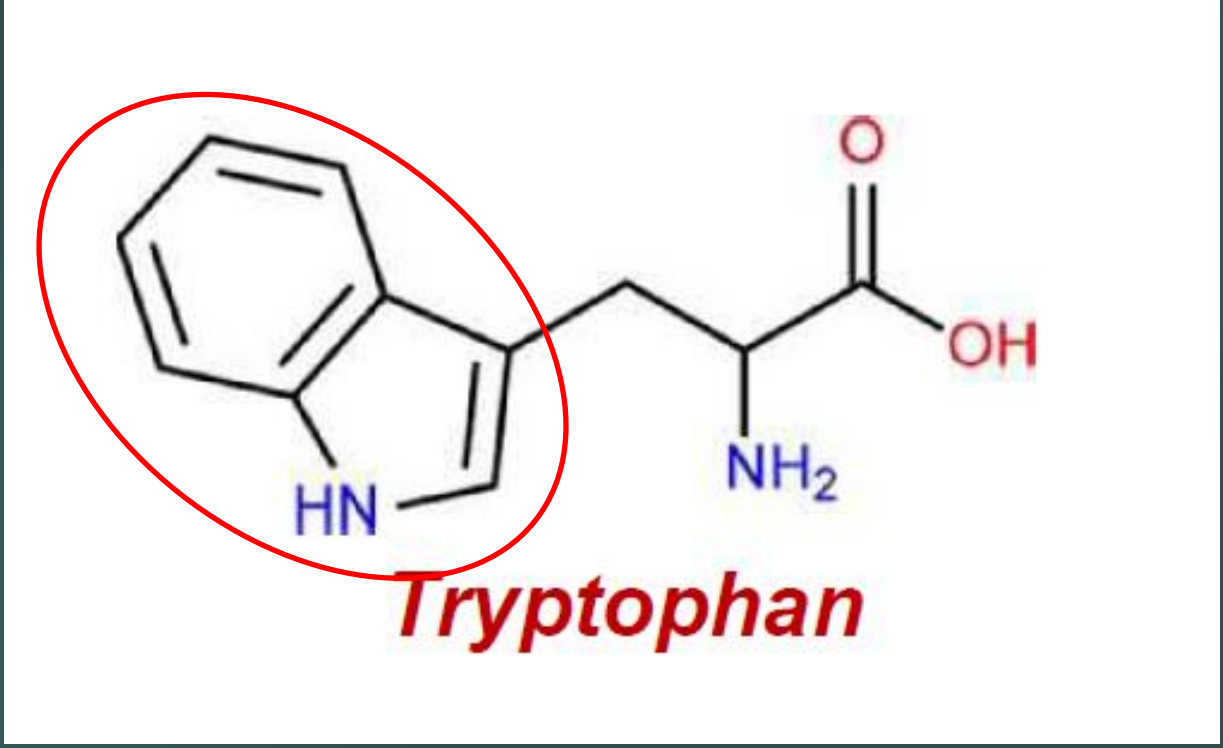
Proteins

Specific Tests for Proteins

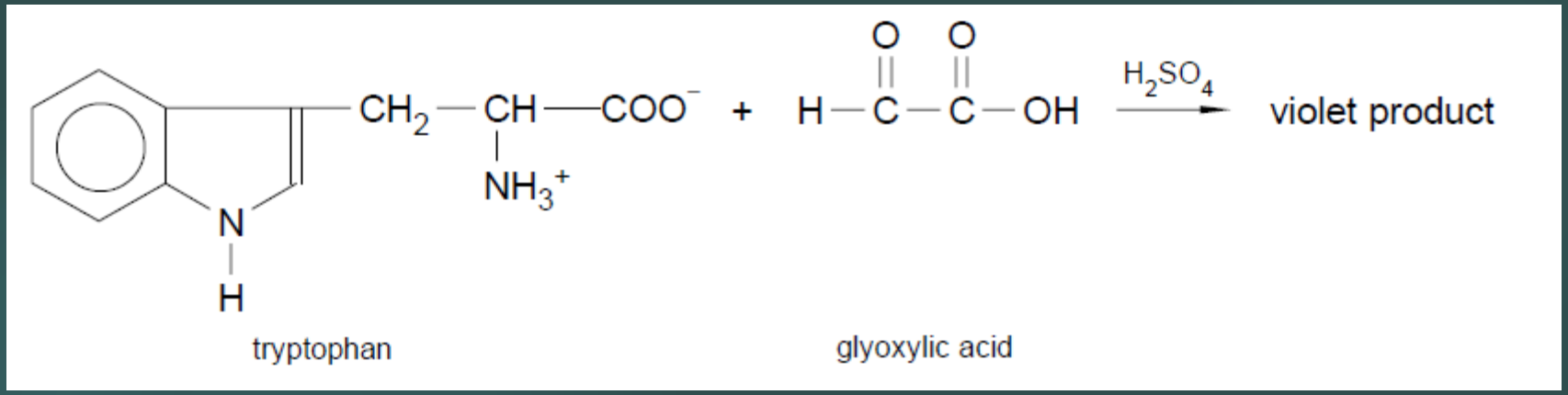
Hopkins Cole Test



Hopkins Cole Test

- **Principle**
- In the presence of strong acid (H_2SO_4) indole group of tryptophan reacts with glyoxylic acid (Hopkins Cole reagent) to form violet colour ring

Hopkins Cole Test

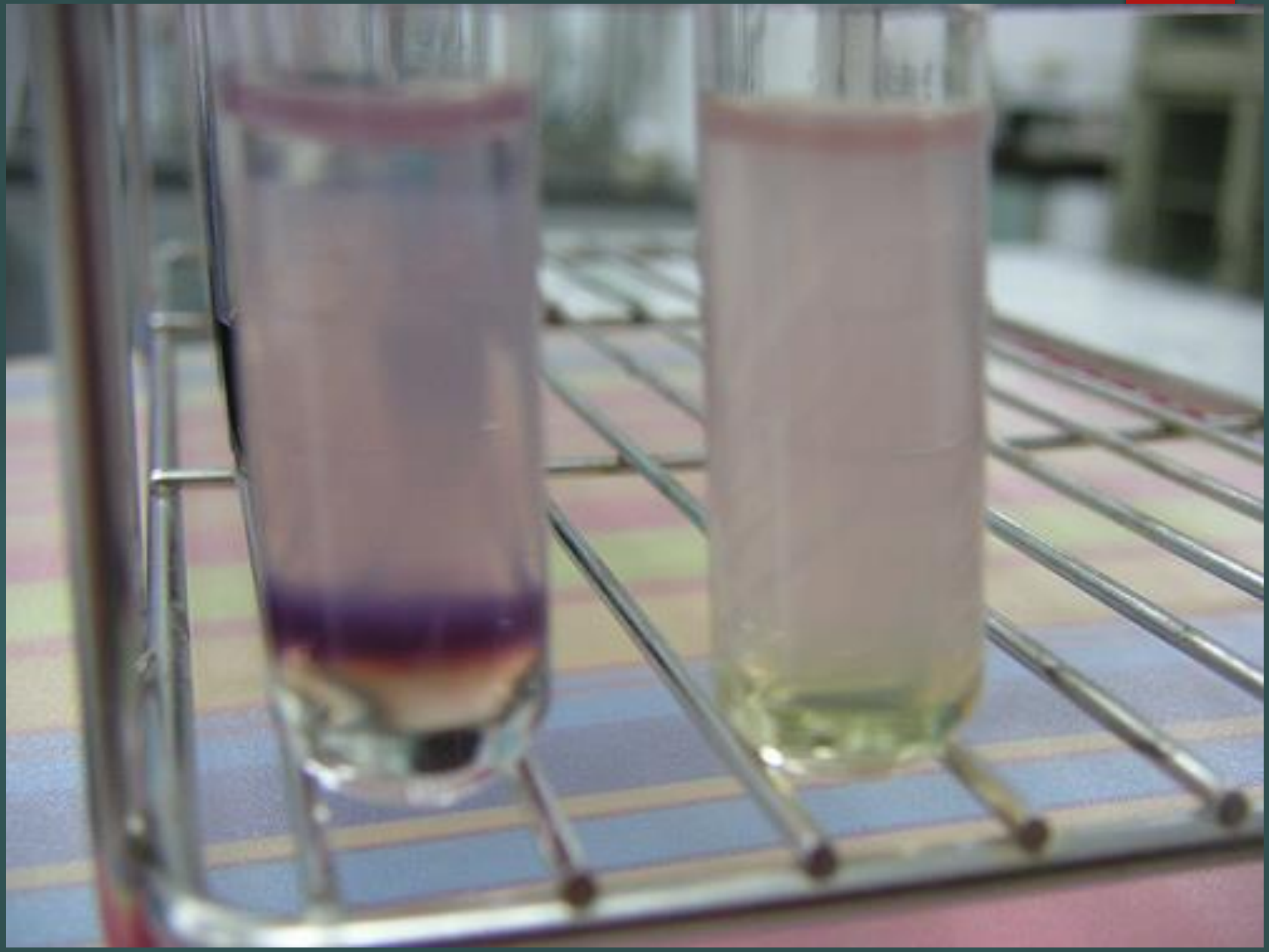


Hopkins-Cole Test

- Procedure

1. 1 ml of each protein
2. 1 ml of Hopkins-cole reagent
3. Mix
4. 20 drops of H_2SO_4 slowly on the wall
5. Do not mix
6. A purple violet ring will form

- **Note**
- All proteins will give positive result with Hopkin-Cole test except gelatin which contain little amount of tryptophan .



Esbasch test

- All protein contain different amino acids

a) Acidic : ?

b) Basic : ?

- Total charge of protein depends on the **composition** of protein and **pH of the solution**

Esbasch test

- **Isoelectric point**
- is the pH at which the positive charge of protein is equal to the negative charge , so the net charge of protein is zero , and the protein precipitate as insoluble salts

Esbasch test

- **Principle**
- Picric acid in Esbasch reagent can bring most proteins to their isoelectric point , and therefore precipitate them as yellow gelatinous ppt.

- Peptone has its isoelectric point in the basic region so cant be precipitated by Esbasch reagent.

Esbasch test

- **Procedure**

1. Add 1 ml of each protein in a test tube
2. Add 1 ml of Esbach reagent
3. Mix
4. A yellow ppt is formed in Albumin , Casein and Gelatin tubes as positive results
5. Peptone tube remains clear as a negative result

Report

Test	A	P	C	G	Conclusion
Hopkins-Cole Test					
Esbasch Test					

Report

Test	A	P	C	G	Conclusion
Hopkins-Cole Test	+				
Esbasch Test					

Report

Test	A	P	C	G	Conclusion
Hopkins-Cole Test	+	+			
Esbasch Test					

Report

Test	A	P	C	G	Conclusion
Hopkins-Cole Test	+	+	+		
Esbasch Test					

Report

Test	A	P	C	G	Conclusion
Hopkins-Cole Test	+	+	+	–	
Esbasch Test					

Report

Test	A	P	C	G	Conclusion
Hopkins-Cole Test	+	+	+	–	
Esbasch Test	+				

Report

Test	A	P	C	G	Conclusion
Hopkins-Cole Test	+	+	+	–	
Esbasch Test	+	–			

Report

Test	A	P	C	G	Conclusion
Hopkins-Cole Test	+	+	+	–	
Esbasch Test	+	–	+		

Report

Test	A	P	C	G	Conclusion
Hopkins-Cole Test	+	+	+	–	
Esbasch Test	+	–	+	+	

- Thanks