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# Some Useful and Practical Tips for Flash Chromatography

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Original procedure: W. Clark Still, *JOC* **1978**, *43*, 2923.

Extensions: *JCE* **1984**, *61*, 645.
*JCE* **1984**, *61*, 649.
*JCE* **1986**, *63*, 361.
*JCE* **1988**, *65*, 459.
*JCE* **1991**, *68*, 790.
*JCE* **1992**, *69*, 939.
*JCE* **1997**, *74*, 1223.
*JCE* **2001**, *78*, 363.
*JCE* **2004**, *81*, 109.

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## 1. Selection of the eluent system:

Arrange the elution strength of the AcOEt / petrol ether system to place the desired compound at $R_f = 0.5$ for best evaluation of $\Delta R_f$.

If less than 5% of AcOEt is required for $R_f = 0.5$ :
switch to less polar solvent as $Et_2O$ or $CH_2Cl_2$

If more than 50% of AcOEt is required for $R_f = 0.5$ :
switch to more polar solvent as THF or aceton

If $\Delta R_f < 0.1$ try 5-100% $CH_2Cl_2$ or 5-30% MeOH in petrol ether

Once the best solvent system is determined ($\Delta R_f$ is maximum), reduce the amount of the polar solvent to get $R_f = 0.2$ (difficult separation, $\Delta R_f < 0.1$) to $R_f = 0.35$ (easy separation, $\Delta R_f > 0.3$) to run the chromatography.

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## 2. Mass of silica gel:

Difficult separation ($\Delta R_f < 0.1$): 80 times the mass of the sample and never more

Easy separation ($\Delta R_f > 0.3$): 20 times the mass of the sample (less is a filtration)

Density of dry 40-63 μm silica gel used for FC is about 0.50

## 3. Height/diameter ratio:

Difficult separation ($\Delta R_f < 0.1$): H/D = 8

Easy separation ($\Delta R_f > 0.3$): H/D = 3

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## 4. Column packing:

- Jam a plug of cotton in the column and cover it with about 1 cm of sand.
- The desired amount of silica gel is introduced in the column as a slurry in the eluent, and pressure is applied to place the solvent head a few millimeters above the bed of silica gel, and 0.5-1 cm of sand is added.
- Ideally, the sample is then added to the top of the adsorbent bed in the minimum amount of the eluent. If the sample is only partially soluble in the eluent, the minimum amount of the more polar solvent of the eluent is added to dissolve the sample. In no case the height of the sample solution should exceed 1 cm over the adsorbent bed.
- The sample solution is adsorbed on the top of the gel column (do not dry the gel!). The walls are washed with a few milliliters of eluent, which are also pushed into the gel before the column is carefully filled with the eluent.
- Very polar impurities, generally insoluble in the eluent, are best removed by silica gel filtration prior chromatography.
- Columns without fritted glass are preferred since they have significantly less dead volume.
- This method is prefered to dry packing, essentially as it avoids powdered silica gel (toxic) dispersion.

## 5. Eluent flow rate

The eluent flow rate should be relatively rapid and increase with the height/diameter ratio; the solvent head should drop **5-7 cm per minute**. For higher viscosity solvents (cyclohexane, dioxane, i-propanol, ethanol…), the flow rate should be reduced. The pressure is controlled by a screwed needle valve or alternatively, the air leak can be realized by simply sticking a 16-18G short needle directly in the Tygon tubing connected to the laboratory air line. As much as possible, the required total amount of solvent to run the chromatography should be introduced in one time (use a solvent tank) to avoid deleterious pressure variations.

## 6. Fraction size

The fraction size should be about 80-100% the volume of silica gel used. Slightly smaller fractions should be used for easy separations. In most cases, the desired product is in the fractions 4-7 (TLC analysis), and a maximum of twelve fractions (generally eight to ten) should be collected. However, make sure all the product is out before drying the column. As noted by Still, the time required to elute the desired component from the column is generally so fast that a simple rack of tube or erlenmeyer is preferred to an automatic collector.

**Warnings:**

It sometimes happens that the $R_f$ observed on TLC is not transposable to the preparative column. This can ultimately result in inversion in the order of elution of the products (the less labile product on TLC is the first out the preparative column). This behavior is due to the differences in the silica gel used for TLC and flash chromatography. Also, the above protocol cannot be applied to deactivated silica gel (for example containing 2.5% v/v $NEt_3$).

**Conclusion:**

If you need more than 15 min. overall to run an efficient FC, you may need to read again this slideshow…