Charges on amino acids and proteins

Acidic side chains: glutamate and aspartate

Basic side chains: arginine, lysine & histidine

Glycine @ pH 1

Glycine @ the pKa of the carboxyl group, pH 2.2

Glycine @ pH 7
Glycine @ the pKa of the amino group, pH 9.6

Glycine @ pH 12

The pl

- The pl is the pH where the charge on the amino acid is exactly zero.
- If the amino acid is placed in an electric field at this pH it will not move.
- If you place the amino acid in a solution at a higher pH it will be negative.
- If the solution has a lower pH the amino acid will be positive.

Calculating the pl of Glycine

- Find the range of pHs where the overall charge of the majority of molecules is zero.
- In the case of glycine this is between the pKas 2.2 and 9.6.
- Find the average = 5.9

pH

The pl of glycine
The pI of glycine

\[ \text{pK}_a = 2.2 \]

\[ \text{pK}_a = 9.6 \]

The pI of basic amino acids

\[ \text{pK}_a = 2.2 \]

\[ \text{pK}_a = 9.6 \]

Glycine will be positive (+)

Glycine will be negative (-)
The pI of acidic amino acids

- **pI 2.2**
  - [H+] =
  - [OH-] =

- **pI 4.3**
  - [H+] =
  - [OH-] =

- **pI 3.3**
  - [H+] =
  - [OH-] =

Note: The diagrams illustrate the changes in pH and the corresponding deprotonation states of acidic amino acids.
The \( pI \) of acidic amino acids

\[
\begin{align*}
\text{[H]} & \quad \text{[OH]} \\
pI & = 3.3 \\
\end{align*}
\]

Charges on proteins

- The formation of the peptide bond ‘neutralises’ the carboxyl and amino group charges on the alpha carbon BUT not the charges on the side chains.
- Not all side chains have a charge (\( \text{only lys, arg, his, glu and asp} \))
- Amino acid sequence dependent

The charge on Proteins

\[
\begin{align*}
\text{Peptide bond} \\
\end{align*}
\]

Charges on proteins

- Different proteins have different native charges.
- The overall charge on a protein will depend on:
  - The sequence
  - The pH

Determining the \( pI \) of a protein

- It can be predicted from the difference between the sum of the acidic side chains (asp + glu) and the sum of the basic side chains (lys + arg + his).
- It is determined experimentally by techniques such as isoelectric focusing. The protein is placed in a pH gradient and subjected to an electric field. The protein moves to its \( pI \).

Determining the \( pI \) of a protein

- Those proteins with more acidic residues will have a lower \( pI \)
- Those proteins with more basic residues will have a higher \( pI \)
Estimating the charge of a protein

- What we really want to know is the charge of a protein at a particular pH, like 7.
- How do we use pI data to predict the charge of our protein?
- Acidic residues lower the pI
- Basic residues raise the pI.

Estimating the charge of a protein

At pH 3 the protein will be +ve

At pH 7 the protein will be -ve

**At a particular pH..**

- If the pH of the environment is below (more acidic >[H+]) the pI then the protein will be positive (+ve)
- If the pH of the environment is above (more basic >[OH-]) the pI then the protein will be negative (-ve).

**Relevance to Molecular Biology**

- If you want a protein to interact with nucleic acid (which has a sugar-phosphate -ve backbone) you will need a protein which has positive charges, at least in the area of the protein where the interaction occurs.
- Examples are histones, proteins which pack the DNA. These proteins have high pI's.
Relevance to Molecular Biology

- Proteins that interact with nucleic acids often use lysine and arginine residues to interact with the sugar-phosphate backbone.
- Glutamates are often involved with the interactions with the bases.