

Date ..... Name ..... Group .....

## Instructions and lab report form for the practical lesson on biochemistry

### Topic: Selected immunochemical methods

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#### Task 1: Immunoprecipitation curve of human albumin and estimation of albumin concentration by means of immunoturbidimetry

Principle:

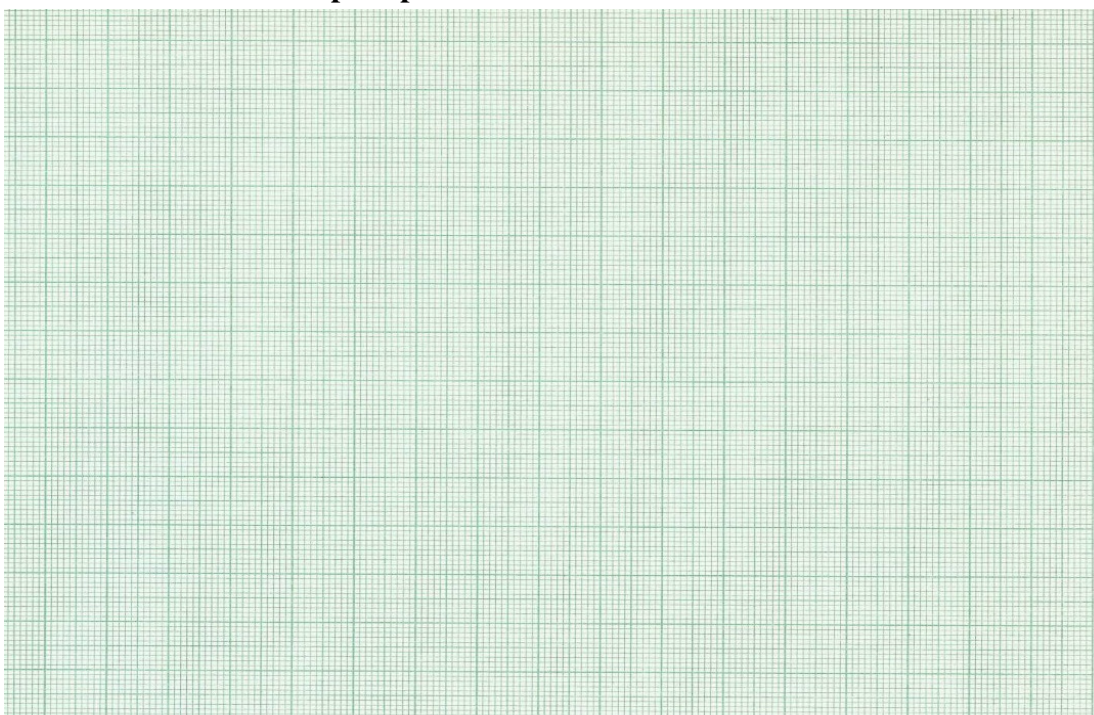
#### Results:

Calibration curve:

For construction of calibration curve use these data:

	Albumin 1,000 mg/l	Albumin 500 mg/l	Albumin 250 mg/l	Albumin 125 mg/l	Albumin 62.5 mg/l	Albumin 31.25 mg/l	Albumin 15.63 mg/l
$A_{400}$	0.19	0.817	0.908	0.647	0.419	0.242	0.137

#### Immunoprecipitation curve of human albumin



Unknown sample:

	Tube 1 (undiluted)	Tube 2 (diluted)
$A_{400}$		

**Concentration of the unknown sample:**

**Conclusion and discussion of results:**

## **Task 2: Evaluation of single radial immunodiffusion for estimation of IgG or IgM**

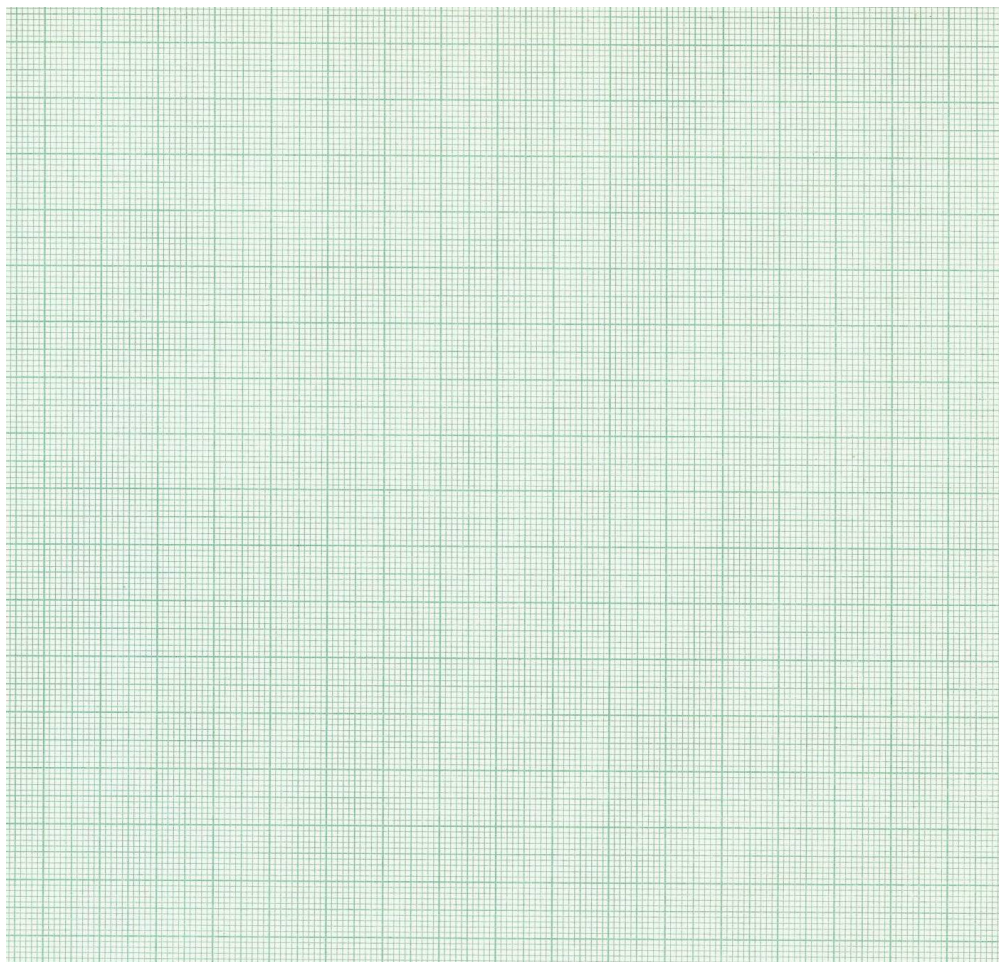
**Principle:**

**Construction of calibration curve:**

(Choose either the plate with IgM or IgG)

Standard No.	Standard concentration for IgM (g/l)	Standard concentration for IgG (g/l)	$d^2$ (mm <sup>2</sup> )
1	1.8	18.0	
2	1.6	16.2	
3	1.4	14.4	
4	1.2	12.6	
5	1.0	10.8	
6	0.8	9.0	
7	0.6	7.2	
8	0.4	5.4	

**Calibration graph for estimation of concentration of total Ig<sup>1</sup>....**



**Concentration of Ig<sup>1</sup>.... in unknown samples:**

Sample	d <sup>2</sup> (mm <sup>2</sup> )	Concentration of Ig <sup>1</sup> ..... (g/l)
1		
2		
3		
4		
5		

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<sup>1</sup> Fill in the class of Ig (written on the plate)

### Task 3: Estimation of antibodies in unknown samples by means of ELISA test

#### Principle:

#### Results and data processing:

The blank absorbance is to be subtracted from the absorbance values obtained for samples and controls. After the subtraction write all the corrected absorbance values to the following table and calculate also the arithmetic means for the two unknown samples measured in doublets.

Blank  $A_{450} = \dots\dots\dots$

	Corrected absorbance		$\bar{\emptyset} A_{450}$
	$A_{450}$	$A_{450}$	
Negative control		–	–
Positive control		–	–
Cut-off control		–	–
Sample 1			
Sample 2			

**Evaluation:**                      **Cut-off value  $\pm 10\%$**   $\dots\dots\dots$

**Sample 1**  $\dots\dots\dots$

**Sample 2**  $\dots\dots\dots$

#### Conclusion:

Decide whether the tested specific antibodies in each of the unknown samples are present (positive), absent (negative), or in the borderline zone.

**Task 4: Estimation of concentration of C-reactive protein in serum by means of turbidimetric POCT test - demonstration**

**Principle:**

**Results and interpretation:**