

(c) Separation techniques

Use of centrifugation to separate substances of differing density

More dense components settle in the pellet

Less dense components remain in the supernatant

Paper and thin layer and affinity chromatography can be used for separating different substances such as:

different amino acids

different sugars

The speed that each solute travels along the chromatogram depends on its differing solubility in the solvent used

Principle of affinity chromatography and its use in separating proteins

A solid matrix or gel is created with specific molecules bound to the matrix or gel

Soluble, target proteins in a mixture, with a high affinity for these molecules, become attached to them as the mixture passes down the column

Other non-target molecules with a weaker affinity are washed out

2. Proteins

(a) The proteome

The proteome is the entire set of proteins expressed by a genome.

The proteome is larger than the number of genes, particularly in eukaryotes, because more than one protein can be produced from a single gene as a result of alternative RNA splicing

Not all genes are expressed as proteins in a particular cell

Genes that do not code for proteins are called non-coding RNA genes and include those that are transcribed to produce tRNA, rRNA, and RNA molecules that control the expression of other genes

The set of proteins expressed by a given cell type can vary over time and under different conditions

Some factors affecting the set of proteins expressed by a given cell type are

- metabolic activity of the cell

- cellular stress

- the response to signaling molecules

- diseased versus healthy cells

The polypeptide folds into a tertiary structure

This conformation is stabilised by interactions between R groups:

- hydrophobic interactions

- ionic bonds

- London dispersion forces

- Hydrogen bonds

- disulphide bridges – covalent bonds between R groups containing sulphur

Quaternary structure exists in proteins with several connected polypeptide subunits

Quaternary structure describes the spatial arrangement of the subunits

A prosthetic group is a non-protein unit tightly bound to a protein, necessary for its function

The ability of haemoglobin to bind oxygen is dependent upon the non-protein haem group

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Interactions of the R groups can be influenced by temperature and pH

Increasing temperature disrupts the interactions that hold the protein in shape; the protein begins to unfold, eventually becoming denatured

The charges on acidic and basic R groups are affected by pH

As pH increases or decreases from the optimum, the normal ionic interactions between charged groups are lost, which gradually changes the conformation of the protein until it becomes denatured

3. Membrane proteins

(a) Movement of molecules across membranes

The fluid mosaic model of cell membranes:

- phospholipid bilayer which acts as a barrier to ions and most uncharged polar molecules
- integral proteins embedded in membranes
- peripheral proteins attached to surface membranes

Regions of hydrophobic R groups allow strong hydrophobic interactions that hold integral proteins within the phospholipid bilayer

Integral membrane proteins interact extensively with the hydrophobic region of membrane phospholipids

Some integral proteins are transmembrane proteins, for example channels, transporters and many receptors.

Peripheral membrane proteins have hydrophilic R groups on their surface and are bound to the surface of membranes, mainly by ionic and hydrogen bond interactions

Many peripheral membrane proteins interact with the surfaces of integral membrane proteins

The phospholipid bilayer as a barrier to ions and most uncharged polar molecules.

Some small molecules such as oxygen and carbon dioxide pass through the bilayer by simple diffusion

Facilitated diffusion is the passive transport of substances across the membrane through specific transmembrane proteins

4. Communication and signalling

(a) Coordination

Multicellular organisms signal between cells using extracellular signalling molecules

Steroid hormones, peptide hormones, and neurotransmitters are examples of extracellular signalling molecules

Receptor molecules of target cells are proteins with a binding site for a specific signal molecule.

Binding changes the conformation of the receptor, which initiates a response within the cell

Different cell types produce specific signals which can only be detected and responded to by cells with the specific receptor

Signalling molecules may have different effects on different target cell types due to differences in the intracellular signalling molecules and pathways that are involved

In a multicellular organism, different cell types may show a tissue-specific response to the same signal.

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