Tests for Carbohydrates

1. **Tollen'ss test**

   All reducing sugars give silver mirror

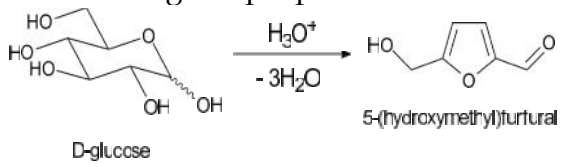
2. **Barfoed's test**

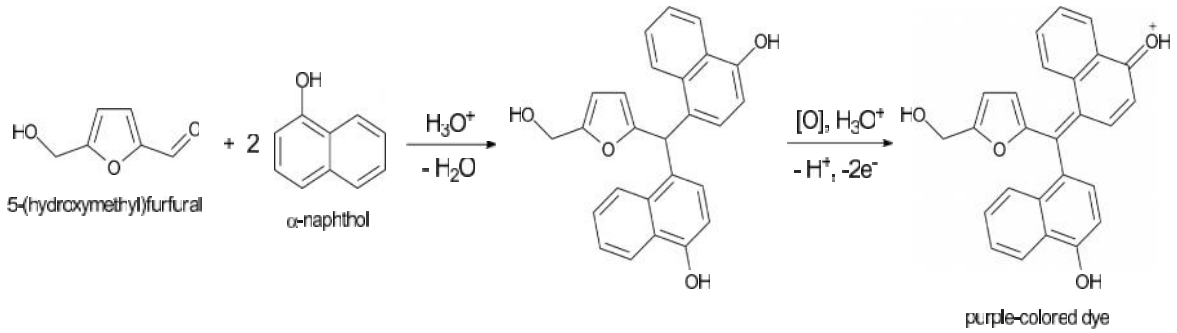
   Reagent: Copper(II) acetate

   Similar to Benedict's and Fehling's test it gives Red ppt with all reducing sugars

3. **Molisch's Test**

Monosaccharides with 5 carbon or more and all oligo and polysaccharides give this test. Saccharide is treated with an alcoholic solution of α-naphthol and con.H_2SO_4 to give purple colouration. Pentoses and hexoses get dehydrated to give furfural and 5-(hydroxymethyl) furfural which gives purple colouration with α-naphthol.



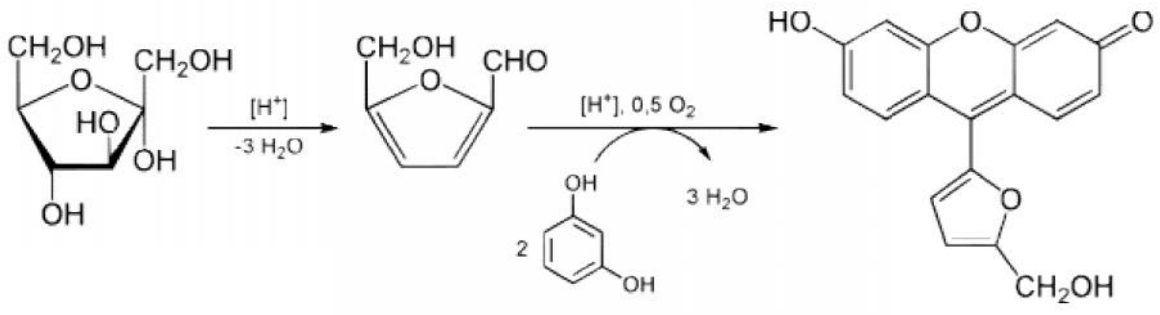


4. **Seliwanoff's test**

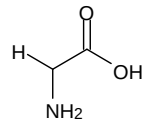
   Used to distinguish ketoses and aldoses

   Reagent: Resorcinol + HCl

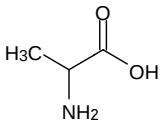
Ketoese get dehydrated faster and hence they give the test faster (Cherry red), Aldoses dehydrated slowly and give colour (Blue-green or peach colour) slowly.



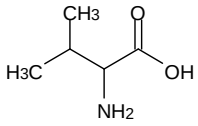
20 Common Amino Acids



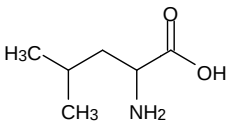
Glycine **(Gly)**



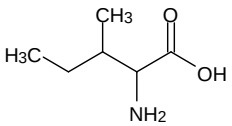
Alanine **(Ala)**



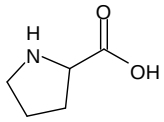
Valine **(Val)**



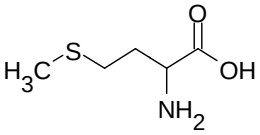
Leucine **(Leu)**



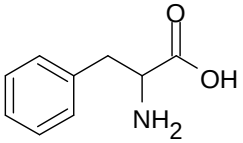
Isoleucine **(Ile)**



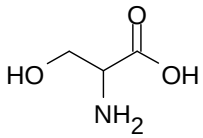
Proline **(Pro)**



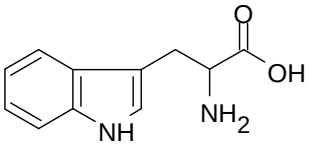
Methionine **(Met)**



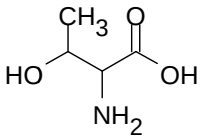
Phenylalanine **(Phe)**



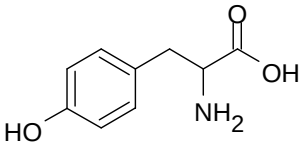
Serine **(Ser)**



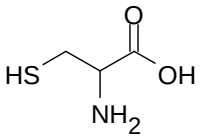
Tryptophan **(Trp)**



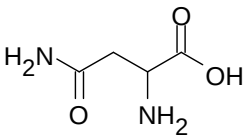
Threonine **(Thr)**



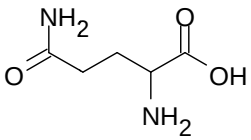
Tyrosine **(Tyr)**



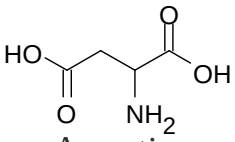
Cysteine **(Cys)**



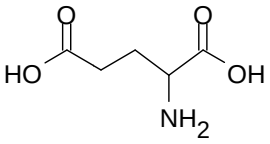
Asparagine **(Asn)**



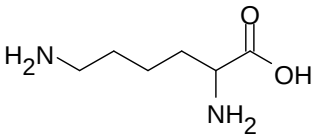
Glutamine **(Gln)**



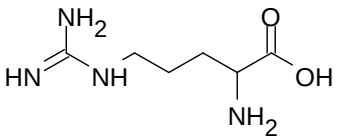
Aspartic Acid **(Asp)**



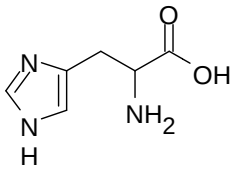
Glutamic Acid **(Glu)**



Lysine **(Lys)**

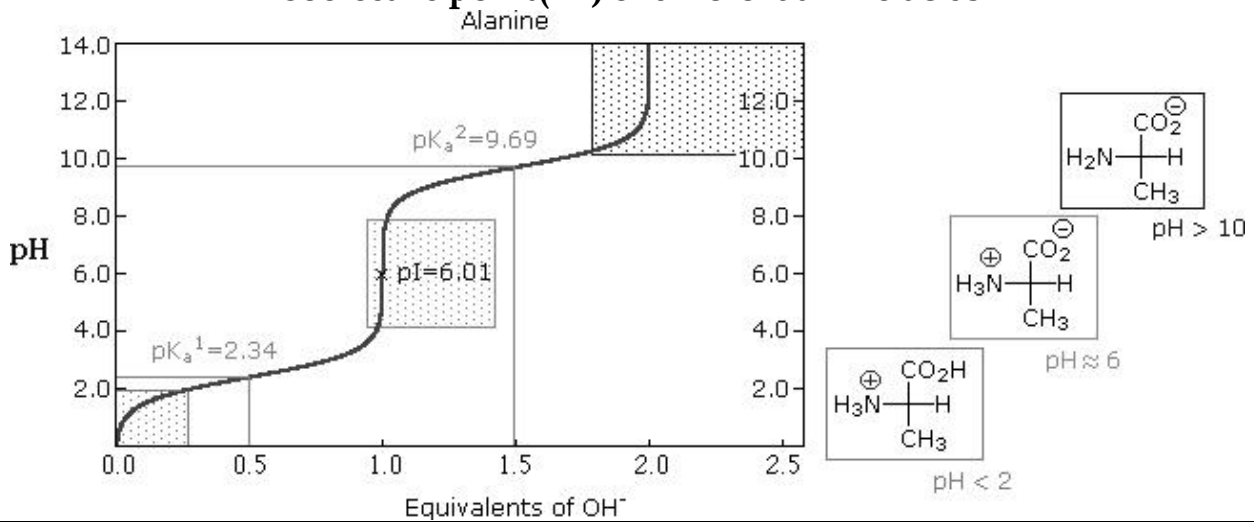


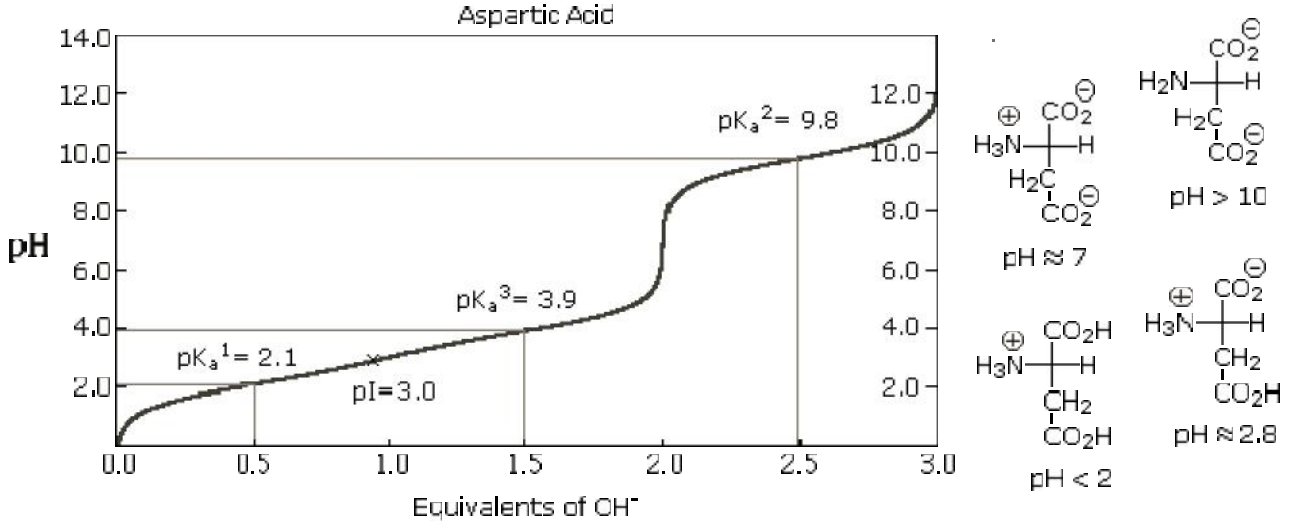
Arginine **(Arg)**

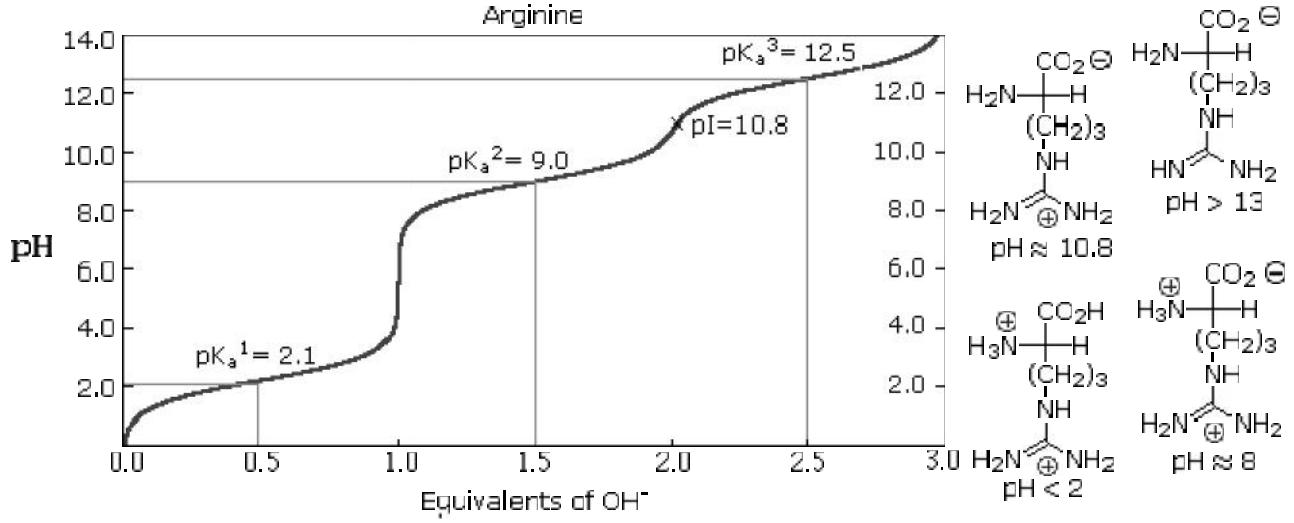


Histidine **(His)**

Isoelectric point(P^I) of different amino acids



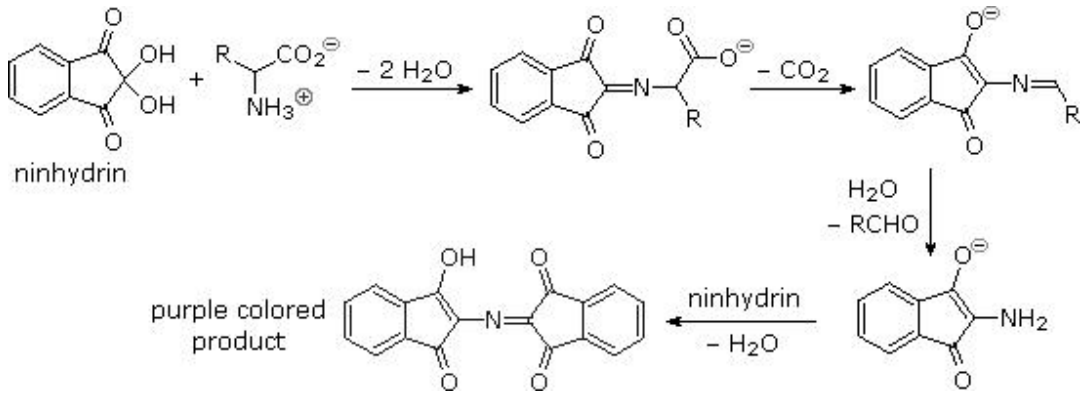




Tests for Amino acids/ Proteins

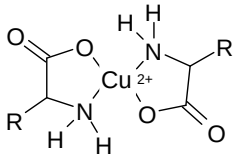
1. The Ninhydrin Reaction

In addition to these common reactions of amines and carboxylic acids, common alpha-amino acids, except proline, undergo a unique reaction with the triketohydrindene hydrate known as ninhydrin. Among the products of this unusual reaction (shown on the left below) is a purple colored imino derivative, which provides as a useful color test for these amino acids, most of which are colorless. A common application of the ninhydrin test is the visualization of amino acids in **paper chromatography**.



2. Biuret test

Peptide bonds complex with Cu^{2+} just like Biuret ($NH_2CONHCONH_2$) to give blue/violet colouration when an alkaline solution of the peptide is treated with $CuSO_4$ solution. There should be minimum two peptide groups for the complex formation and hence dipeptides do not give this reaction.



3. Xanthoproteic test

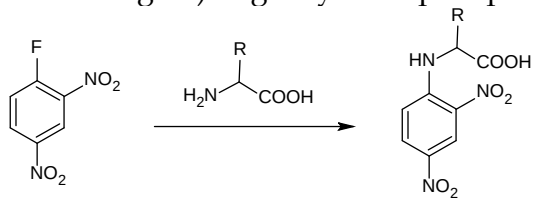
When treated with con.HNO_3, amino acids having benzene ring get nitrated to give yellow colouration when turns orange on treatment with NaOH.

4. Millon's test

When treated with Con.HNO_3 and Hg(II) nitrate Tyrosine (as it contains a phenolic group) gets nitrated to give a white precipitate which turns red on heating.

5. Sanger's test

Amino acids act as nucleophiles and substitute F in 2,4-dinitroflourobenzene (DFNB – Sanger's reagent) to give yellow precipitate.



6. Sodium nitroprusside test

Cysteine complexes with sodium nitroprusside with its –SH group to give violet coloured complex.

The following table gives the pKa values for the α-carboxylic acid group, the α-amino group, and any ionizable side chains.

Amino Acid pKa Values

Amino Acid	~-carboxylic acid	~-amino	Side chain	Amino Acid	~-carboxylic acid	~-amino	Side chain
Alanine	2.35	9.87		Lysine	2.18	8.95	10.53
Arginine	2.01	9.04	12.48	Methionine	2.28	9.21	
Asparagine	2.02	8.80		Phenylalanine	2.58	9.24	
Aspartic Acid	2.10	9.82	3.86	Proline	2.00	10.60	
Cysteine	2.05	10.25	8.00	Serine	2.21	9.15	
Glutamic Acid	2.10	9.47	4.07	Threonine	2.09	9.10	
Glutamine	2.17	9.13		Tryptophan	2.38	9.39	
Glycine	2.35	9.78		Tyrosine	2.20	9.11	10.07
Histidine	1.77	9.18	6.10	Valine	2.29	9.72	
Isoleucine	2.32	9.76		Lysine	2.18	8.95	10.53
Leucine	2.33	9.74					