

High Performance Liquid  
Chromatography (HPLC)

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HPLC

- Collect blood sample
- Separate plasma (partial clean-up)
- Extraction into organic solvent (polarity/pH)
- Injection onto column
- Separation on column
- Quantitation with detector
- Record and analyse results

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High Performance Liquid  
Chromatography

- Pump
- Injector
- Column
- Detector
- Recorder

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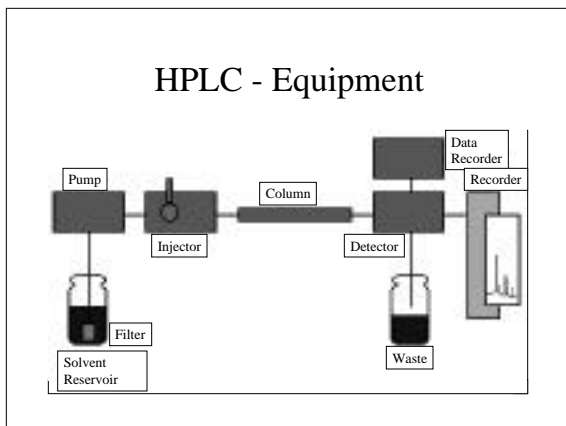
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### Solvent / Pump / Injector

- Solvent reservoir
- High pressure - 1000 to 5000 psi
- Injector
  - Low pressure - stop flow
  - High pressure valve

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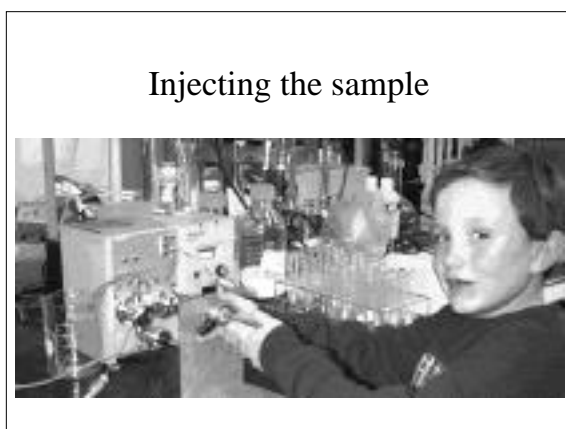
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### Column

- Normal Phase
  - Silica gel - non-aqueous
  - Adsorption
- Reverse phase (C8, C18) - aqueous
  - Partitioning
- Ion-exchange - aqueous
- Molecular sieve - aqueous
  - Size

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### Detector

- Specific
  - Absorbance
  - Fluorescence
  - Electrochemical
- Non-specific
  - Refractive index
  - Radioactivity
  - Conductivity

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### Detector



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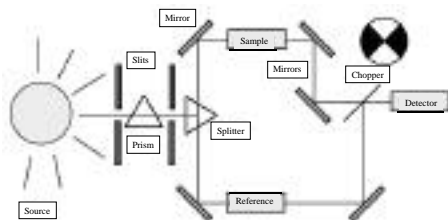
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### UV-Visible Double Beam



Redrawn from: Instrumental Analysis by Bauer, Christian and O'Reilly, 1978, page 187

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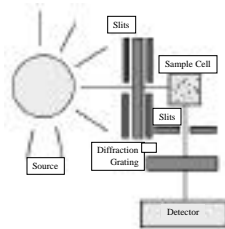
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### Fluorescence Detector



Redrawn from: Instrumental Analysis by Bauer, Christian and O'Reilly, 1978, page 235

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### Recorder



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