

Using molecular marker technology in studies on plant genetic diversity

Protein-based technologies Protein basics

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Protein basics

- ▶ The DNA's base sequence instructs the cell on how to make the different proteins it needs to function as part of the organism in which it exists
- ▶ Proteins have many different functions, both structural and functional
- ▶ Proteins are complicated molecules made by assembling simple building blocks (amino acids) together in a chain (polypeptide chain)

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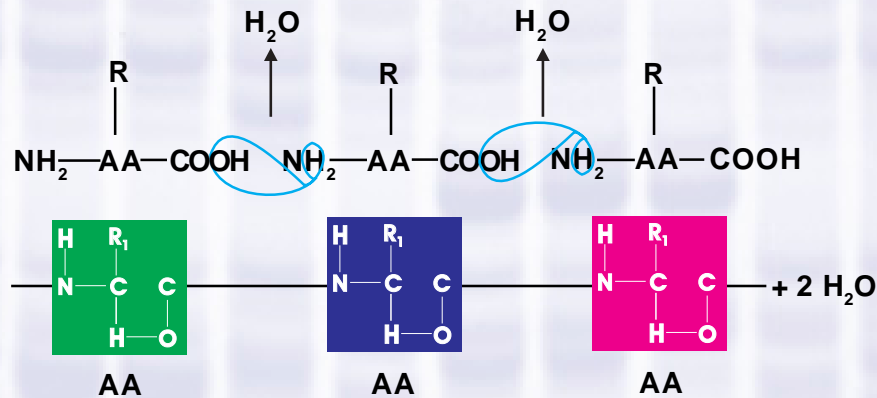
The information carried by the DNA bases translates into proteins. The DNA molecule is copied into a different type of nucleic acid—the RNA or ribonucleic acid. The RNA moves to the 'ribosome', an organelle in charge of making proteins. Every set of three bases in the RNA determines which amino acid is added to the protein molecule in progress. The RNA chain passes through the ribosome until the protein is complete.

This process has been called 'the central dogma', as it is the basis of all biological life. Amino acids are bi-functional organic compounds that contain a basic amino group ($-\text{NH}_2$) and an acidic carboxyl group ($-\text{COOH}$). In proteins, 20 different amino acids are commonly found, varying in function and property according to the nature of the R-group. For instance, in alanine, the R-group is ($-\text{CH}_3$), whereas, in cysteine, it is ($-\text{CH}_2\text{-SH}$).

Protein structures: primary

The primary structure of a protein is the order of amino acids on the polypeptide chain

AA = Amino acid



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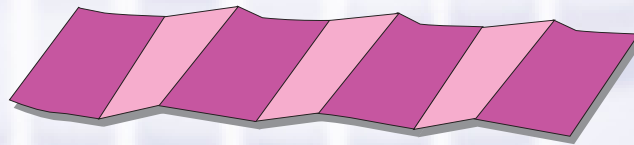
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In proteins, amino acids are joined together in chains by peptide (amide) bonds that form the molecule's backbone. A peptide bond is formed between a basic amino group ($-\text{NH}_2$) on one amino acid and an acidic carboxyl group ($-\text{COOH}$) on another. The general formula of an amino acid is $\text{H}_2\text{N}-\text{CHR}-\text{COOH}$. The R group can be anything from an atom to a complex molecule. The term 'polypeptide' simply refers to a long chain of amino acids.

Once the protein chain has been made, it must fold up properly before it can do its job. The structure and folding of each protein is specific. How the amino acid sequence causes the protein to fold is not yet completely understood. A 'protein' can be made up of one or more separate polypeptides; it can be made of sheets (amino acid chains lining up together) that contain spiral structures. Clearly, the properties of polypeptides and proteins depend on their amino acid composition.

Protein structures: secondary

The secondary structure is the result of local hydrogen bonds being created along the polypeptide backbone



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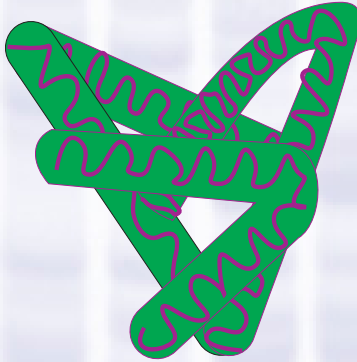
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The secondary structure is the result of local hydrogen bonds being created along the polypeptide backbone. This gives the protein strength and flexibility. Common structures found are:

- *Alpha-helix*, caused by hydrogen-bonding within the polypeptide chain, for example, muscle proteins.
- *Beta-pleated sheet*, caused by hydrogen-bonding between adjacent polypeptide chains, for example, silk fibroin.

Protein structures: tertiary

The tertiary structure results from interactions between the R-groups in a polypeptide: the non-covalent and covalent bonds



Adapted from Griffiths *et al.* 1996

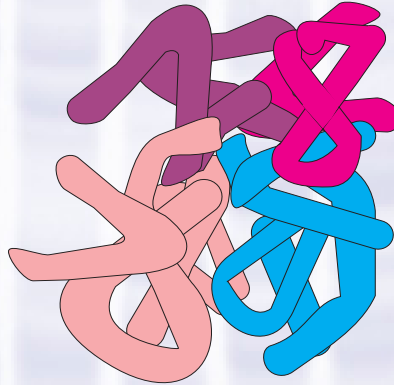
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The tertiary structure results from interactions between the R-groups in a polypeptide, such as non-covalent bonds (hydrogen bonds, ionic bonds, hydrophobic interactions) and weak, covalent bonds (disulphide bonds between cysteine residues).

Protein structures: quaternary

The quaternary structure results from interactions between two or more polypeptide chains to form dimers, trimers, tetramers, etc.



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Sometimes a single polypeptide is sufficient for the protein to be active; we then talk of a protein that acts as a monomer. Often, however, two or more polypeptides need to interact to allow a protein to perform its particular function. If this is the case, we talk of a dimer; and so on through trimers, etc.

The quaternary structure results from interactions between two or more polypeptide chains to form dimers, trimers, tetramers, etc. They are held together by hydrogen bonds, ionic bonds and, less commonly, hydrophobic interfaces and inter-chain disulphide bonds.

Proteins functions

- ▶ The three-dimensional structure of proteins is a direct result of interactions with their internal environment
- ▶ Diversity of protein function is a result of the complexity of protein structure

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The three-dimensional structure that proteins have is a direct result of interactions with its internal environment. As a consequence, knowing how proteins are structured tells us a lot about how they perform their tasks in the cell.

For instance, in aqueous environments, the hydrophobic R-groups are positioned towards the protein's interior. Changes in temperature or pH can interfere with the non-covalent bonding, causing disruption in the three-dimensional structure and loss of activity. This process is called 'denaturation'. Denatured proteins can also clump together to become insoluble in a process called coagulation.

The diversity of protein function, as facilitated by the complexities of protein structure, include (with examples):

- *Structural* (collagen, muscle fibres)
- *Storage* (wheat gliadins, barley hordeins)
- *Enzymes* (hydrolases, transferases, isomerases, polymerases, ligases)
- *Transport* (oxygen transfer with haemoglobin)
- *Messengers* (insulin and certain other hormones)
- *Antibodies* (proteins that bind to specific foreign particles)
- *Regulation* (proteins involved in regulating DNA synthesis)

Enzymes: description

- ▶ Enzymes are a particular type of protein that act as catalysts
- ▶ Each enzyme is highly specific with regard to the type of chemical reaction that it will catalyse and to the substances (called substrates) on which it will act

Enzymes: allozymes and isozymes

The multiple forms that enzymes take fall into two main classes according to how they are coded:

- Allozymes—enzymes coded by different alleles at one gene locus
- Isozymes—enzymes coded by alleles at more than one gene locus

Usually, the term 'isozymes' refers to both classes

Polymorphisms are generated by changes in the amino acids that may result in changes in the primary structure of the enzyme.

In summary

- ▶ Proteins are the primary product of genes
- ▶ Proteins may have different structures, which are closely related to the tasks they perform in the cell
- ▶ Enzymes are a particular type of protein that act as catalysts
- ▶ Enzymes may have multiple forms, belonging to either allozymes or isozymes

By now you should know

- ▶ What a protein is
- ▶ The mechanisms involved in shaping proteins into different structures
- ▶ What an enzyme is
- ▶ The difference between allozymes and isozymes

Basic reference

Griffiths, A.J.F., J.H. Miller, D.T. Suzuki, R.C. Lewontin and W.M. Gelbart. 1996.
The nature of the gene. Pp. 345-358 *in* An Introduction to Genetic Analysis (6th
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Next

Protein-based technologies **Isozymes**

- ▶ DNA-based technologies
- ▶ Complementary technologies
- ▶ Final considerations
- ▶ Glossary