BIOC 384: M03.T01-Miesfeld

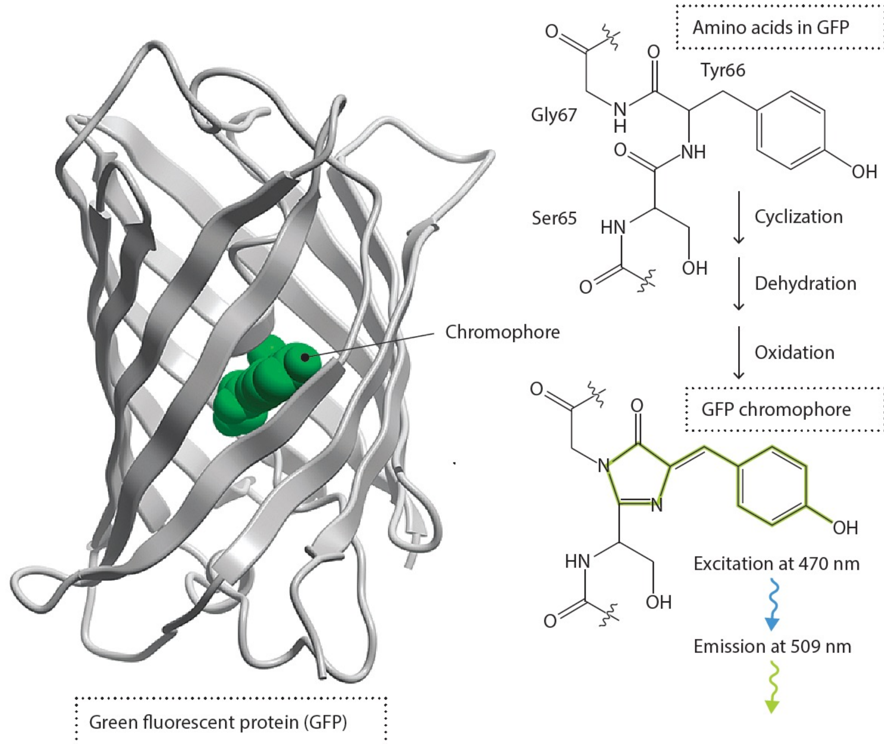
Assigned Reading: *Biochemistry* Chapter 4.1a

THE UNIVERSITY OF ARIZONA

Amino Acid Functional Groups

The Big Picture

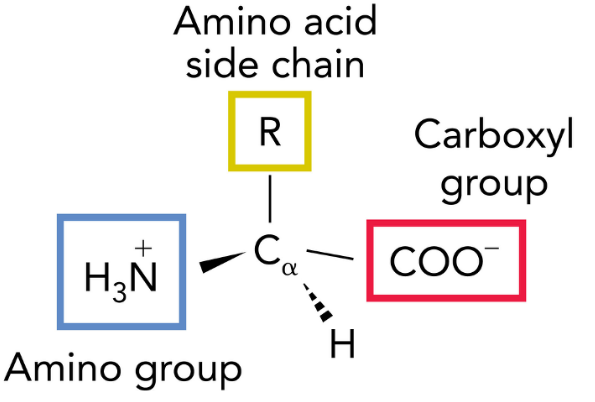
- The 20 amino acids can be grouped into four subfamilies based on the chemistry of their R groups (charged, hydrophilic, hydrophobic, and aromatic).
- Understanding how these R groups facilitate protein structure and function can provide insights into how proteins fold, interact with water, and function in diverse cellular environments.



Chemical Grouping of Amino Acids

- The 20 amino acids can be divided into four subfamilies based on their R group that emphasize the chemical interaction of amino acids with the aqueous environment of the cell at neutral pH (7.2-7.6):

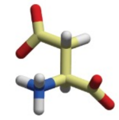
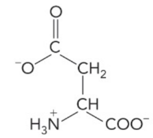
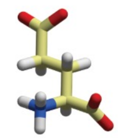
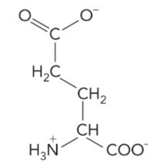
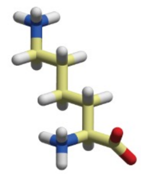
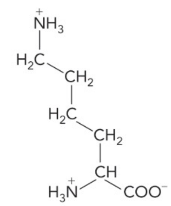
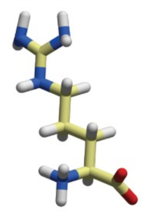
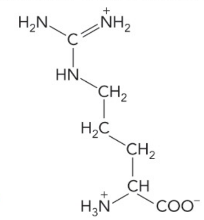
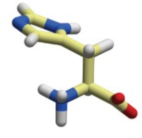
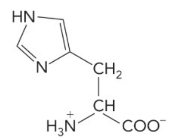
- **Charged** (Asp, Glu, Lys, Arg, His)
- **Hydrophilic** (Ser, Thr, Cys, Asn, Gln)
- **Hydrophobic** (Gly, Ala, Pro, Val, Leu, Ile, Met)
- **Aromatic** (Phe, Tyr, Trp)



- Some amino acids could fit into more than one subfamily when considering other chemical attributes. For example, the charged amino acids are polar and hydrophilic and the aromatic amino acids are nonpolar and hydrophobic.

Charged Amino Acids

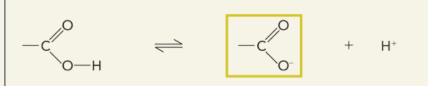
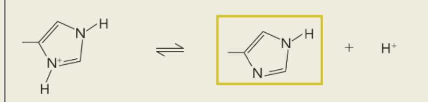
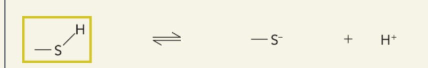
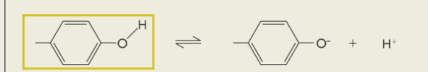
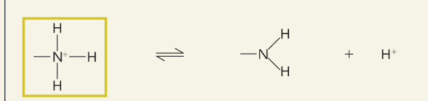
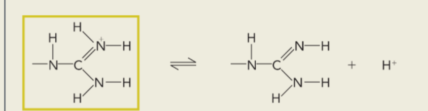
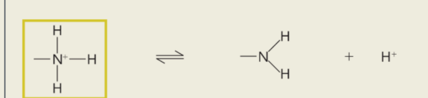
- The five charged amino acids—Asp, Glu, Lys, Arg, His—carry net positive or negative charges at neutral pH due to ionizable side chains.
- These five amino acids are key to ionic interactions, active site chemistry, and protein solubility in aqueous environments.

Name	Molecular structure	Chemical structure	Molecular mass (Da)	Percent frequency in proteins (%)	Comments
Negatively charged					
Aspartate Asp D			133	5.3	Ionizable side chain has a pK_a of ~3.9. In proteins, Asp often has a net negative charge at pH 7.
Glutamate Glu E			147	6.3	Ionizable side chain has a pK_a of ~4.1. In proteins, Glu often has a net negative charge at pH 7.
Positively charged					
Lysine Lys K			146	5.9	Ionizable side chain has a pK_a of ~10.5. In proteins, Lys often has a net positive charge at pH 7.
Arginine Arg R			174	5.1	Ionizable side chain has a pK_a of ~12.5. In proteins, Arg often has a net positive charge at pH 7.
Histidine His H			155	2.3	Ionizable side chain has a pK_a of ~6.0. In proteins, the imidazole ring in His can be either positively charged or neutral depending on the environment.

Ionization of Amino Acids

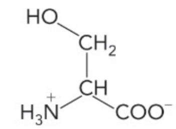
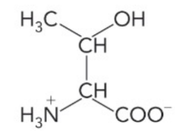
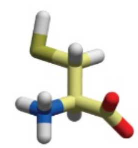
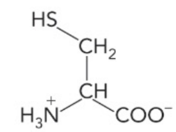
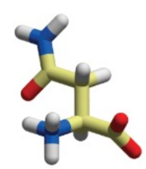
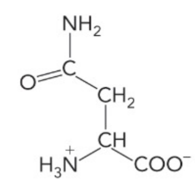
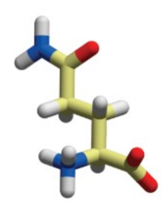
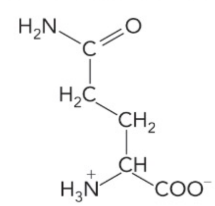
- pK_a values are affected by the local chemical environment of the amino acid side chains, so the actual pK_a may be higher or lower, depending on the protein.
- Boxed ionization states are the most abundant at neutral pH.

Table 4.3 Ionization States and Approximate pK_a Values of Titratable Groups Within Proteins

Amino acid	Group	Acid ⇌ Base[a]	Typical pK_a[b]
Aspartic acid Glutamic acid	Side-chain carboxyl group		4.0
Histidine	Imidazole group		6.0
Cysteine	Thiol group		8.3
Tyrosine	Aromatic hydroxyl group		10.1
Lysine	ε amino group		10.5
Arginine	Guanidino group		12.5
Terminal residues	α carboxyl group		3.1
	α amino group		8.0

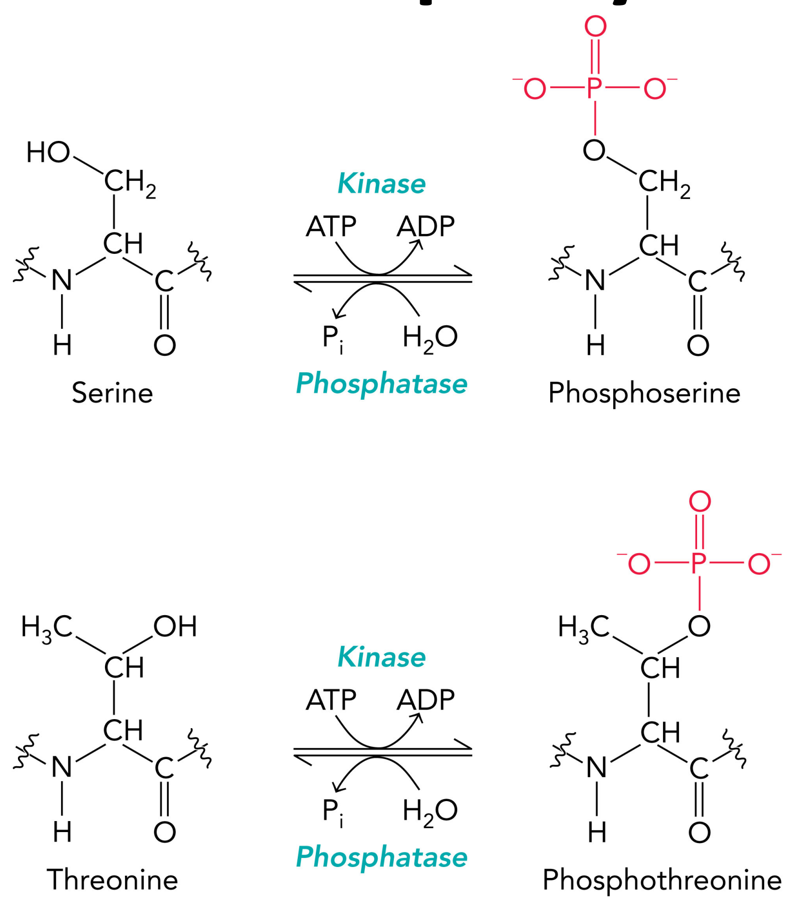
Hydrophilic Amino Acids

- Five amino acids in the hydrophilic amino acid family (serine, threonine, cysteine, asparagine, glutamine) are polar and uncharged at physiologic pH.
- These five amino acids all contain functional groups that participate in hydrogen bonding with H_2O and have short aliphatic side chains.

Name	Molecular structure	Chemical structure	Molecular mass (Da)	Percent frequency in proteins (%)	Comments
Serine Ser S			105	6.8	Hydroxyl group in Ser can form hydrogen bonds and may be a substrate in proteins for kinase-mediated phosphorylation.
Threonine Thr T			119	5.9	Chemically similar to Ser in that the hydroxyl group can form hydrogen bonds and may be phosphorylated by kinases.
Cysteine Cys C			121	1.9	The ionizable side chain has a pK_a of ~8.3 in proteins. The sulfhydryl can form a disulfide bond with other cysteines and can form weak hydrogen bonds. It is the only L-amino acid with an absolute configuration of *R*.
Asparagine Asn N			132	4.3	Often found on the surface of globular proteins, the side chain of Asn can form numerous hydrogen bonds. It is structurally similar to the charged amino acid aspartate.
Glutamine Gln Q			146	4.2	Like asparagine, the side chain of Gln can form numerous hydrogen bonds. It is structurally similar to the charged amino acid glutamate.

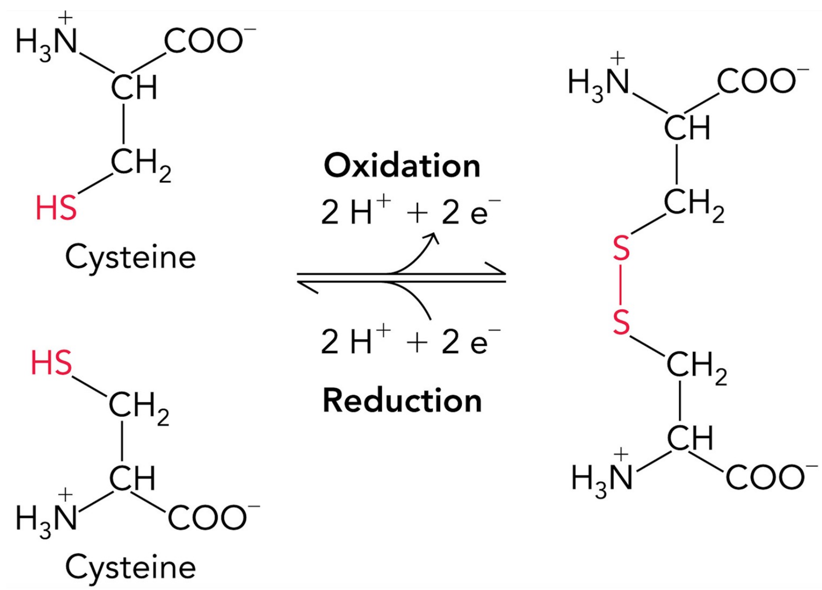
Serine and Threonine can be Phosphorylated

- The hydroxyl group on serine and threonine residues can be phosphorylated by enzymes called serine/threonine kinases, which use ATP as a phosphate donor.
- Phosphatases are enzymes that remove phosphoryl groups from biomolecules by catalyzing a hydrolysis reaction.



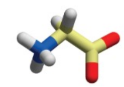
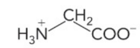
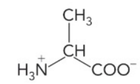
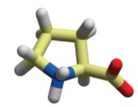
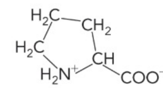
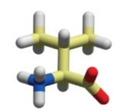
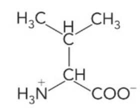
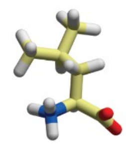
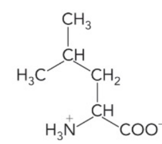
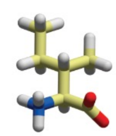
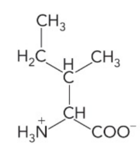
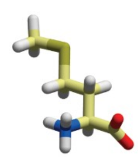
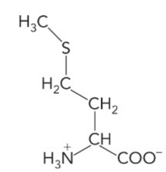
Cysteine Residues Form Disulfide Bonds

- Cysteine has a reactive sulfhydryl group (pKa ≈ 8.3) that can form covalent disulfide bonds in oxidizing environments, which can stabilize protein structure.
- A reduction reaction readily restores the two sulfhydryl groups.



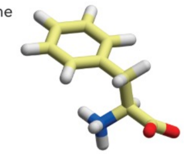
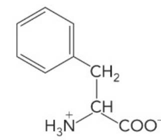
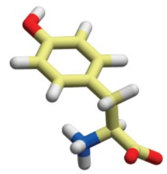
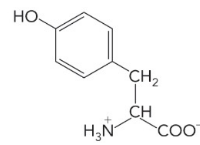
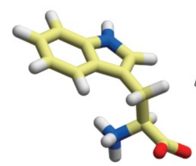
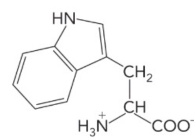
Hydrophobic Amino Acids

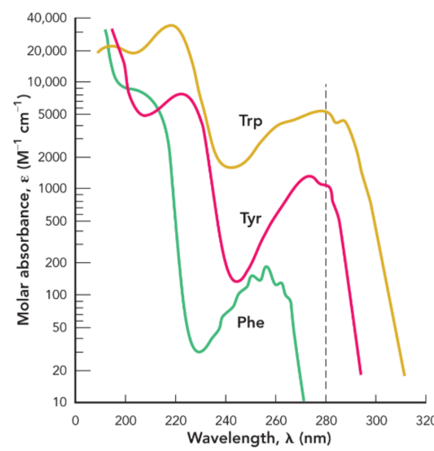
- The six amino acids alanine, proline, valine, leucine, isoleucine, and methionine are nonpolar and have hydrophobic properties and reside in the protein interior.
- Glycine is not a charged, hydrophilic, or aromatic amino acid and included in the hydrophobic subfamily as an orphan.

Name	Molecular structure	Chemical structure	Molecular mass (Da)	Percent frequency in proteins (%)	Comments
Glycine Gly G			75	7.2	The smallest amino acid; the hydrogen side chain makes Gly the most chemically neutral of all the amino acids.
Alanine Ala A			89	7.8	Minimally hydrophobic, Ala is one of the more abundant amino acids in proteins.
Proline Pro P			115	5.2	Cyclic ring limits the conformations that proline can adopt. In a polypeptide, the backbone nitrogen lacks a hydrogen, which limits hydrogen bonding.
Valine Val V			117	6.6	Val is a small hydrophobic amino acid often found in the hydrophobic core of globular proteins.
Leucine Leu L			131	9.1	Leu is the most abundant amino acid in proteins and plays a major role in promoting hydrophobic interactions in the core of globular proteins.
Isoleucine Ile I			131	5.3	Ile is often found in close proximity to Leu and other hydrophobic amino acids in proteins.
Methionine Met M			149	2.3	Unlike the sulfur in Cys, the sulfur in Met is unreactive. Met is often the amino-terminal amino acid in nascent polypeptides.

Aromatic Amino Acids

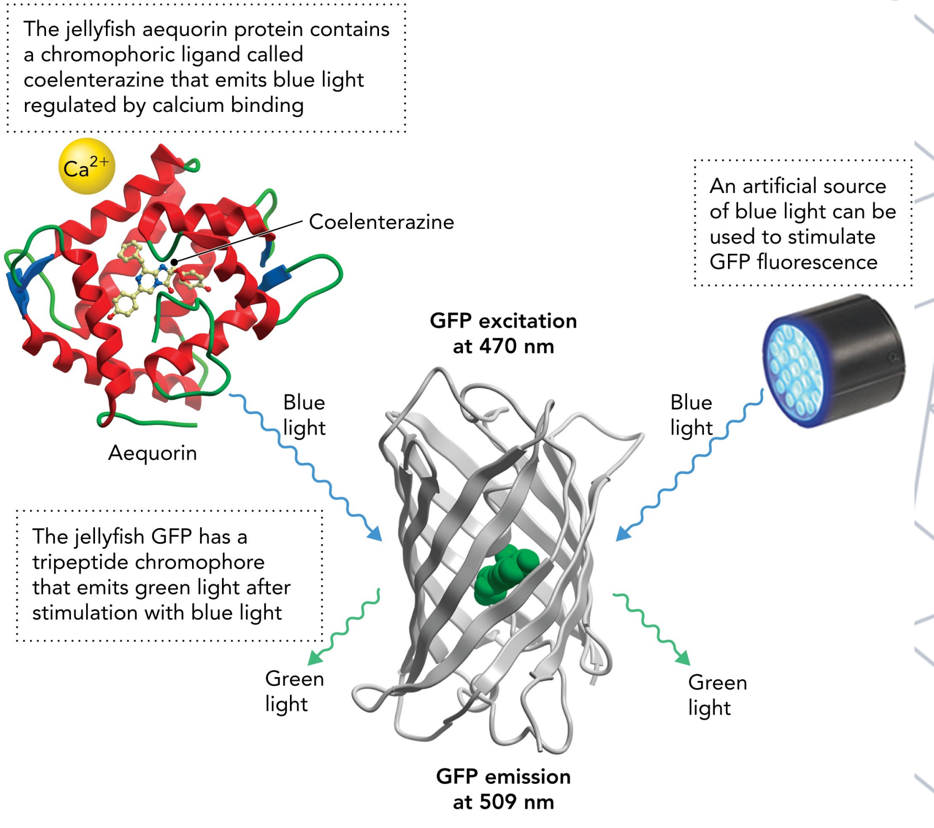
- The aromatic amino acids are phenylalanine, tyrosine, and tryptophan. Tyrosine can be phosphorylated by kinases.
- The aromatic amino acids absorb ultraviolet light at 280 nm, which is used to measure protein concentration in solutions.

Name	Molecular structure	Chemical structure	Molecular mass (Da)	Percent frequency in proteins (%)	Comments
Phenylalanine Phe F			165	3.9	Phenyl ring is hydrophobic and chemically inert. Unlike Tyr and Trp, Phe only weakly absorbs ultraviolet light.
Tyrosine Tyr Y			181	3.2	The ionizable hydroxyl group (pK_a ~10.1 in proteins) can form hydrogen bonds. Tyr is an amphipathic amino acid and absorbs light at 280 nm. Phosphotyrosine is functional in signaling pathways
Tryptophan Trp W			204	1.4	Trp is the largest amino acid and strongly absorbs light at 280 nm. The indole ring is amphipathic, and the ring nitrogen can form hydrogen bonds.



GFP: A Case Study in Aromatic Chemistry

- Green fluorescent protein (GFP) from jellyfish derives its chromophore from Ser65, Tyr66, and Gly67.
- It emits a green light at 509 nm after excitation with blue light at 470 nm from either aequorin that is located in the jellyfish light organ or a light source.



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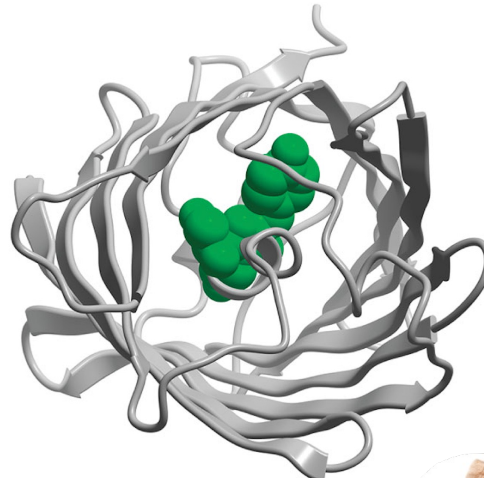
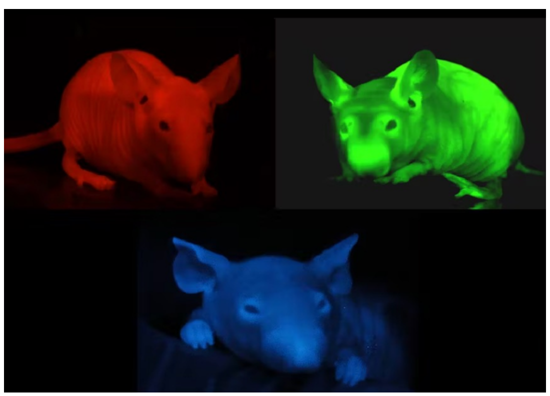
GFP: A Case Study in Aromatic Chemistry

- When cells expressing a recombinant GFP–target protein fusion are illuminated at a wavelength of 470 nm, it results in peak emission at 509 nm that can be visualized as green light.





A variety of autofluorescence proteins have been engineered that emit light at different wavelengths and the genes encoding these proteins incorporated into the genomes of lab animals.

Key Concepts to Guide Your Learning

- The 20 amino acids can be divided into four subfamilies on the basis of shared chemical properties: charged, hydrophilic, hydrophobic, and aromatic.
- Some amino acids can fit into more than one subfamily, for example, the charged amino acids are polar and hydrophilic, whereas the aromatic amino acids are nonpolar and have hydrophobic properties.
- Glycine does not fit easily into any of the four subfamilies because its hydrogen side chain is chemically inert; Gly is put into the hydrophobic subfamily because it is not a charged, hydrophilic, or aromatic amino acid.
- Green fluorescent protein (GFP) is an autofluorescent protein from jellyfish that undergoes a spontaneous cyclization reaction involving Ser65, Tyr66, and Gly67, which generates an intrinsic chromophore.