

ORGANIC CHEMISTRY

Score Advance : Guided Revision Plan - Question Bank- (02) TOPIC : TEST OF AMINO ACIDS AND PROTEINS

Test Of Amino acids and Proteins

1. Biurate Test 2. Nin hydrin Test 3. Xanthoproteic test

4. Sakaguchi test 5. Millon's Test

1. Biurate Test

Addition of a very dilute solution of CuSO₄ to an alkaline solution of a protein is done. A positive test is indicated by the formation of a pink violet to purple violet color.

The name of test is derived from a specific compound, biuret, which gives a positive test with this reagent

When a protein reacts with copper (II) sulfate (blue), the positive test is the formation of a violet colored complex.

The biuret test works for any compound containing two or more of the following groups.

2. Nin hydrin Test

The ninhydrin test is a test for amino acids and proteins with a free –NH₂ group.

Amino acids are detected by ninhydrin test. All amino acids give violet - coloured product with ninhydrin (triketo hydroindene hydrate) except proline and 4 - hydroxy proline, which gives yellow colour with it.

When such an –NH₂ group reacts with ninhydrin, a purple-blue complex is formed.

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The same violet coloured dye forms from all α - AA's with 1° amino groups because only their nitrogen is incorporated into it. The 2° amines proline and 4 - hydroxyproline give different adducts that absorb light at a different and thus have a different yellow colour.

3. Xanthoproteic test

This test is used for aromatic amino acids which give positive result from other amino acids. Such as tyrosine, and tryptophan gives Xanthoproteic test, phenyl alanine does not respond with this test.

Principle:

Xanthoproteic test is used to detect amino acids containing an aromatic nucleus (tyrosine, tryptophan and phenylalanine) in a protein solution which gives yellow color nitro derivatives on heating with conc. HNO₃. The aromatic benzene ring undergoes nitration to give yellow colored product. Phenylalanine gives negative or weakly positive reaction though this amino acid contains aromatic nucleus because it is difficult to nitrate under normal condition. On adding alkali to these nitro derivative salts, the color change for yellow to orange.

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4. Sakaguchi test

The Sakaguchi test is a chemical test used for detecting the presence of arginine in proteins Sakaguchi reagent consists of α -Naphthol and a drop of sodium hypobromite. The guanidine group in arginine reacts with Sakaguchi reagent to form a red-coloured complex.

Red compound

5. Millon's Test

Millon's reagent is an analytical reagent used to detect the presence of soluble proteins. A few drops of the reagent are added to the test solution, which is then heated gently. A reddish-brown coloration or precipitate indicates the presence of tyrosine residue which occur in nearly all proteins

Tyrosine Here
$$R = -CH_2CH(NH_2)COOH$$
.

 $R \longrightarrow OH \longrightarrow R \longrightarrow OH$
 $N=O$
 $N=O$

reddish-brown colored complex

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Sanger's Method of sequencing of Amino acids of polypeptide chain

It is N-terminal amino acid analysis

Sanger's reagent (1-fluoro-2,4-dinitrobenzene)

- 1. React the peptide with a reagent that will selectively label the N-terminal amino acid.
- 2. Hydrolyse the protein.
- 3. Determine the amino acid by chromatography and comparison with standards.

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QUESTION BANK 2 ADDITIONAL QUESTION

1. Vitamin **Biochemical name Deficiency disease** (a) Vitamin-B₁ (P) Ascorbic acid (i) Pernicions Anemia

- (b) Vitamin-D (Q) Thiamine
- (c) Vitamin-B₁₂ (R) Calciferol (iii) Rickets
- (d) Vitamin C (S) Cynocobalamine (iv) Beri-Beri

Correct match of following is -

- (A) a Q, iv
- (B) b-P, iii
- (C) c S, i

(ii) Scurvy

- (D) d-P, ii
- 2. If the Molecular weight of a pentapeptide chain is X, Find out the value of total molecular weight of all Amino acids after complete acidic hydrolysis of that pentapeptide chain.
 - (A) X + 90
- (B) X + 72
- (C) X + 18
- (D) X
- **3.** Chargaff's rule state that DNA from any cell of any organism should have a 1:1 stoichiometric ratio of pyridine and purine bases in double helix. Find out the correct statement(s) using Chargaff's rule? (A) Double helix DNA contain, Adenine + Guanine = Thymine + Cytosine
 - (B) $\frac{\text{Purine}}{\text{Pyrimidine}} = 1$
 - (C) $\frac{\text{Adenine} + \text{Guanine}}{\text{Thymine} + \text{Cytosine}} = 1$
 - (D) Cell contains adenine = Thymine and Cytosine = Guanine
- 4. Which of the following compound gives Biurate test

$$(B) \ H_2N-C-NH \ (-CH_2)_3- \begin{matrix} H & O & O \\ I & II \\ C-C-NH-CH_2-C-OH \\ NH_2 \end{matrix}$$

- (C) $H_{2}N CH_{2} CH_{3} NH_{4}$
- (D) Insulin hormone
- 5. α -Helix sheet and β - Pleated structure of protein is
 - (A) Primary Structure

(B) Secondary Structure

(C) Tertiary Structure

(D) Quaternary Structure



6	Which of the following is Sanger reagent?			
	(A) 2,4-Di-nitro flurobenzene		(B) Phenyl isocyanide	
	(C) 2,4-Di-nitro chlororbenzene		(D) 2,4-Di-nitro-iodobenzene	
7.	Which of the following Amino acid gives +ve Xanthoproteic test.			
	(A) Tyrosine	(B) Tryptophan	(C) Histidine	(D) Cystein
8.	Which of the following Amino acid gives +ve Sakaguchi test.			
	(A) Glycine	(B) Phenyl Alanine	(C) Aspartic acid	(D) Arginine
9.	 A polypeptide chain gives reddish brown colored complex with Millon's reagent, It indicates (A) Alanine is present in polypeptide chain. (B) Phenyl alanine is present in polypeptide chain (C) Tyrosine is present in polypeptide chain (D) Methionine is present in polypeptide chain 			
10.	Which of the following Amino acid do not gives Ninhydrin test.			
	(A) Glycine	(B) Alanine	(C) Lysine	(D) Proline.

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