edible colors are permitted for use. That's how quantification and identification becomes more important.

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Analyzing complex mixture

• Certain organic compounds such as carbohydrates and amino acids are identified or detected from a complex mixture of organic compounds with the help of paper chromatography. It is useful in the separation of anions and amino acids. Ranging from forensic investigations, pharmaceuticals, environmental monitoring and foods, paper chromatography have retained their ground in various fields. For matching the requirements of high throughput laboratories, chromatography has seen phenomenal growth in terms of software's, applications and increased automation and separation in general.

II. CONCLUSION

Paper chromatography is a useful technique in the separation and identification of differentplant pigments. In this technique, the mixture containing the pigments to be separated is firstapplied as a spot or a line to the paper about 1cm from the bottom edge of the paper. Solventis absorbed by the paper and moves up the paper by capillary action. As the solvent crossesthe area containing plant pigment extract, the pigments dissolve in and move with the solvent.The solvent carries the dissolved pigments as it moves up the paper. The pigments are carriedalong at different rates because they are not equally soluble.

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