

iv. Chemistry and metabolism of Proteins and Amino acids...

iv. Chemistry and metabolism of Proteins and Amino acids, Ramachandran plot, primary, secondary, tertiary and quaternary structure of proteins, Mechanisms and specificity of Enzymes, Coenzymes and Cofactors, Disorders associated with protein and amino acid metabolism, proteomics

Chemistry and Metabolism of Proteins and Amino Acids

Amino Acid Structure and Classification

1. General Structure

- Each amino acid has a central (α) carbon, an amino group ($-\text{NH}_2$), a carboxyl group ($-\text{COOH}$), a hydrogen atom, and a variable side chain (R-group).
- At physiological pH (~ 7.4), the amino group is protonated ($-\text{NH}_3^+$) and the carboxyl group is deprotonated ($-\text{COO}^-$), making amino acids zwitterions.

2. Classification

- Nonpolar (Hydrophobic):** e.g., Ala, Val, Leu, Ile, Phe.
- Polar, Uncharged:** e.g., Ser, Thr, Asn, Gln, Tyr.
- Positively Charged (Basic):** Lys, Arg, His.
- Negatively Charged (Acidic):** Asp, Glu.

3. Essential vs. Nonessential Amino Acids

- Humans can synthesize some amino acids (**nonessential**); others (**essential**, e.g., Val, Leu, Ile, Lys, etc.) must be obtained from the diet.
- Semi-essential or conditionally essential amino acids (e.g., Arg, His) may be required in certain physiological states (e.g., infancy).

Protein Turnover and Amino Acid Metabolism

1. Protein Turnover

- Continuous synthesis and degradation of proteins; helps maintain amino acid pools and remove damaged or misfolded proteins.
- Ubiquitin-Proteasome System** and **Autophagy-Lysosome Pathway** key to selective protein degradation.

2. Amino Acid Catabolism

- Transamination:** Transfer of α -amino group to α -ketoglutarate, forming glutamate and an α -keto acid (catalyzed by transaminases).
- Deamination:** Removal of the amino group (e.g., oxidative deamination by glutamate dehydrogenase).
- Urea Cycle:** Detoxifies ammonia into urea (ornithine cycle in the liver). Dysfunction leads to hyperammonemia.

3. Carbon Skeleton Utilization

- Glucogenic** amino acids: Carbon skeletons can yield substrates for gluconeogenesis (e.g., pyruvate, TCA intermediates).
- Ketogenic** amino acids: Carbon skeletons form acetyl-CoA or acetoacetyl-CoA, can generate ketone bodies or lipids (e.g., Leu, Lys).
- Some are both glucogenic and ketogenic (e.g., Ile, Phe, Tyr, Trp).

Ramachandran Plot

1. Definition and Utility

- A Ramachandran plot plots the dihedral angles ϕ (**phi**) ($\text{N}-\text{C}\alpha$) vs. ψ (**psi**) ($\text{C}\alpha-\text{C}'$) for amino acid residues in a polypeptide.
- Conformational space is restricted by steric hindrance, so only certain angle combinations are energetically allowed (e.g., α -helical, β -sheet, collagen triple helix regions).

2. Significance

- Helps evaluate protein structures (e.g., from X-ray crystallography or NMR) for sterically disallowed

conformations that indicate errors.

- Gly and Pro are exceptions: **Gly** is more flexible, **Pro** is more restricted due to its ring structure.

Levels of Protein Structure

Primary Structure

- **Definition:** The linear sequence of amino acids in a polypeptide chain, read from the N-terminus to the C-terminus.
- **Importance:** Determines how the chain folds and what higher-order structures form.
- **Peptide Bond Properties:** Partial double-bond character (resonance) restricts rotation, typically in trans configuration except for certain X-Pro bonds.

Secondary Structure

1. **α-Helix**
 - Right-handed helix stabilized by **intrachain hydrogen bonds** (C=O of residue n with N-H of residue n+4).
 - Side chains project outward. Proline disrupts α-helices; Gly can increase flexibility.
2. **β-Sheet**
 - **Extended conformation** stabilized by hydrogen bonds between backbone amides of adjacent strands.
 - Can be **parallel** or **antiparallel**. Side chains alternate above and below the sheet.
3. **Turns and Loops**
 - **β-Turns** reverse the polypeptide direction, often found on protein surfaces.
 - Essential for compact folding, connecting secondary structure elements.

Tertiary Structure

- **Definition:** The overall 3D conformation of a single polypeptide chain, including interactions among side chains (hydrophobic effects, hydrogen bonds, ionic interactions, disulfide bonds).
- **Domains:** Distinct structural/functional units within a polypeptide.
- **Stabilizing Forces:** Hydrophobic core, salt bridges, hydrogen bonds, **disulfide bonds** (covalent links between cysteine residues), metal ion coordination.

Quaternary Structure

- **Definition:** Assembly of multiple polypeptide subunits into a functional complex (e.g., hemoglobin's α₂β₂ structure).
- **Forces:** Similar noncovalent interactions as tertiary structure; sometimes disulfide bonds.
- **Functional Significance:** Cooperativity, allostery, subunit regulation, structural stability.

Mechanisms and Specificity of Enzymes

Enzyme Kinetics and Catalysis

1. **Active Site**
 - Specialized pocket where substrate binding and catalysis occur. Conformational changes can enhance substrate alignment (induced fit).
2. **Transition State Stabilization**
 - Lowering activation energy (ΔG^\ddagger) is central to catalysis. Enzymes often bind transition states more tightly than substrates, fostering product formation.
3. **Michaelis-Menten Kinetics**
 - $v = \frac{V_{\max} [S]}{K_m + [S]}$
 - **K_m** (Michaelis constant) relates to substrate affinity; **V_{max}** is maximum rate at saturating [S].
4. **Catalytic Mechanisms**
 - Acid-base catalysis, covalent catalysis, metal ion catalysis, proximity/orientation effects.



Specificity and Regulation

1. Substrate Specificity

- Determined by shape, charge, polarity of the active site.
- **Lock-and-key** vs. **Induced fit** models.

2. Allosteric Regulation

- Effector molecules bind sites other than the active site, altering enzyme activity.
- Sigmoidal kinetics typical of cooperative enzymes (e.g., ATCase in pyrimidine synthesis).

3. Covalent Modification

- Phosphorylation, methylation, acetylation modulate enzyme activity.
- Zymogen activation by proteolytic cleavage (e.g., trypsinogen → trypsin).

Coenzymes and Cofactors

1. Definition

- **Cofactors:** Non-protein chemical compounds (metal ions or organic molecules) required for enzymatic activity.
- **Coenzymes:** Organic cofactors (often derived from vitamins) that transiently associate with the enzyme (e.g., NAD⁺ from niacin).

2. Examples

- **Metal Ions:** Fe²⁺, Zn²⁺, Mg²⁺ in various enzymes (e.g., iron in cytochromes).
- **Vitamins:** B vitamins → coenzyme forms (e.g., Thiamine pyrophosphate (B1), FAD (B2), NAD⁺ (B3), CoA (B5), Pyridoxal phosphate (B6), Biotin (B7), Folate derivatives (B9), Cobalamin (B12)).

3. Function

- Participate in redox reactions, group transfers, electron carriers, assisting in enzyme conformational changes.
- **NAD⁺/NADH** and **FAD/FADH₂** vital for oxidative metabolism.
- **CoA** for acyl group transfers, etc.

Disorders Associated with Protein and Amino Acid Metabolism

Amino Acid Metabolic Defects

1. Phenylketonuria (PKU)

- Deficiency of **phenylalanine hydroxylase** or tetrahydrobiopterin cofactor. Elevated phenylalanine → neurological damage if untreated.
- Dietary phenylalanine restriction essential.

2. Maple Syrup Urine Disease (MSUD)

- Defect in branched-chain α-ketoacid dehydrogenase complex. Accumulation of Leu, Ile, Val and their α-keto acids leads to neurological symptoms.
- Control via dietary restriction of branched-chain AAs.

3. Alkaptonuria

- Defect in **homogentisate oxidase**, leading to buildup of homogentisic acid. Characterized by dark urine on standing, ochronosis (pigmentation in cartilage).

4. Homocystinuria

- Often due to **cystathionine β-synthase** deficiency. High homocysteine can cause cardiovascular and connective tissue problems.
- B6 supplementation, dietary methionine/homocysteine management.

Urea Cycle Disorders

- **Ornithine Transcarbamylase (OTC) Deficiency:** X-linked, leads to hyperammonemia, elevated orotic acid.
- Other enzyme defects (e.g., carbamoyl phosphate synthetase I deficiency) also cause hyperammonemia, neurotoxicity.

Protein Misfolding and Aggregation

- **Neurodegenerative Diseases:** Alzheimer's (β -amyloid), Parkinson's (α -synuclein), prion diseases (PrP^{Sc}).
- Protein quality control failures (chaperones, proteasome) implicated in pathology.

Proteomics

Definition and Approaches

1. **Proteomics:** The large-scale study of the entire set of proteins (proteome) expressed by a cell, tissue, or organism, including their modifications, interactions, and functions.
2. **Techniques**
 - **2D Gel Electrophoresis:** Separate proteins by isoelectric point (pI) and molecular weight.
 - **Mass Spectrometry:** Identify proteins/peptides (e.g., MALDI-TOF, LC-MS/MS).
 - **Protein Microarrays or Affinity-based methods:** Assess protein-protein or protein-ligand interactions.

Biological and Clinical Significance

1. **Biomarker Discovery**
 - Proteomic profiles in diseases (e.g., cancer) can reveal diagnostic or prognostic markers.
2. **Functional Proteomics**
 - Identification of protein-protein interaction networks, post-translational modifications (phosphorylation, glycosylation, etc.).
3. **Systems Biology Integration**
 - Proteomics complements genomics and metabolomics to yield a holistic view of cellular function and disease mechanisms.

Concluding Remarks

Proteins and amino acids are central to life's chemistry, fulfilling roles in **enzymatic catalysis**, **structural frameworks**, **signaling**, and **transport**. Their **primary** sequence determines higher-level folding (secondary, tertiary, quaternary), governed by **steric constraints** (Ramachandran plot) and diverse **intermolecular interactions**. **Enzymes** exemplify the sophistication of **specificity and catalysis**, often reliant on **cofactors** and **coenzymes** derived from dietary vitamins and minerals.

Disorders in protein/amino acid metabolism—from enzymatic defects (e.g., PKU, MSUD, urea cycle deficiencies) to misfolded protein aggregation—underscore the delicate balance required for normal physiology. Finally, **proteomics** offers an unparalleled lens through which to interrogate protein expression, modifications, and interactions on a systems-wide scale, paving the way for **precision medicine** and deeper insights into the molecular underpinnings of health and disease.