Date……..  Name……  Group ……

## Lab report from practical lesson in biochemistry

## Topic: Spectrophotometry

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## Task 1: Spectrophotometric estimation of total serum protein with the biuret method

**Principle:**

**Results:**

Absorption spectrum of colored complex of $Cu^{2+}$ with peptidic bonds:

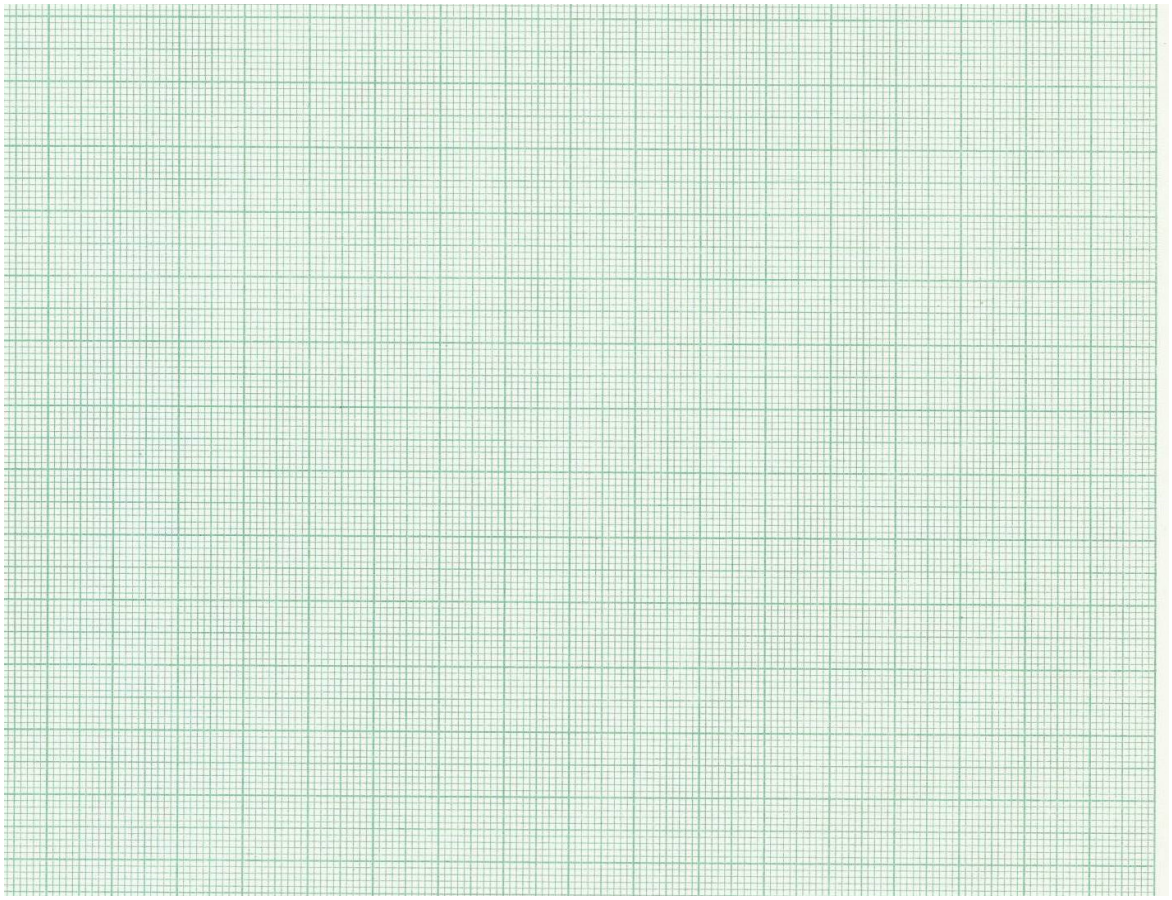
Absorption maximum at……nm

Fill in the measured absorbances and calculate the calibration factors for all standards and then the average calibration factor:

| | **Tube 1** | **Tube 2** | **Tube 3** | **Tube 4** | **Tube 5** | **Tube 6** |
|---|---|---|---|---|---|---|
| | Standard 1 20 g/l | Standard 2 40 g/l | Standard 3 60 g/l | Standard 4 80 g/l | Standard 5 100 g/l | Unknown sample |
| Absorbance | | | | | | |
| Calibration factor | | | | | | |

Average calibration factor F: ……

Calibration graph for estimation of total protein with the biuret method:



**Evaluation:**

1. Graphically from the calibration graph (indicate in graph above)
2. Calculation using the average calibration factor:

3. Calculation using one standard method (use the Standard No. 3):

**Conclusion – summary of results:**

| Evaluation method | Concentration (g/l) |
|---|---|
| Calibration graph | |
| Calibration factor | |
| One standard | |

**Discussion:**
Compare the advantages and disadvantages of the three different methods of evaluation.
Which one do you think is the most accurate?

## Task 2: Estimation of catalytic activity of lactate dehydrogenase in serum by means of Warburg optical test

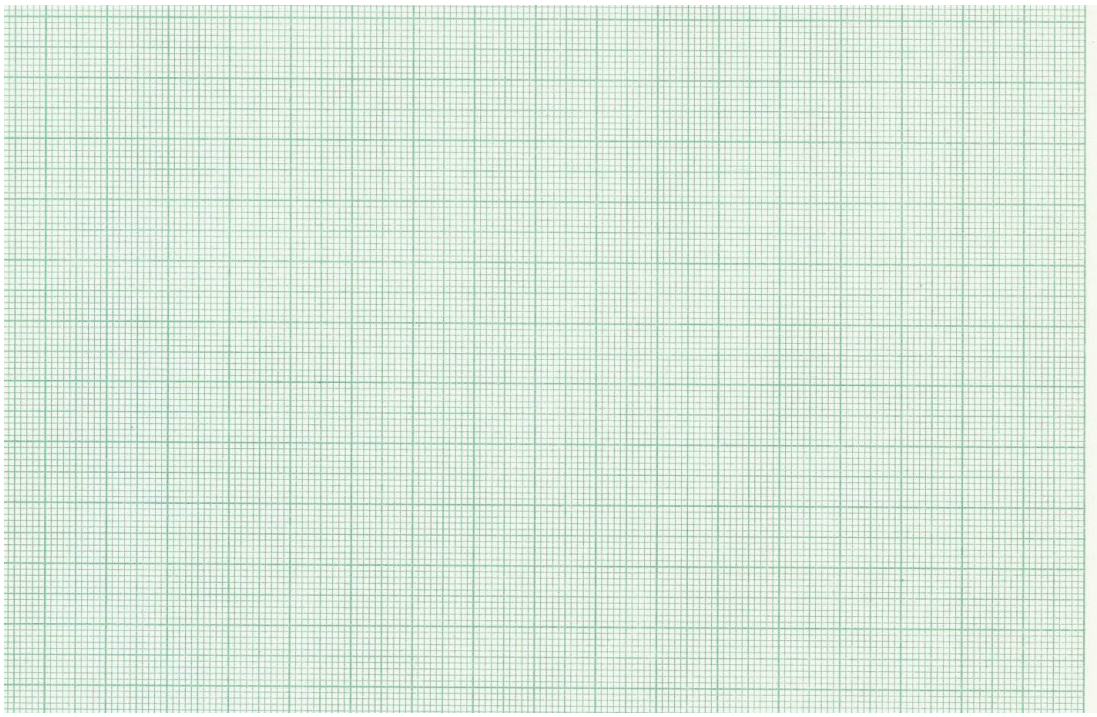
**Principle:**

**Results:**

| Minute | $A_{340}$ | $\Delta A_{340}$ |
|---|---|---|
| 1 | | – |
| 2 | | |
| 3 | | |
| 4 | | |
| 5 | | |
| 6 | | |

**Evaluation:**

1. In order to check whether the data really follow the zero order kinetics, plot the absorbance readings at 340 nm as a time-dependent plot (choose a suitable scale!):



Is the increase in absorbance linear?

2. Calculate the catalytic activity of lactate dehydrogenase:

## Task 3: Spectroscopic examination of cerebrospinal fluid

**Principle:**

## Results:

**Spectrum of sample 1:**

Absorption maxims (nm):

Evaluation:

**Spectrum of sample 2:**

Absorption maxims (nm):

Evaluation:

**Spectrum of sample 3:**

Absorption maxims (nm):

Evaluation: