Proteins

Profa. dra. Édira Castello Branco de Andrade Gonçalves

http://www.unirio.br/analisedealimentos
Reaction

carbon-nitrogen bonds

methyl amine

Amides

nitrogen atoms connected to the carbon atom of a carbonyl group

\[
\text{CH}_3\text{CH}_2\text{C}\text{OH} + \text{H}\text{N}\text{CH}_3 \rightarrow \text{CH}_3\text{CH}_2\text{C}\text{N}\text{CH}_3 + \text{H}_2\text{O}
\]

carboxylic acid

amine

amidation

amide

water

POLYMER TO FORM PROTEINS

https://opentextbc.ca/chemistry/chapter/20-4-amines-and-amides/

http://www.unirio.br/analisedealimentos
Amino acids

POLYMER

peptide bond

peptide bond
Peptide bond

Amino acid + Amino acid → Peptide bond

Condensation reaction

H₂O

Polypeptide chain

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Hydrolysis of Amides

\[ \text{R} \text{C} = \text{NR}_2' \xrightarrow{\text{H}_2\text{O}, \text{heat}} \text{R} \text{C} \text{OH} + \text{R}_2'\text{NH} \]

Amides → carboxylic acid + amine

Reduction of Amides

\[ \text{LiAlH}_4 + \text{R} \text{C} = \text{NH}_2 \rightarrow \text{H} - \text{C} - \text{NH}_2 \]

Amides → amine

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PROPERTIES OF AMINO ACID

L-Amino Acid  D-Amino Acid

Chirality of amino acids

Proteins L configuration

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PROPERTIES OF AMINO ACID

ZWITTER ION

Eletric charge – zero
Isoelectric point - pl
pH - specific each amino acid

http://www.unirio.br/analisedealimentos
PROPERTIES OF AMINO ACID

https://www2.chemistry.msu.edu/faculty/reusch/virttxtjml/proteins.htm
PROPERTIES OF AMINO ACID

a) At pH values above the isoelectric point the protein is negatively charged

b) pH=pl, the number of negative and positive charges is equal

c) At pH values below the isoelectric point the protein is positively charged

d) pH-dependence of the solubility of the β-lactoglobulin protein

ionic repulsion between the protein molecules is minimal, intermolecular attracting ionic interactions is at its maximum
PROPERTIES OF AMINO ACID

Interaction competition

- Ions
- Water
- Proteins

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The distribution of charged species in a sample can be shown experimentally by observing the movement of solute molecules in an electric field, using the technique of **electrophoresis**.

Properties of Amino Acid

https://www2.chemistry.msu.edu/faculty/reusch/virttxtjml/proteins.htm

[Diagram showing electrophoresis with amino acids and their pIs: alanine (pI=6.01), arginine (pI=10.77), isoleucine (pI=6.02), aspartic acid (pI=2.80)].
Carboxylic Acid Esterification

\[
\text{R-CO}_2^\text{CH}_3 + \text{NaHCO}_3 \rightarrow \text{R-CO}_2^\text{CH}_3 + \text{NH}_2
\]

Amine Acylation

\[
\text{R-CO}_2^- + \text{C}_6\text{H}_5\text{COCl} \rightarrow \text{R-CO}_2^- + \text{C}_6\text{H}_5\text{NH}_3^+
\]

https://www2.chemistry.msu.edu/faculty/reusch/virttxtjml/proteins.htm

http://www.unirio.br/analisedealimentos
The Ninhydrin Reaction

[Chemical reaction diagram]

Paper Chromatography

color test for these amino acids

Oxidative Coupling

[Chemical reaction diagram]

Cysteine-Cystine Interconversion

https://www2.chemistry.msu.edu/faculty/reusch/virttxtjml/proteins.htm

http://www.unirio.br/analisedealimentos
THE 20 TYPES OF AMINO ACIDS CLUSTERED BY THEIR PHYSICAL-CHEMICAL PROPERTIES

https://amit1b.wordpress.com/the-molecules-of-life/about/amino-acids/

http://www.unirio.br/analisedealimentos
Primary Structure

- C-N double bond character in amide (peptide) bonds
- Planar peptide bond segments

**β-Pleated Sheets**

- Extended Stick Model

Helical Coiling

- Ala-Thr-Gly-Ala-Phe-Leu-Ala-Phe-Ser-Ile-Gly
- N-Terminus
- One turn of Ala-Thr-Gly-Ala-Phe-Leu-Ala-Phe-Ser-Ile-Gly in the α-helix
- Hydrogen bonds

The Secondary and Tertiary Structure

- Model

https://www2.chemistry.msu.edu/faculty/reusch/virttxtjml/protein2.htm#aacd6

http://www.unirio.br/analisedealimentos
Quaternary Structures

ATP synthase
symmetry mismatch

Hemoglobin

Beta chains
Heme units with iron atom

Alpha chains

RNA polymerase II
asymmetric

https://www2.chemistry.msu.edu/faculty/reusch/virttxtjml/proteins.htm

http://www.unirio.br/analisedealimentos

http://www.biochemsocietrans.org/content/ppbiost/40/3/475/F6.large.jpg
Schematic diagram of protein cross-linking and three types of cross-links including dead-end. Cross-link, intra-peptide cross-link and interpeptide cross-link (RG - Reactive groups).

http://www.unirio.br/analisedealimentos
Denaturing

Denaturation

Normal protein

Denatured protein

Renaturation

loss of biological activity

regains activity

Protein Thermal Irreversible Denaturation

Native albumen

Denaturation

Crosslinking

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## Denaturing

<table>
<thead>
<tr>
<th>Denaturing Action</th>
<th>Mechanism of Operation</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Heat</strong></td>
<td>hydrogen bonds are broken by increased translational and vibrational energy.</td>
</tr>
<tr>
<td></td>
<td>(coagulation of egg white albumin on frying.)</td>
</tr>
<tr>
<td><strong>Ultraviolet Radiation</strong></td>
<td>Similar to heat (sunburn)</td>
</tr>
<tr>
<td><strong>Strong Acids or Bases</strong></td>
<td>salt formation; disruption of hydrogen bonds.</td>
</tr>
<tr>
<td></td>
<td>(skin blisters and burns, protein precipitation.)</td>
</tr>
<tr>
<td><strong>Urea Solution</strong></td>
<td>competition for hydrogen bonds.</td>
</tr>
<tr>
<td></td>
<td>(precipitation of soluble proteins.)</td>
</tr>
<tr>
<td><strong>Some Organic Solvents</strong></td>
<td>change in dielectric constant and hydration of ionic groups.</td>
</tr>
<tr>
<td>(e.g. ethanol &amp; acetone)</td>
<td>(disinfectant action and precipitation of protein.)</td>
</tr>
<tr>
<td><strong>Agitation</strong></td>
<td>shearing of hydrogen bonds.</td>
</tr>
<tr>
<td></td>
<td>(beating egg white albumin into a meringue.)</td>
</tr>
</tbody>
</table>
Heat stabilization uses rapid conductive heating - permanent elimination of all enzymatic activity

http://www.denator.com/what-hs/how
Molecular interpretation of gluten network development after water addition associated with mixing

Scanning electron micrograph of the gluten network

Ortolan & Steel 2017

http://www.unirio.br/analisedealimentos
The foaming properties of camel and bovine whey: The impact of pH and heat treatment

The effect of heat treatment (70°C or 90°C for 30 min) on the foaming and interfacial properties of acid and sweet whey obtained from bovine and camel fresh milk was examined. The maximum foamability and foam stability were observed for acid whey when compared to sweet whey for both milks, with higher values for the camel whey. This behavior for acid whey was explained by the proximity of the pI of whey protein (4.9–5.2), where proteins were found to carry the lowest negative charge as confirmed by the zeta potential measurements. Interfacial properties of acid camel whey and acid bovine whey were preserved at air water interface even after a heat treatment at 90°C. These results confirmed the pronounced foaming and interfacial properties of acid camel whey, even if acid and sweet bovine whey exhibited the highest viscoelastic modulus after heating.

Lajnaf et al. 2018
Acid wheys were obtained after acidification of fresh bovine and camel milks until pH 4.6 and 4.3.

Sweet wheys – rennet enzyme

ABW (acid bovine whey),
SBW (sweet bovine whey),
ACW (acid camel whey)
SCW (sweet camel whey)

Foam capacity: FC (A) and foam stability: FS (B) of camel and bovine wheys; at a concentration of 5 g L\(^{-1}\) as function of the temperature of the heat treatment (70 °C and 90 °C) for 30 min.

http://www.unirio.br/analisedealimentos
Time-dependent changes in interfacial tension $\gamma$ (mN m$^{-1}$) at air/water interface for 0.011 g L$^{-1}$ of camel and bovine whey solution in native conditions (A), after a heat treatment of 70 °C (B) and 90 °C (C) for 30 min (ABW ○ SBW ● ACW Δ and SCW ▲).

Lajnaf et al. 2018
Schematic overview of enzymatic hydrolysis of protein

This induces a higher availability of hydrophobic regions, an decreased average molecular mass (MM), and liberation of ionizable groups.

Schematic overview of the mechanism of water-holding capacity (WHC) by proteins, and the effect of enzymatic hydrolysis

http://www.unirio.br/analisedalimentos

Wouters et al. 2016
Schematic overview of the mechanism of fat-holding capacity (FHC) by proteins, and the effect of enzymatic hydrolysis.

http://www.unirio.br/analisedealimentos

Wouters et al. 2016
Schematic overview of diffusion to adsorption at and stabilization of a hydrophilic–hydrophobic interface by proteins and the effect of enzymatic hydrolysis

<table>
<thead>
<tr>
<th>Native protein</th>
<th>Hydrolyzed protein</th>
<th>Determining factor</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nonsoluble</td>
<td>Nonsoluble</td>
<td>Diffusion speed $v: 0 = v_0 &lt; v_1 &lt; v_2$</td>
</tr>
<tr>
<td>Soluble</td>
<td>Soluble</td>
<td>$MM_{mag} \downarrow$</td>
</tr>
</tbody>
</table>

Affinity towards the interface: $0 = A_0 < A_1 < A_2 < A_3 < A_4$

- Hydrolysis can weaken the protein film
- More electrostatic interactions can improve/worsen interface stability
- More hydrophobic interactions can strengthen the protein film

Wouters et al. 2016

[Image: http://www.unirio.br/analisedealimentos]
Properties

Without an emulsifier  
With an emulsifier

- Hydrophilic molecule
- Hydrophobic molecule
- Emulsifying molecule

http://www.unirio.br/analisedealimentos
References


