

## 7.10: Reverse Phase Chromatography

Silica and alumina are not the only possible solid phases. Stationary phases can be purchased that have long carbon chains bonded to silica beads. For example, a C18 column contains beads that have 18-carbon chains attached to them. These stationary phases are powders, like silica, and they can be loaded into a column just like silica can.

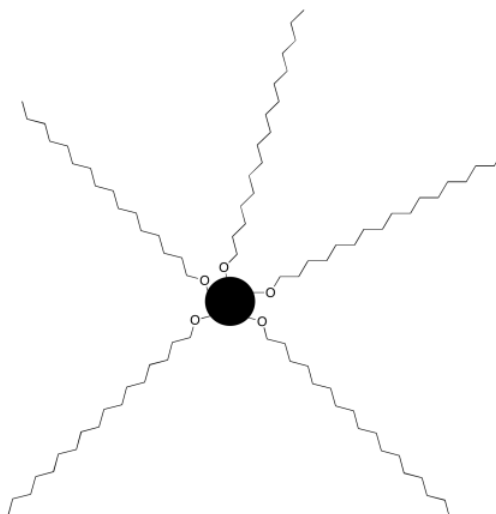


Figure 7.10.1: A cartoon of a C18 bead

A C18 column is an example of a "reverse phase" column. Reverse phase columns are often used with more polar solvents such as water, methanol or acetonitrile. The stationary phase is a nonpolar hydrocarbon, whereas the mobile phase is a polar liquid.

The same approach can also be used in TLC. If a plate is sprayed with a layer of C18 beads, then we can elute the plate in a polar solvent and separate compounds in a sample. Of course, things are reversed now. The most polar compounds will spend the most time in the mobile phase, and move most quickly. The least polar compounds will spend the most time in the stationary phase, and move most slowly.

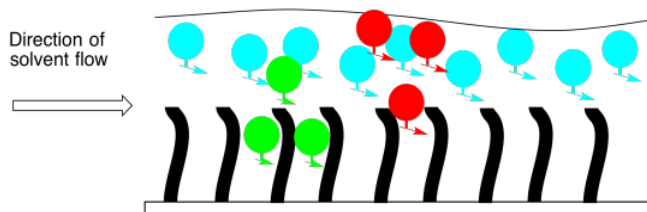


Figure 7.10.2 A cartoon of reverse phase chromatography.

### ? Exercise 7.10.1

On a silica ( $\text{SiO}_2$ ) column, three compounds were eluted in the following order using a hexanes / ethyl acetate mixture: p-dimethylbenzene, p-dimethoxybenzene, then p-methoxyphenol. What might you expect the order of elution would be on a C18 column?

#### Answer

p-methoxyphenol, then p-dimethoxybenzene, then p-dimethylbenzene

### ? Exercise 7.10.2

You are trying to elute a sample on a C18 column using 20:80 mixture of water:acetonitrile, but the compounds are taking too long to come through the column. What should you do?

**Answer**

The compounds are not spending much time in the mobile phase, and have a much higher affinity for the non-polar stationary phase; use more acetonitrile and less water in the mobile phase.

One of the advantages of reverse phase chromatography is that there are many kinds of stationary phase from which to choose. By changing the kind of chain that is to the bead, we can alter how strongly it will interact with certain molecules. Maybe a completely saturated hydrocarbon packing interacts well with saturated hydrocarbons in the sample, making them elute more slowly. On the other hand, maybe a more rigid packing that contains aromatics will interact better with aromatic hydrocarbons in the sample, making those elute more slowly. We might even have some polar groups mixed in, to get a mixture of interactions.

**? Exercise 7.10.3**

Predict the order in which the following compounds would elute from a reverse phase column.

- a. butylbenzene and benzylamine
- b. 2-decanol and decanoic acid
- c. 1-heptanol and 1-heptene
- d. octanal and methyl octyl ether

**Answer****Answer a**

benzylamine then butylbenzene

**Answer b**

2-decanol then decanoic acid

**Answer c**

1-heptanol then 1-heptene

**Answer d**

octanal then methyl octyl ether

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