Ultra High Performance Liquid Chromatography

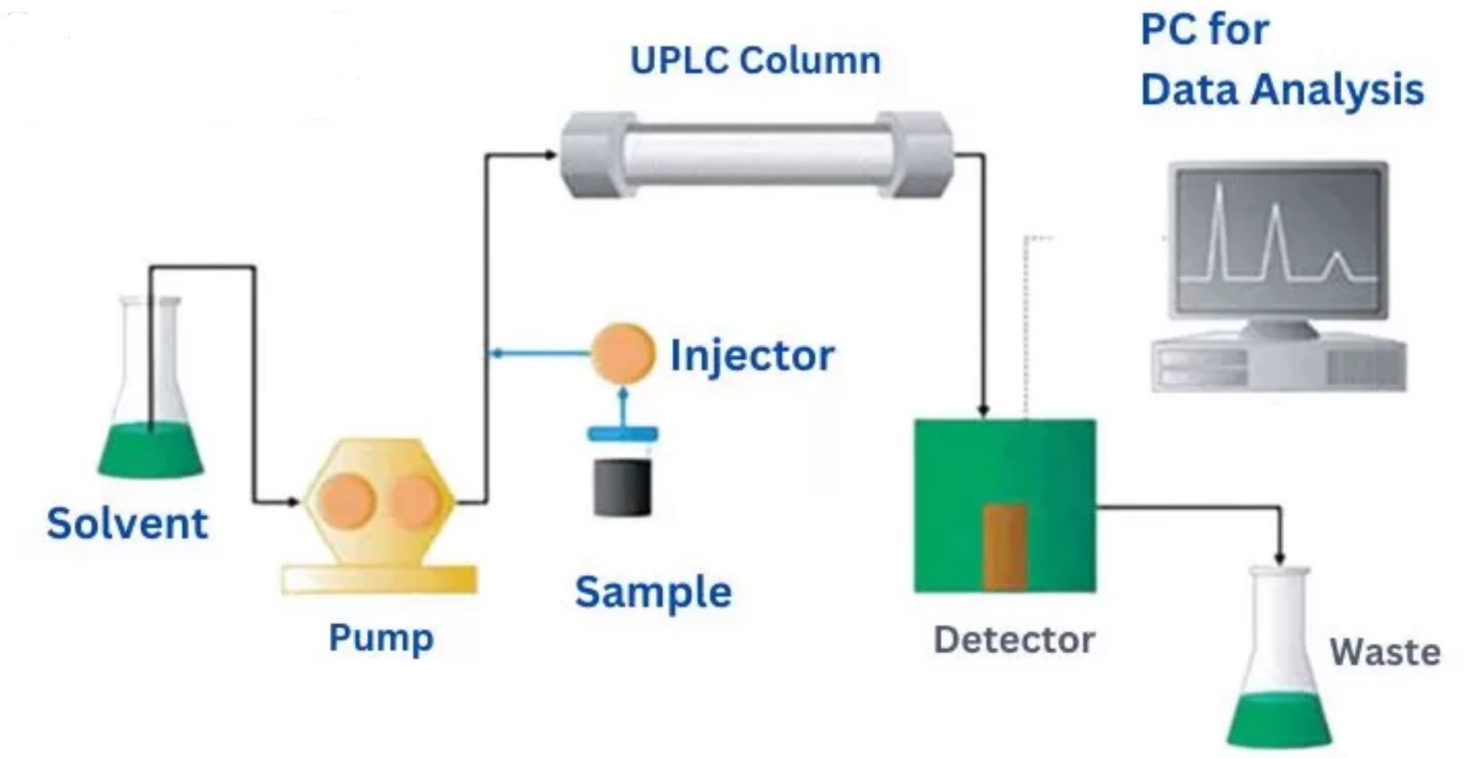
Ultra-High-Performance Liquid Chromatography (UHPLC) is a new and advanced form of liquid chromatography used for separation, identification, and quantification of compounds in complex mixtures.

It's similar to high-performance liquid chromatography (HPLC) but operates at higher pressures (typically above 600 bar or 60 MPa), with finer column particles (sub-2 μm) and lower flow rates.

This results in-

Higher resolution,
Faster separations,
Greater sensitivity,
Higher throughput,
and lower solvent consumption.

Ultra-Performance Liquid Chromatography



Solvent (Mobile Phase):

This liquid (typically a mixture of water, acetonitrile, or methanol) carries the sample through the column. It's kept in a solvent reservoir.

Pump:

The pump pushes the solvent forward under high pressure (up to 15,000 psi or even higher).
This high pressure lets the solvent move faster and carry the sample through the column efficiently.

Injector:

The injector introduces a small volume of your sample into the flow of solvent.
This happens in a controlled and reproducible manner (so you get consistent results).

UPLC Column:

Inside this column, there are tiny particles (typically < 2 μm).
These particles create a large surface area for compounds to interact with, allowing them to separate based on their properties (like size, polarity, or affinity).

Detector:

As the compounds elute (come out of the column) at different speeds, they pass through the detector.
The detector measures their signals — typically UV absorption — which shows their concentration.

PC for Data Analysis:

The signals are then processed by a computer, generating a chromatogram with peaks.
Each peak corresponds to a separate compound in your sample.

Waste:

After the compounds pass through the detector, the liquid exits into a waste container.

ADVANTAGES OF UHPLC-

Higher resolution and faster separations — due to small particle size of column media.

Higher sensitivity and better peak capacity.

Shorter run time — faster method (sometimes 5–10 minutes instead of 30-minute runs with traditional HPLC).

Small solvent consumption, which reduces solvent waste and operational costs.

Enhanced robustness and reproducibility.

Ability to separate complex mixtures with greater precision.

DISADVANTAGES OF UHPLC -

➤ Higher pressure requirement — necessitates specialized, high-pressure equipment.

➤ Higher cost of instrumentation and maintenance.

➤ Column fragility — small particles can generate high backpressure and may be prone to blockage.

➤ Method transfer from HPLC might require extensive re-validation.

➤ Small particle columns can shorten column lifespan due to pressure stress.

APPLICATIONS OF UHPLC -

-Pharmaceutical Analysis – for high-resolution quantitation of drugs, related compounds, or degradation products.

-Proteomic and Metabolomic Research – for complex mixture separations in biotechnology.

-Food and Beverage Quality Control – for detection of pesticides, food colorants, or contaminants.

-Environmental Analysis – for traces of pesticides, heavy metals, or industrial waste.

-Forensic Toxicology – for drug abuse testing, toxin identification, or metabolic studies.

-Clinical Research and Biomedical Applications – for biomarker, disease-related, or metabolic profile studies.