Chapter:
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AIM: TO CALCULATE THE RF VALUE OF GIVEN SAMPLE

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Materials Required

- A thin- paper of 'Aluminium' sheet.
- Thin layer object with costing of adsorbent material.

Theory

TLC is a tool for analyzing mixtures, by separating the mixture's constituent chemicals. TLC has the potential to identify compounds, establish their identities, and ascertain the purity of a chemical. It may also be used to trail the development of a reaction by keeping an eye on the arrival of a product or the disappearance of a reactant. TLC is a quick and sensitive method that can examine microgram (0.000001 g) amounts in a short amount of time (about 5-10 minutes). Three phases make up TLC: spotting, development, and visualizing. To create an extremely diluted (approximately 1%) solution, the sample to be studied is first dissolved in a volatile (easily evaporated) solvent.

A competition is set up between the silica gel plate and the development solvent for the spotted material. The outcome depends upon a balance among three polarities - that of the plate, the development solvent and the spot material.

Calculating the RF value of a compound

The relationship between the distance traveled by the solvent front and the compound is usually expressed as ;

The Rf value:

R_f value = distance moved by compound ÷ distance moved by solvent front.

Running the two samples side by side on the same TLC plate, ideally at the same concentration, is important to evaluate whether an unknown chemical and a compound with known

structure are same. Rf values are characteristically greater for low polarity chemicals compared to higher polarity molecules.

Principle

The relative affinity of chemicals for the two phases is what drives the separation. The substances in the mobile phase pass across the stationary phase's surface. The compounds that have a stronger attraction for the stationary phase move slowly whereas the other compounds move quickly during the movement. As a result, the mixture is successfully detached. After the separation procedure is complete, the mixture's constituent components show up as spots at the adequate levels on the screens. Suitable detecting techniques are used to determine their type and character.

Procedure

a. Start by drawing a line with a pencil approximately 1/4 inch from the plate's bottom. Spot locations for each position along the line (pure reference mark and sample).

b. Quickly and gently place a spot on the plate in the location you've marked by dipping the capillary into the solution. The spots should be tiny.

c. Place a piece of filter paper in a screw-cap jar, add approximately 3 mL of solvent, and wet the paper with the solvent to saturate the air. In order for the solvent to be below the spot line on your plate, it must be shallow enough.

d. Cautiously place the plate into the chamber using tongs or tweezers, taking care not to submerge the areas in the solvent. Place the jar's lid on it and wait for the plate to emerge.

e. Remove the TLC plate when the solvent front is almost at the top and mark the solvent front with a pencil line right away.

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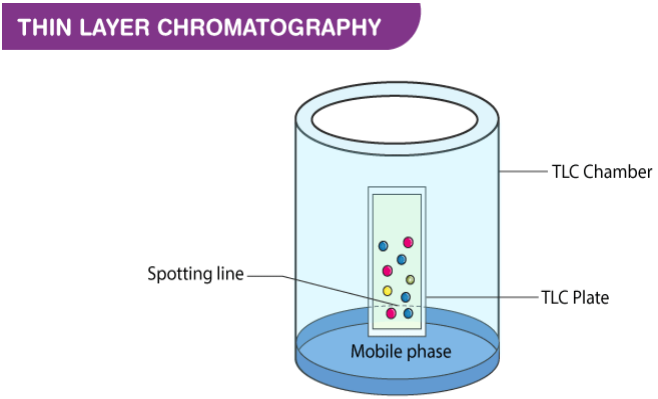
f. In a fume hood, dip the TLC plate using tweezers into the PAA stain solution, smoothly dip the plate in one motion.

g. Remove any extra stain from the back of the plate (the glass portion) and heat it for about 20 seconds. Examine the areas after letting the solvent drain.

h. Draw pencil rings around the places and use a ruler to get the Rf values.

Diagram



Applications

- TLC does qualitative testing on a variety of drugs, including sedatives, local anaesthetics, anticonvulsant tranquillizers, analgesics, antihistamines, steroids, and many other categories of medicines.
- Separating or isolating biochemical metabolites from their blood plasma, urine, serum, etc. is one use of TLC that is highly valuable in biochemical analysis.
- Natural materials companies used it for such volatile oils, alkaloids, glycosides, waxes, essential oils as well as

cosmetics industry employs it for purification and characterization.

- It is frequently labouring to separate complex medicinal compositions.
- Whether a reaction is completed or not, it is utilised**.**

Advantages of Thin Layer Chromatography

Thin layer chromatography has the following benefits:

- It is a quick and easy method.
- It facilitates the simple display of separated compound locations.
- The technique aids in locating the distinct chemicals.
- It aids in the isolation of the majority of compounds.
- The separation process moves more quickly, and the chemicals are more selective (even small differences in chemistry is enough for clear separation).
- It is simple to evaluate the purity standards of the provided sample.
- It is a less expensive chromatographic method.

Disadvantages of Thin Layer Chromatography

- Thin layer chromatography plates lack a longer stationary phase, which is one of its drawbacks.
- The separation time is constrained as compared to other chromatographic methods.
- TLC produces outcomes that are challenging to duplicate.

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- Because of the high detection limit, TLC cannot be used to reduce the detection limit.
- It is a qualitative analysis method; it is not a quantitative analysis method.

Result: The R_F value was determined to be……..