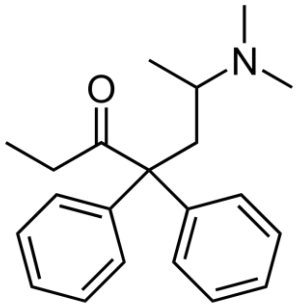
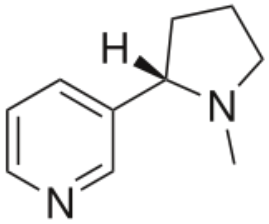
Chemical Analysis Problem

Hair analysis is frequently used for the long-term monitoring of drug and alcohol users.

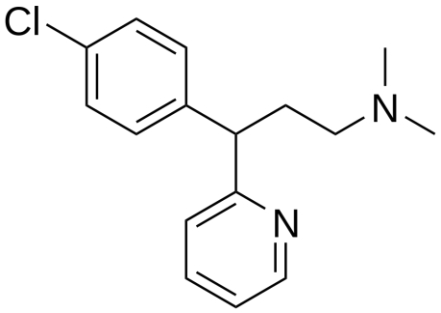
You are working at a forensics laboratory and have been given the task of developing a method for the analysis of hair samples from suspects for alcohol and drug usage. How would you go about doing this to determine if an individual has been using drugs?



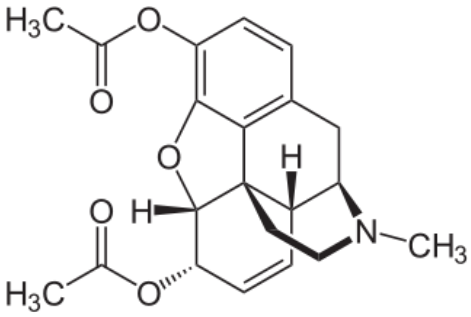
Methadone



Nicotine



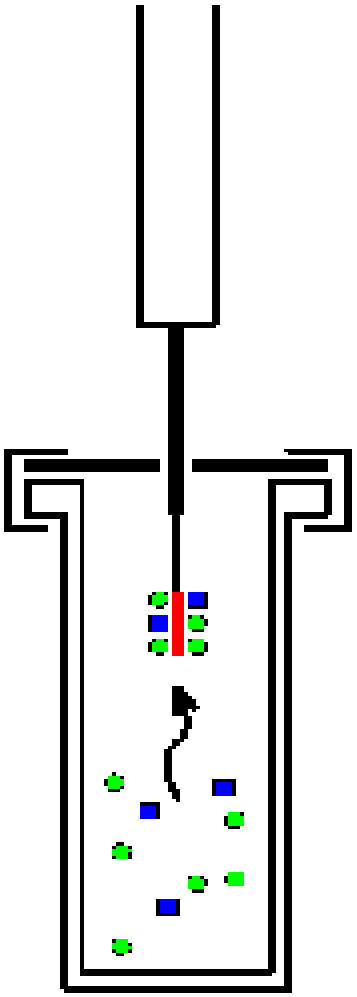
Chlorphenamine



Heroin

Lipophilic – able to dissolved in oils, lipids and non-polar solvents

Solid Phase Microextraction (SPME)



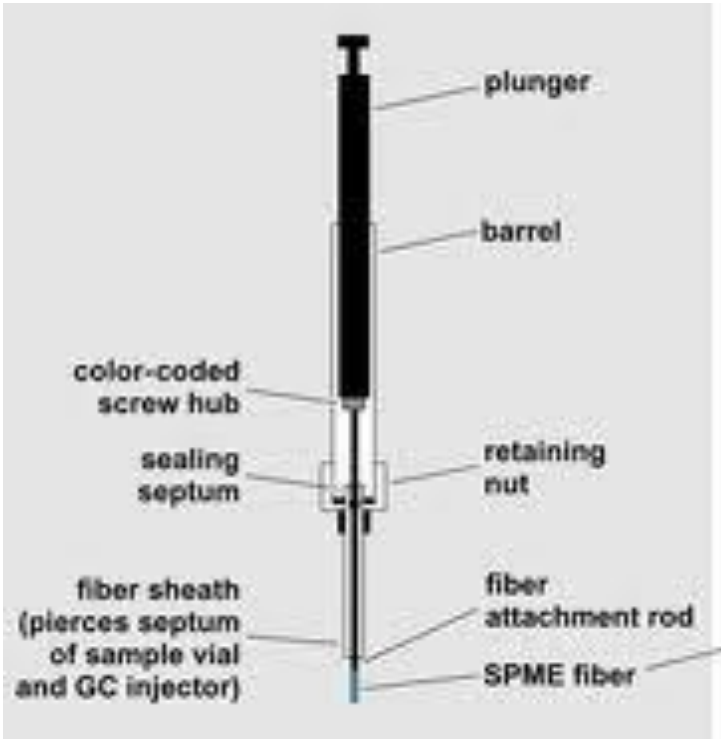
Challenge **– Can the measurement scientist use sample preparation for analyte preconcentration (to lower detection limits) and or to remove analytes from a complex matrix?**

Sample preparation is an essential step in analysis, greatly influencing the reliability and accuracy of resulted the time and cost of analysis. Solid-Phase Microextraction (SPME) is a very simple and efficient, solventless sample preparation method, invented by J. Pawliszyn in 1989.

Hair differs from other traditional biological samples used for human toxicological analysis such as urine, blood, liver or saliva with its significantly longer detection window (months) allowing retrospective investigation and measurement of drug consumption.

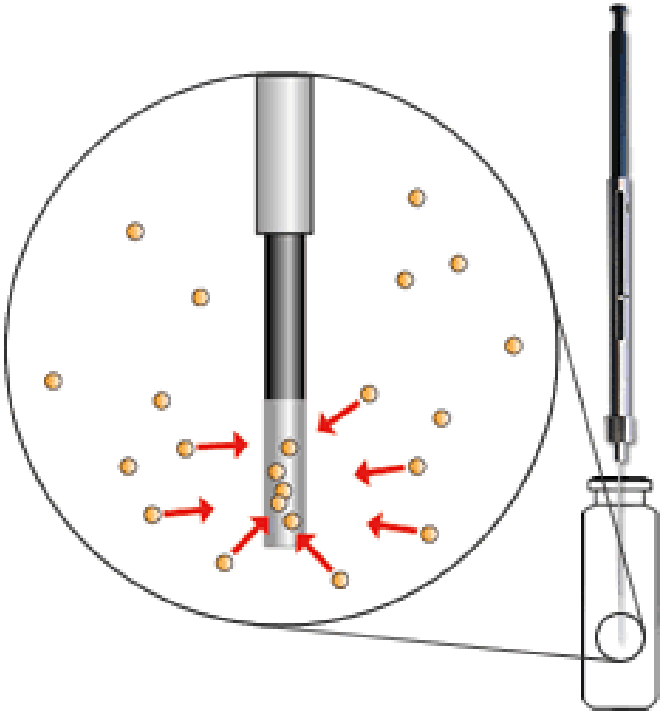
Solid Phase Microextraction (SPME)

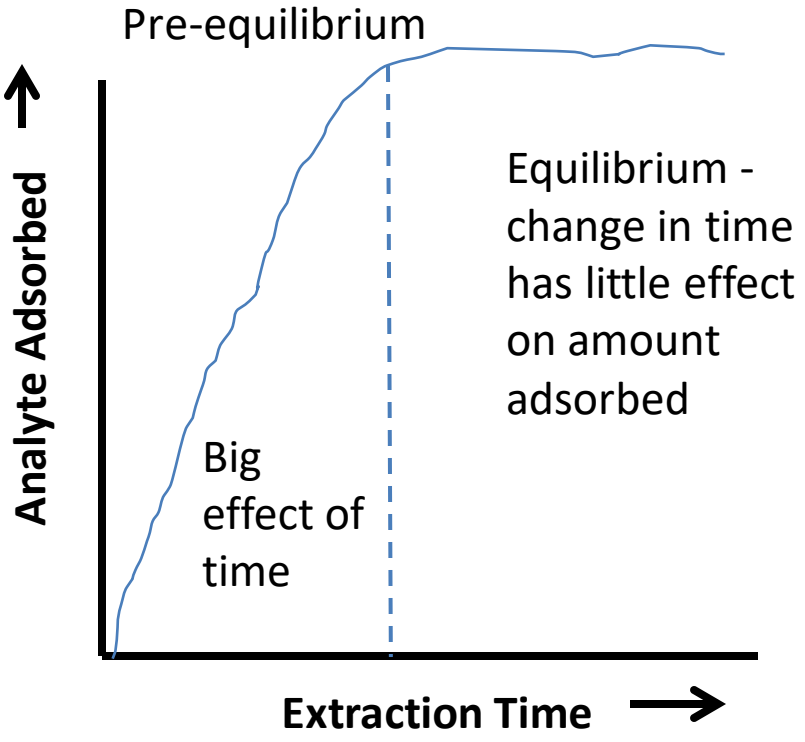
SPME is a fast, solventless alternative to conventional sample extraction techniques. Analytes establish equilibria among the sample matrix, the headspace above the sample, and a polymer-coated fused fiber, then are desorbed from the fiber to a chromatography column.



$$K_{fs} = \frac{C_f^{\infty}}{C_s^{\infty}}$$

Theory of SPME



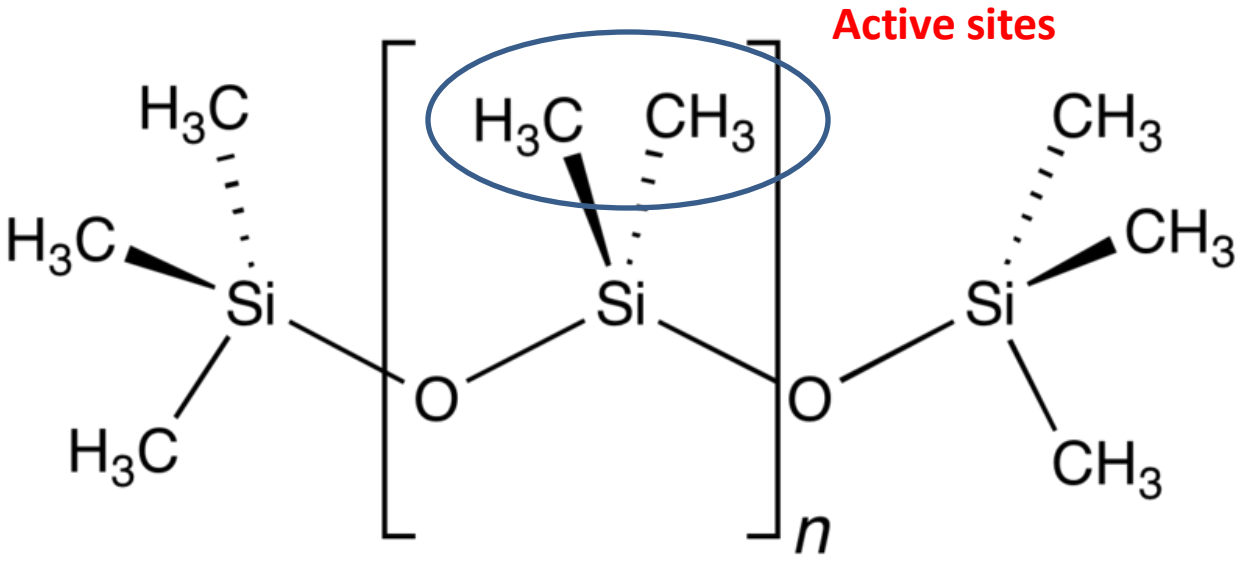


Molecular adsorption onto the solid phase from either head space or solution.

Moles adsorbed (i) = $K_{eq,SPME}$ x V_{SPME} x C(i)

Solid Phase Material

Polydimethylsiloxane (PDMS)



Nonpolar sorbent phase – medium to non-polar volatile and semi-volatile analytes

280-300 °C stability

Useful for both GC and HPLC analysis

100 µm thick

The chemistry of the sorptive SPME layer plays a significant role in enhancing or discriminating against classes of compounds!!

Chemical Problem – Hair Analysis

Apart from external drug deposition on hair, drugs are mainly enclosed tightly in the hair shaft and to a certain extent maybe bound to proteins, melanin or lipids of the cell membrane complex. Therefore, hair matrix type, structure of the drug, method and duration of extraction, and solvent used are all important factors affecting the final extraction yield.

1. **Hair sample collection and preparation (washing, drying and weighing).**

2. **Digestion or removal of drugs from hair matrix (NaOH, CH_3OH)**

3. **Control measurements to determine recovery of drugs. Why important?**

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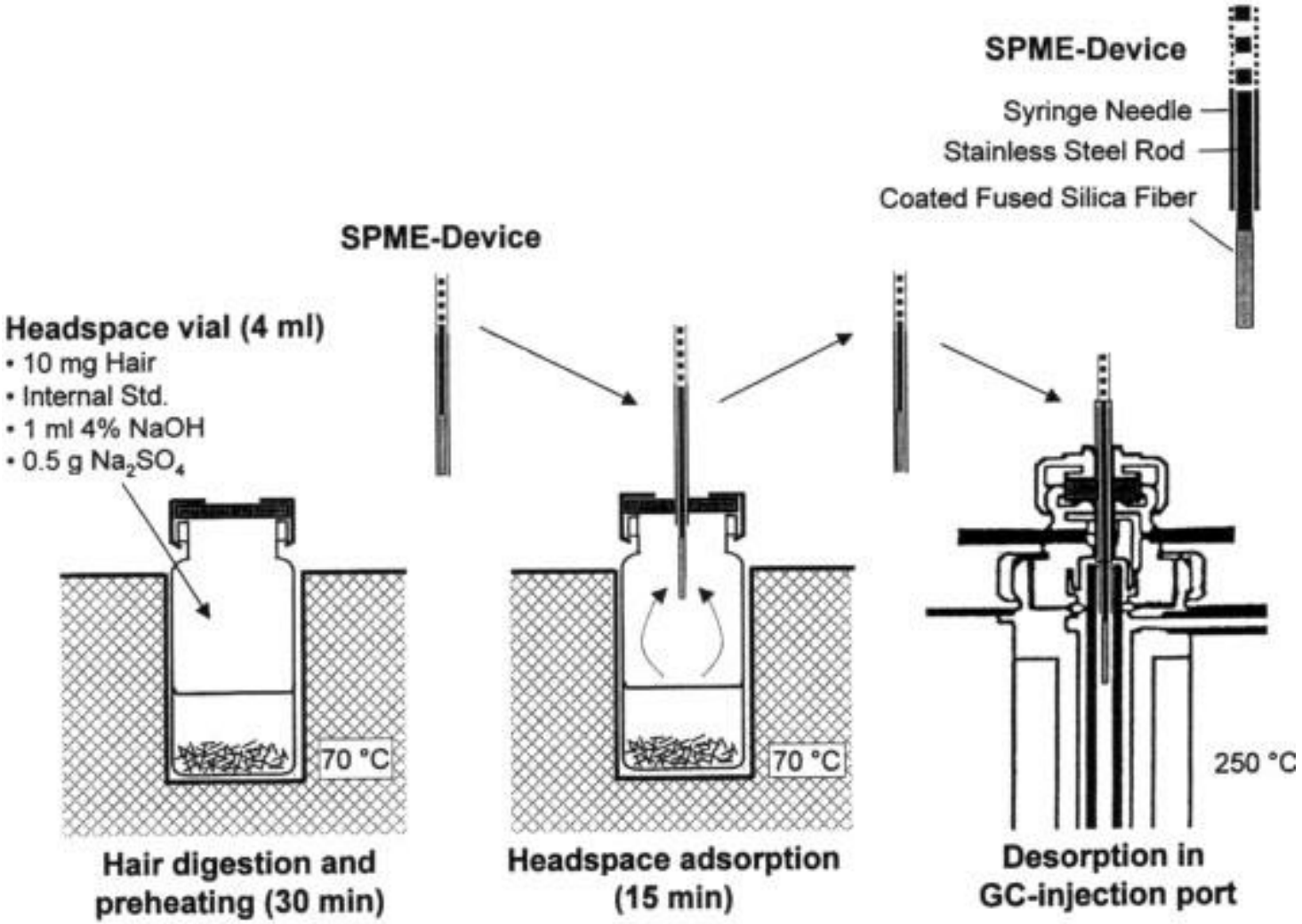
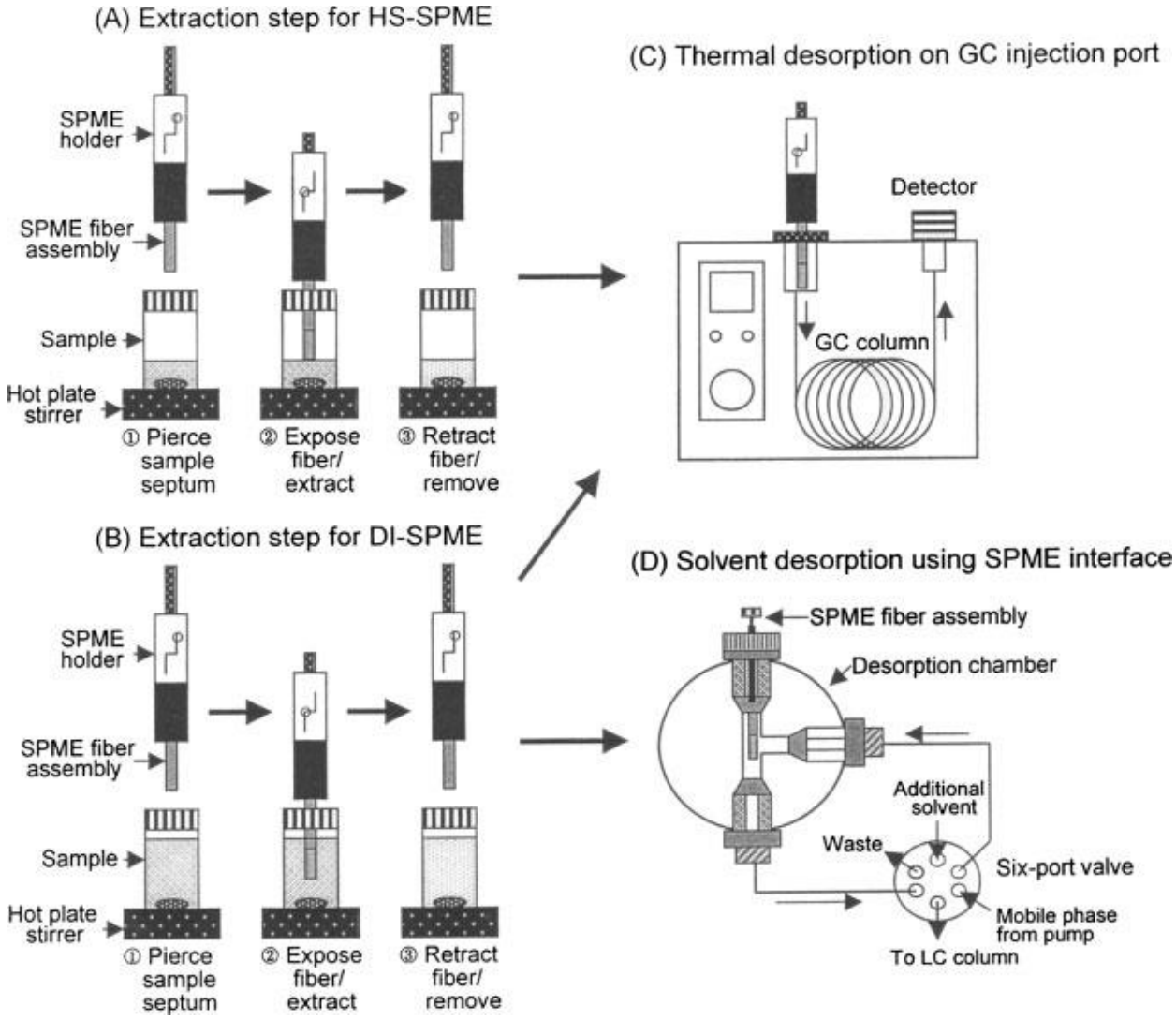
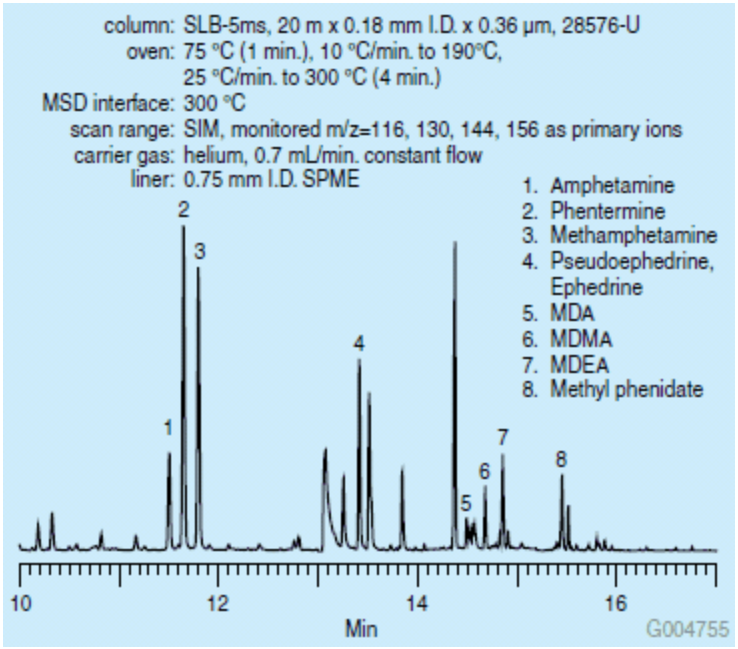
Approach for Hair Analysis



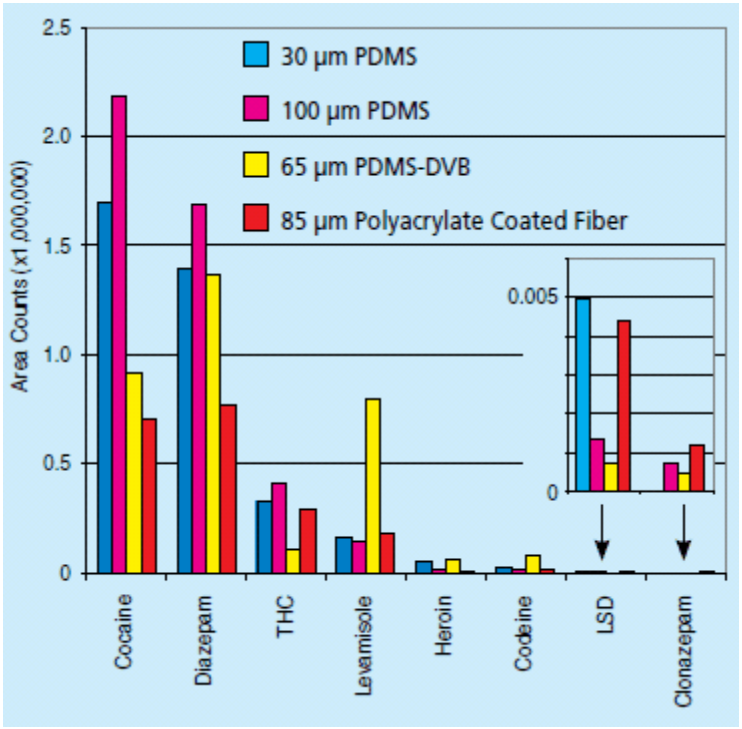
Solid Phase Microextraction – Head Space



Example Data from Hair Analysis



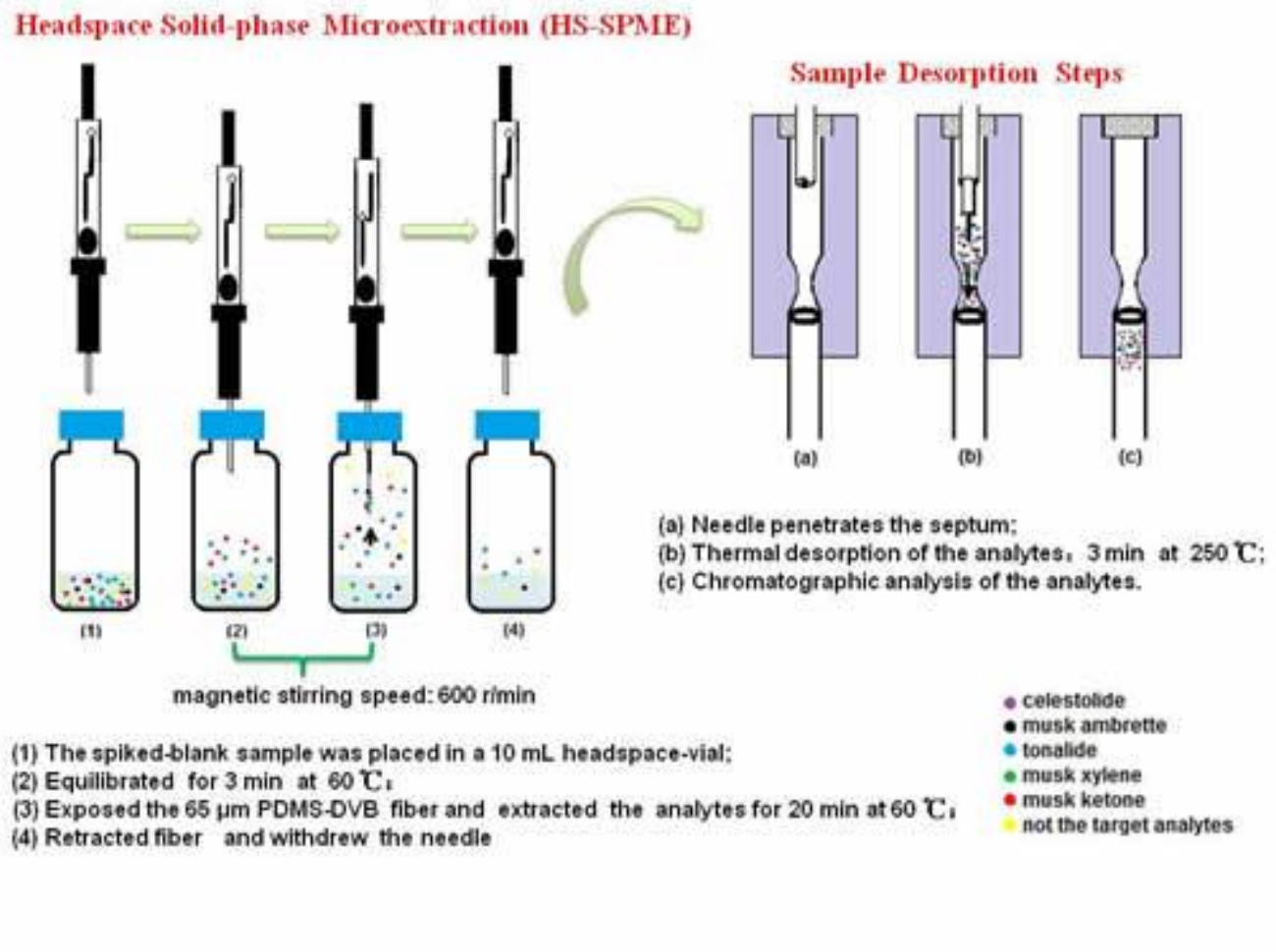
50 µg/L or 50 ppb samples



Fiber chemistry and thickness can make a difference in extraction efficiency.

Sigma-Aldrich

Solid Phase Microextraction (SPME)



25-50 µL injection volume

Percent Recovery

Percent recovery is used in cases where no chemical reaction is taking place, as in purification of a sample. It is calculated as follows:

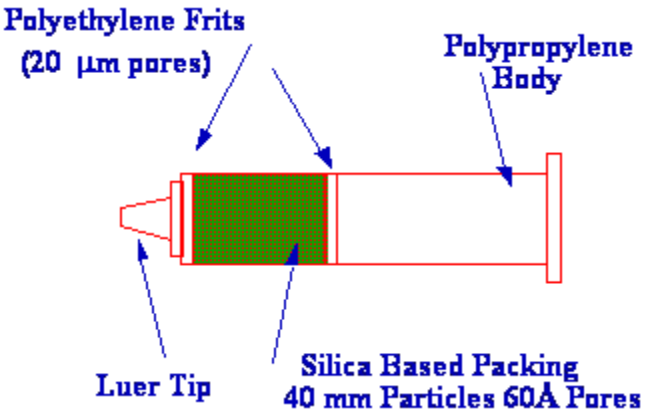
$$\text{Percent Recovery (\%)} = \frac{\text{amount of pure product recovered (g)}}{\text{amount of crude product used (g)}} \times 100$$

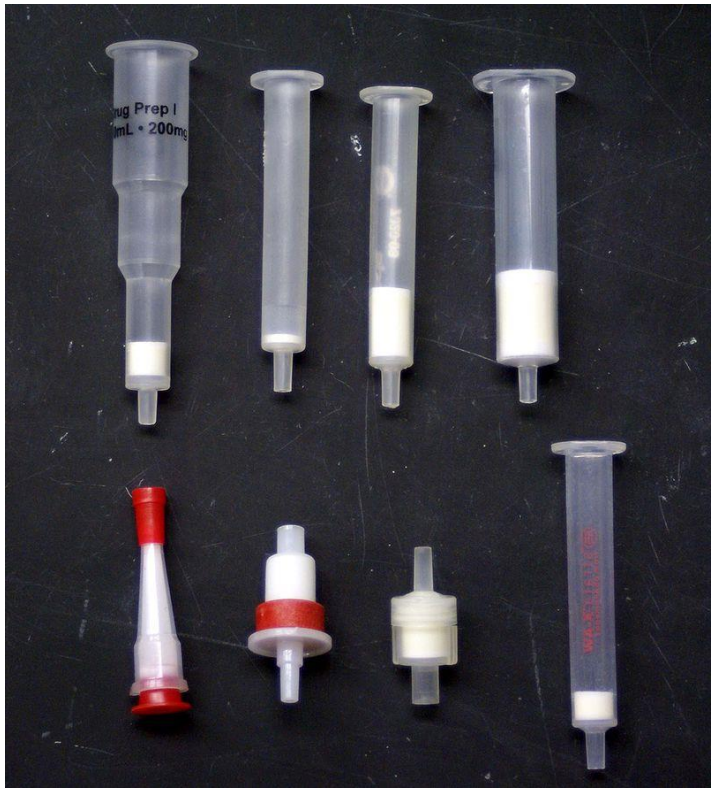
For quantitative analysis of samples using SPME, one must know the % percent recovery. Experiments can be done to determine this. Alternately, one can generate a response curve using the SPME on standard solutions and this would take into account the percent recovery for the specific extraction conditions.

Solid Phase Extraction

Solid-phase extraction (**SPE**) is a sample preparation process by which compounds that are dissolved or suspended in a liquid mixture are separated from other compounds in the mixture according to their physical and chemical properties. Analytical laboratories use solid phase extraction to concentrate and purify samples for analysis.

- Normal phase (polar)
- Reversed phase (non-polar)
- Anion exchange (anions)
- Cation exchange (cations)

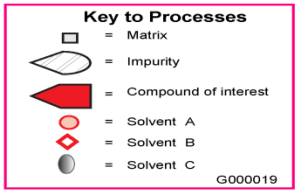


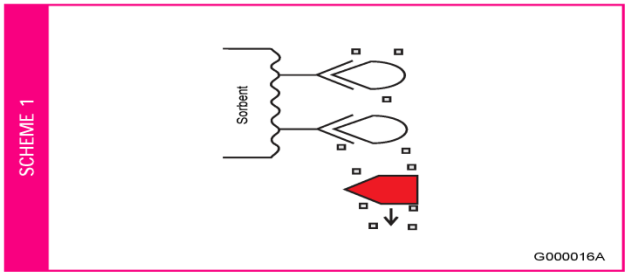


Solid Phase Extraction (SPE)

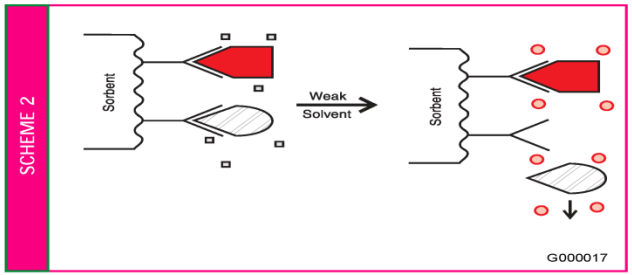
How to Use SPE

Solid phase extraction is used to separate compounds of interest from impurities in three ways. Choose the most appropriate scheme for your sample:

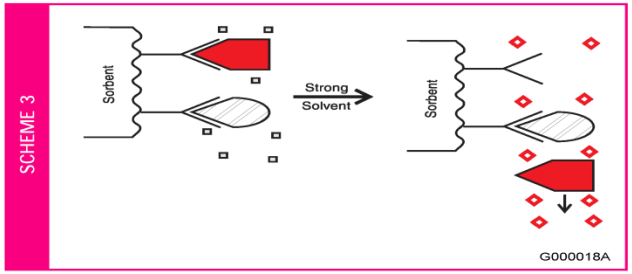




Selective Extraction. Select an SPE sorbent that will bind selected components of the sample — either the compounds of interest or the sample impurities. The selected components are retained when the sample passes through the SPE tube or disk (the effluent will contain the sample minus the adsorbed components). Then, either collect the adsorbed compounds of interest through elution, or discard the tube containing the extracted impurities.



Selective Washing. The compounds of interest and the impurities are retained on the SPE packing when the sample passes through; the impurities are rinsed through with wash solutions that are strong enough to remove them, but weak enough to leave the compounds of interest behind.



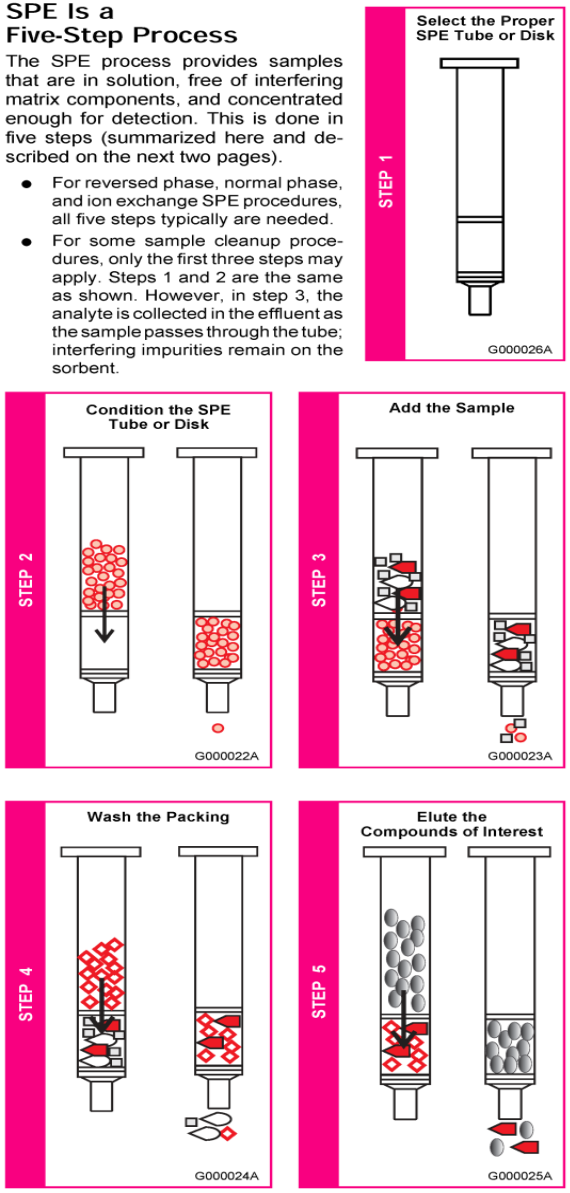
Selective Elution. The adsorbed compounds of interest are eluted in a solvent that leaves the strongly retained impurities behind.

Analytes must have some chemical affinity for the packing material or the stat. phase.

Must select the packing material to have some specific chemical interactions or affinity for the analytes of interest.

Process is very much like that which occurs during an analytical separation on a column, often with some preconcentration used.

Solid Phase Extraction (SPE)



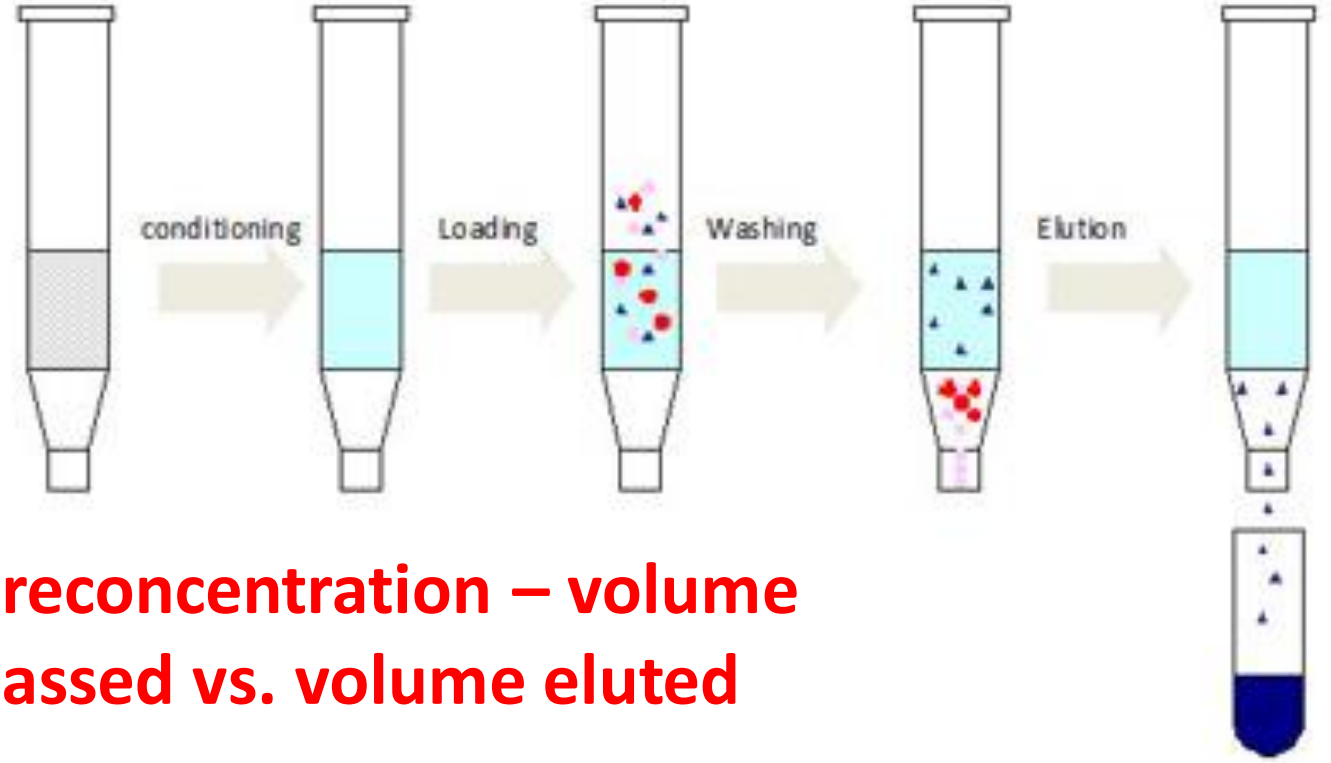
Steps in Process

- Select the proper stat. phase
- Condition the stat. phase
- Add the sample (mL)
- Wash the stat. phase
- Elute the compounds of interest

Solvent or solution selection for the elution depends on the chemistry of the stat. phase and chemical nature of the analytes.

Solid Phase Extraction

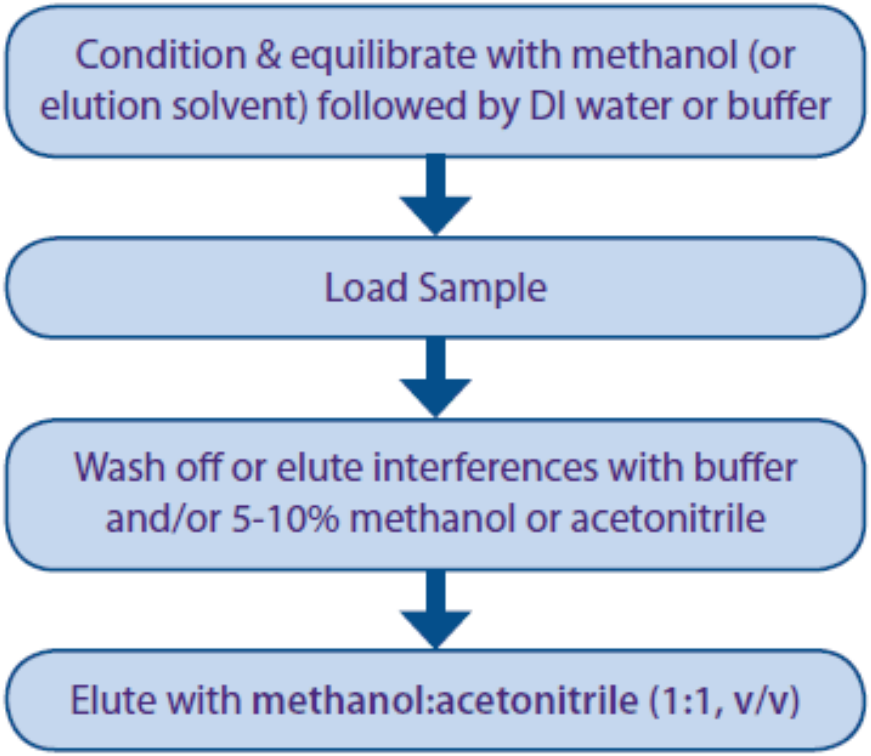
Solid-phase extraction (SPE) is a sample preparation process by which compounds that are dissolved or suspended in a liquid mixture are separated from other compounds in the mixture according to their physical and chemical properties.



Preconcentration – volume passed vs. volume eluted

If 10 mL of sample is passed through the packing and then elution is done with 100 µL of solvent, then preconcentration factor is 100x!!!

Solid Phase Extraction



Analysis often by HPLC. Excellent for sample clean up and analyte preconcentration. Process is much like that which occurs on the stationary phase of an analytical column.