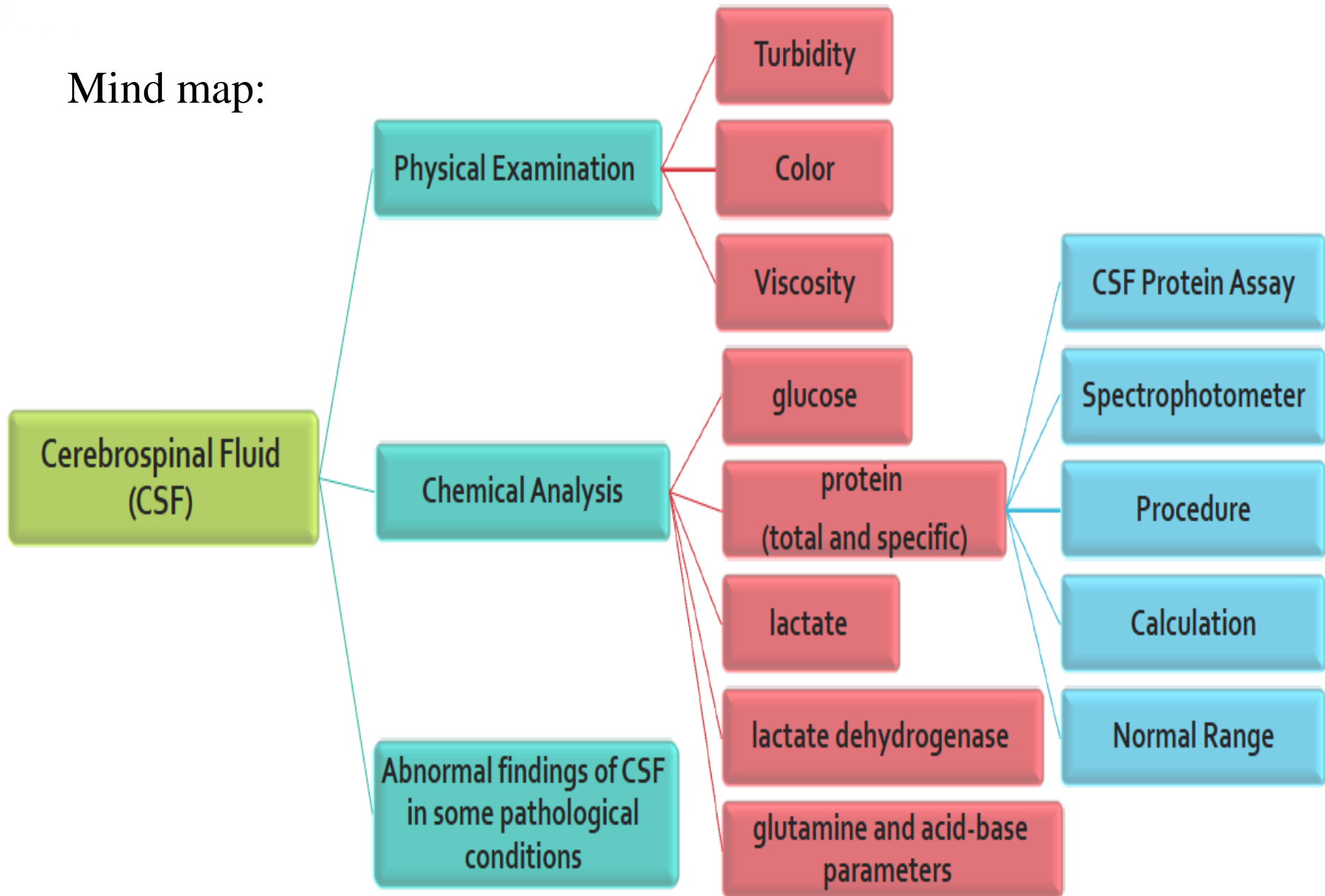Biochemistry Team 434

Cerebrospinal Fluid (CSF) Analysis for total protein

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Mind map:

Glucose and protein estimations should be performed as soon as possible after drawing the CSF specimen

If testing is to be delayed, the specimen should be frozen at – 20^0 C.

The specimen should be delivered to the laboratory immediately after collection

CSF sample

Remember !!

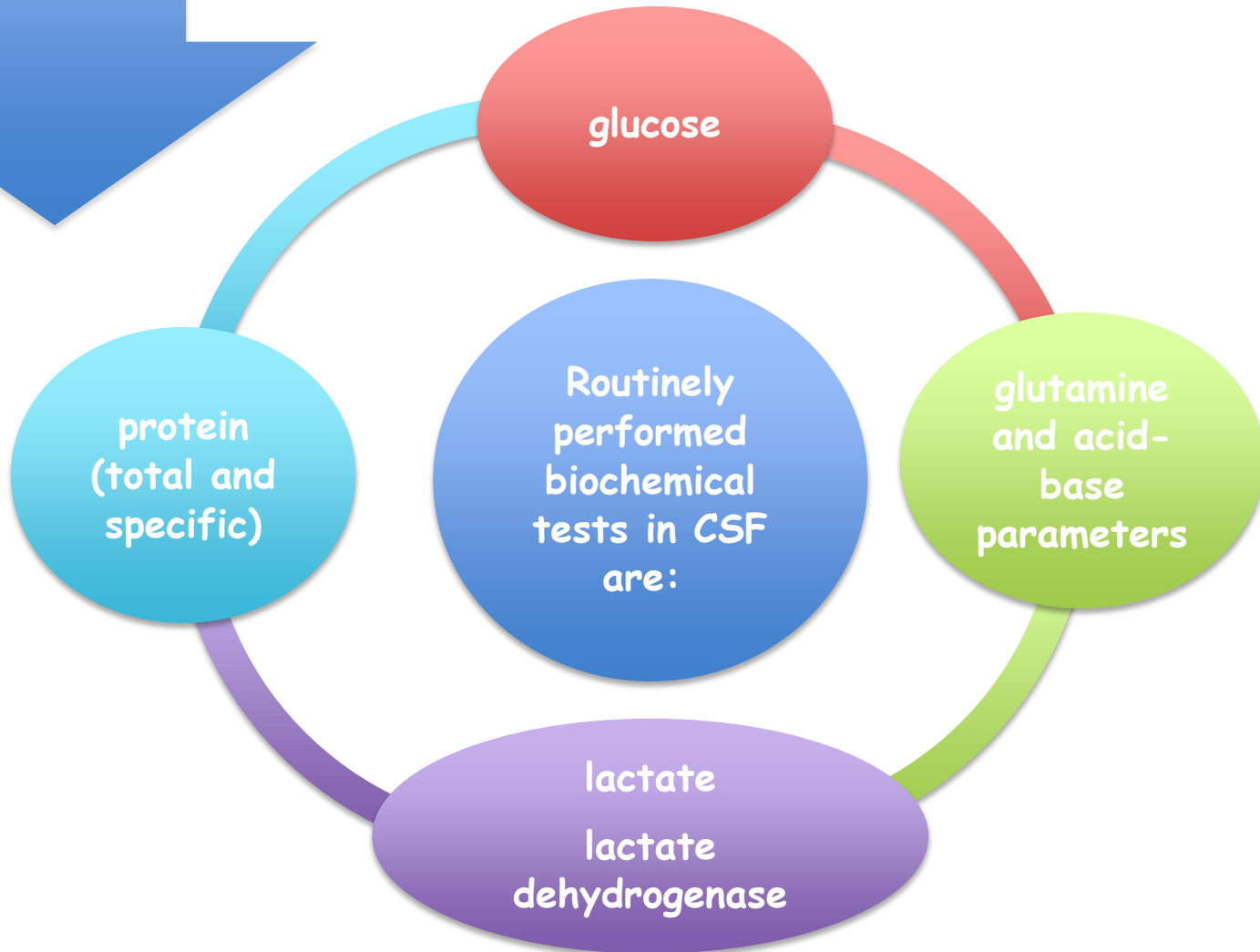**Before any analysis, the fluid should be centrifuged to avoid contamination by cellular elements**

CSF is the most precious biological material. Often, only small volumes of CSF are available for analysis due to difficulty in collection; hence handle this with care

The specimen may contain virulent organisms, so strict safety precautions should be followed.
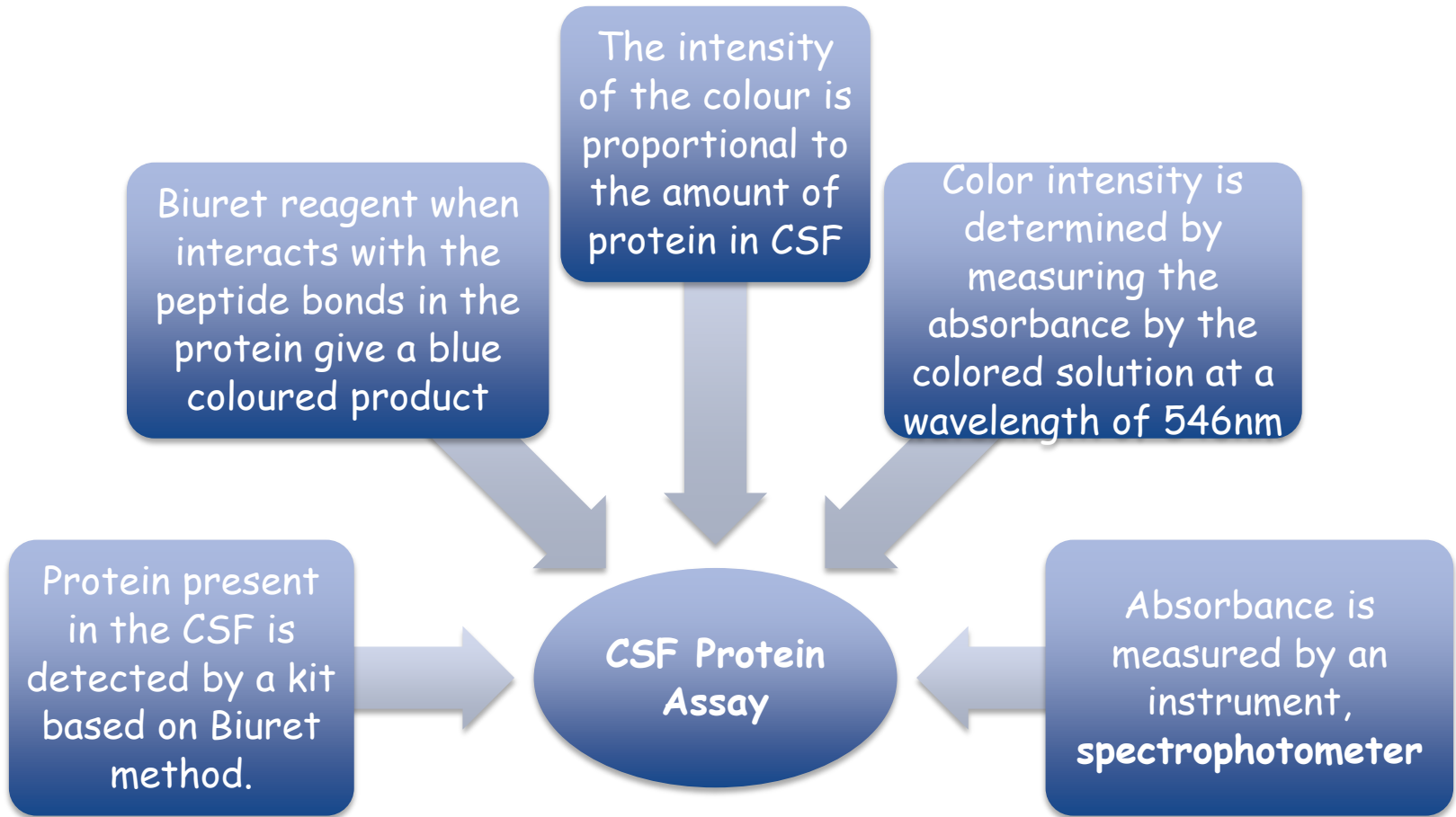
Physical examination

Physical Examination	Normal	Abnormal
Turbidity	Clear	Cloudy/ turbid- may indicate the presence of white, or red blood cells, microorganisms, or an increase in protein level
Color	Colorless	Yellow, orange-brown, or red- may indicate the presence blood
Viscosity	should have the same consistency as water	CSF may be seen in patients Thicker with certain types of cancers or infections (meningitis)

Chemical Analysis

Routinely performed biochemical tests in CSF are:

- glucose
- protein (total and specific)
- glutamine and acid-base parameters
- lactate
- lactate dehydrogenase

Abnormal findings of CSF in some pathological conditions

Parameter	Condition			
	Bacterial Meningitis	**Tuberculous Meningitis**	Viral Meningitis	**Brain Tumor**
Protein	↑ ↑	↑ ↑	Normal	↑
Glucose	↓ ↓	↓ ↓	Normal or slightly ↓	↓
Chlorides	↓ ↓	↓ ↓	Normal or ↓	Normal or ↓

Biuret reagent when interacts with the peptide bonds in the protein give a blue coloured product

The intensity of the colour is proportional to the amount of protein in CSF

Color intensity is determined by measuring the absorbance by the colored solution at a wavelength of 546nm

Protein present in the CSF is detected by a kit based on Biuret method.

CSF Protein Assay

Absorbance is measured by an instrument, **spectrophotometer**

Spectrophotometer:

Most of visible spectrophotometers are composed of:

- Light source which works with visible wavelengths (400-700 nm)
- Monochromator filter for choosing desired wavelength
- Sample holder (cuvette)
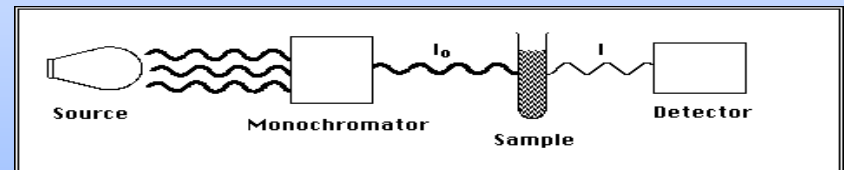- Detector
- Meter or recorder

Source   Monochromator I_0 Sample I Detector

Figure I

	Test	Standard	Blank
Reagent	2 ml	2 ml	2 ml
CSF sample	40 μl	-	-
Standard	-	40 μl	-
H$_2$O	-	-	40 μl

Normal Range

Normal reference values for CSF protein:
15 - 45 mg/dL   (0.1 - 0.4 g/L)

1. Mix and incubate for 15 minutes at room temperature
2. Measure absorbance at 546 nm

Calculation

Protein conc (g/L) =

$$\frac{\text{Abs of sample}}{\text{Abs of standard}} \times \text{Conc of standard (60 g/L)}$$

Case scenario 1 (Bio + Micro)

A 12-years old child was brought to the emergency department in KKUH by his mother. She said that her child has been suffering form fever and headache symptoms for the last two weeks, and developed a stiffness in the neck recently.

A CSF sample was drown from the patient and sent to the microbiology and biochemistry labs.

The microbiology lab results: The culture reveals the growth of ….. Etc.

Biochemistry lab results for analysis of CSF:

Parameter	Result	Normal state
Predominant cell	Neutrophils	Nill
Protein	100 mg/dl	15-45 mg/dL
Glucose	35 mg/dl	50 - 80 mg/dL

Q1) What is the most likely diagnosis?
Bacterial meningitis.

Q2) What other relevant finding is expected to be seen in such condition ?
Decreased Cl levels.

433 Biochemistry team

Case scenario 2

A 50-years old male presented to the ER with excruciating headache.
He said "It is the worst headache I have ever had in my life".
The diagnosis of subarachnoid hemorrhage was suspected, the GP ordered a sample of the cerebrospinal fluid (CSF) to be drawn for examination.
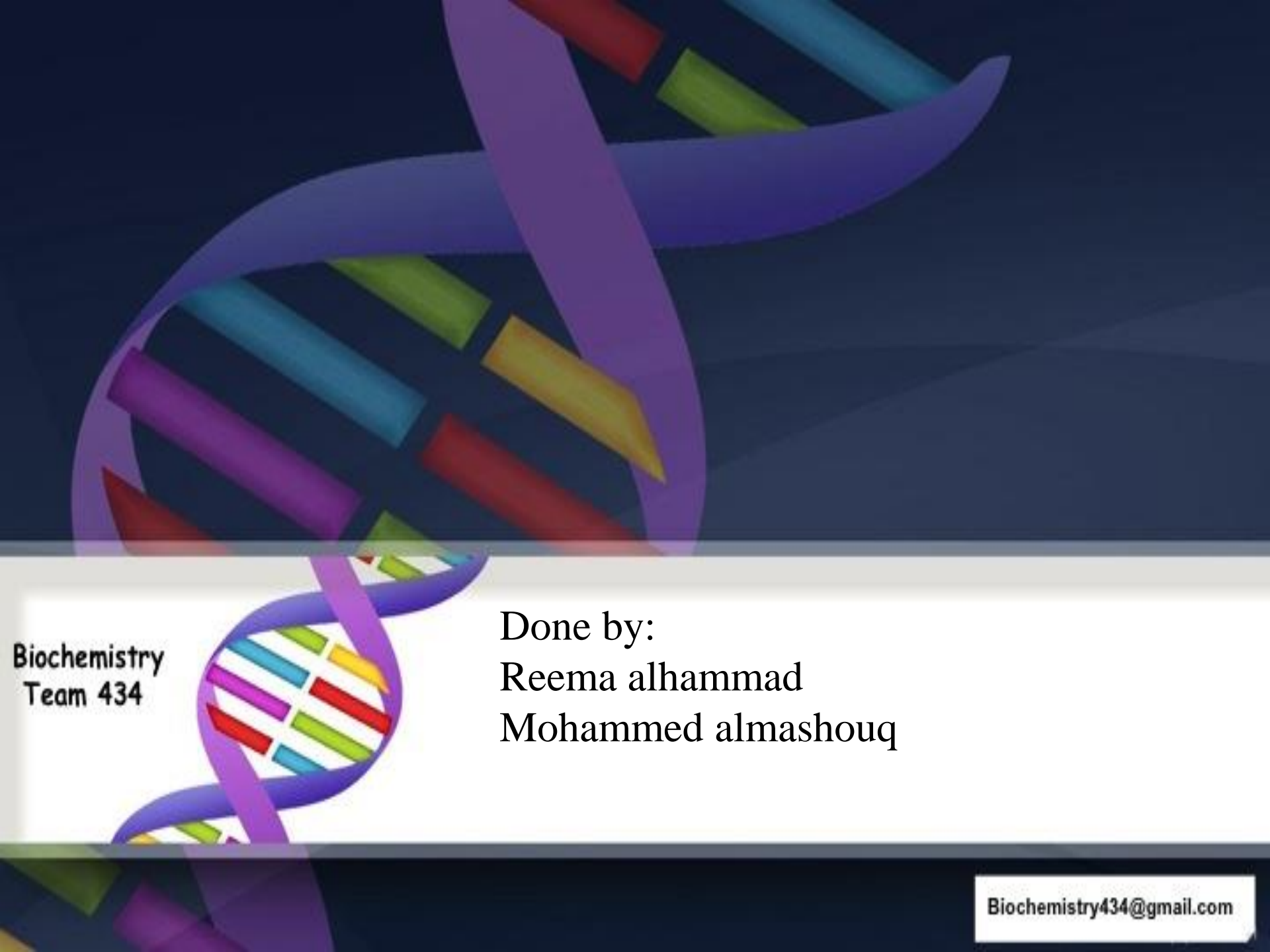
The result of physical examination are shown below :

Parameter	Result	Normal
Appearance	Cloudy	Clear
Color	Yellow	Colorless
Viscosity	As water	As water

Q1) The yellowish CSF color is referred as ? What is the pathogenesis underlying it ?
Xanthochromia , due to hemoglobin pigment breakdown and RBCs lysis.
Q2) What is the level of spinal cord in which the CSF sample can be drawn ?
The interspace L3-L4.

Biochemistry
Team 434

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