HPLC/UHPLC Detectors

Detectors for HPLC/UHPLC can be grouped as follows:

General: FID, refractive index, thermal conductivity
Spectroscopic: UV, photodiode array, fluorescence, chemiluminescence
Mass: ESI (electrospray ionization), MALDI, ambient ionization (DESI, DART, ASAP-MS)

The detector choice determines how samples are prepared for HPLC/UHPLC. For example, samples that don't burn are not suitable for FID detection. Fluorescence detection requires samples that are fluorescent. And, hydrocarbon-only samples are require an EI (electron impact) source.

What if your sample contains compounds that burn and some that don't? Why isn't FID okay? Stop and think! What happens to a flame, when it is clobbered with solids? Nothing good. Few if any runs are going to be possible before the detector has be replaced. Solution: remove the compounds that don't burn before HPLC/UHPLC.

What about samples that contain UV absorbing and non-UV absorbing compounds? How useful is diode array detection? If the challenge is to determine the ratio of absorbers to non-absorbers, the answer is not useful. If the challenge is to follow the concentration of absorbers over time, then an additional set of experiments is needed to look at how the non-absorbers change (or don't change) the absorbances measured and if the non-absorbers collect on the column and what the non-absorbers do to the detector window. Extra materials on columns change retention times and reproducibility of runs.

A popular mass detector for HPLC/UHPLC uses ESI (electrospray ionization) to generate signals., but it seriously limits the the scope of the HPLC/UHPLC. For example, trifluoroacetic acid (TFA) is a well known ion-pairing agent used to sharpen reversed phase separated peaks. However, it is such a good ion-pairing agent, negative ion ESI ions are often completely suppressed, and positive ions are partially suppressed. Suppression means the signal intensity is reduced. Also TFA tends to stick to the LCMS flow path, so it can take many runs to clear it from an instrument. Sodium phosphate is another useful HPLC/UHPLC additive for reversed phase chromatography. However, inorganic salts are not ESI friendly. They dirty up and clog the spray region of the instrument quickly as they come off the column. Volatile chromatography additives like formic acid or ammonium acetate can be used. DMSO, DMF, and other viscous high boiling liquids stick to the LC flow path and take a long time to disappear. Again, extra stuff on the columns change retention times and reproducibility of runs.

The LCMS instruments the Mass Spectrometry laboratory (Room 2142 in the Chemistry Instrument Center) (CIC), all have ESI detectors and diode array detectors. They are therefore limited to samples that are ESI detectable. **If diode array detection (or UV detection) is all that is needed, you should NOT use the LCMS instruments in the Mass Spectrometry laboratory.**

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With diode array detection, **abuse** is failure to consider where non-UV absorbing compounds are. With ESI detection, **abuse** is failure to consider where neutral (non-ionizable) compounds are.