Chromatography can be used to determine the types and quantities of amino acids in a polypeptide, but cannot be used to determine a sequence since to prepare for chromatography a polypeptide is first hydrolyzed.

Ninhydrin is used to visualize amino acids, but not identify them (product of ninhydrin has no R group).

Fractions — different portions of the solvent eluting (coming out) from the column that may contain one or (non-ideally) more solutes.

\[ A = \varepsilon \cdot C \cdot l \]

Beer's law — Unique property of an individual compound since the absorbance and volume of a fraction can be measured, and since the path length used to measure a sol'n would be known, the concentration (and therefore moles) of an amino acid can be determined.

Remember: In chromatography, the identity of an amino acid is determined by retention time.
Edman degradation - Allows individual amino acids to be cleaved from a polypeptide chain in order, allowing sequencing of that chain.

Reagent: Phenyl isothiocyanate (PITC)

\[ R-N=\overset{C=O}{\ddots} + H_2O \quad \rightarrow \quad \text{thiourea} \]

At this point of the mechanism, the remainder of the polypeptide is ejected from the PITC derivative. The PITC derivative can therefore be used to identify the amino acid that had been pulled off the chain.

Thiazolinone

\[ N-H \quad \rightarrow \quad \text{phenylhydantoin} \]

(Can be isolated and used to identify the amino acid removed.)
Synthesis of amino acids

23.6

[Chemical structures and reactions]

Diethyl N-phthalimidomalonate ester will decarboxylate.

\[ R \xrightarrow{\text{HCl}} H_2N\text{R} \text{NH} \text{COOH} \]

Will be hydrolyzed.