

High-Performance Liquid Chromatography Demonstration

Practical 11 | Instrumental Methods of Analysis and Novel Drug Delivery Systems

Aim

To study High-Performance Liquid Chromatography Demonstration, and interpret the recorded observations.

Principle

- HPLC pumps a liquid mobile phase through a column and measures separated components with the selected detector. Retention, peak area and system suitability have different roles.
- The construction and operating principle determine the function of the equipment.
- The practical distinguishes the input, working zone, controlled variables and output.
- Major parts: solvent reservoirs, degassing arrangement, pump, injector, column, detector and data system.
- Trace the flow path and identify where separation and detection occur.
- A reference calibration can relate peak response to amount; retention time alone is not sufficient identity evidence.

Materials and Apparatus

- Instructor-operated HPLC system, reference/test chromatograms and method record.
- Assigned equipment demonstration, labelled schematic or manufacturer-approved training material.
- Observation sheet and any instructor-supplied performance readings.

Method

1. Identify the major components on the actual equipment or training diagram.
2. Trace the material, energy or signal path through the system.
3. Observe the instructor demonstration and note the purpose of each control.
4. Record the operating variables and the output relevant to the exercise.
5. Relate the equipment principle to its pharmaceutical application.
6. Summarize the variables that influence performance and the checks required before operation.

Observation Record

Complete this record using actual laboratory findings or the provided teaching data.

Observation / parameter	Record / finding
Mobile phase	_____
Column	_____
Flow and detector setting	_____
Retention time	_____
Peak area	_____
Suitability observations	_____

Calculations and Interpretation

- For a validated calibration $y = mC + b$, concentration $C = (\text{peak response} - b)/m$.
- Apply the stated preparation/dilution correction and reference-standard potency.

Result

Record the actual finding, calculated value with units where relevant, and a conclusion supported by the observations.

Finding: _____

Conclusion: _____

Precautions

- Use equipment demonstrations and trained operation for moving, hot or pressurized components.
- Do not bypass guards, interlocks or the assigned operating limits.
- Distinguish operating performance from the appearance of the machine.

Viva Questions

1. What does the HPLC column do?

It provides the stationary phase through which different interactions create separation.

2. Why trace the material or energy path?

It connects the equipment construction to the process it performs.

3. Why are operating variables recorded?

They are needed to explain or reproduce the observed performance.

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