UNIVERSITY of LIFE SCIENCES in Lublin

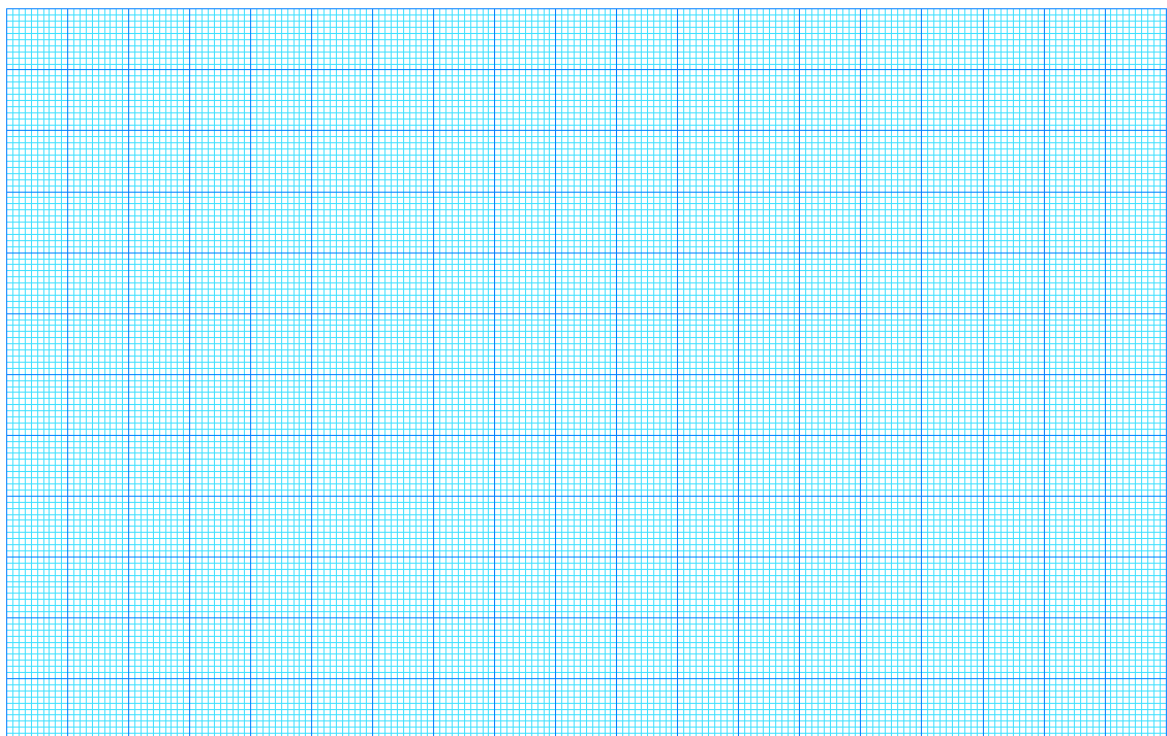FACULTY OF VETERINARY MEDICINE

Quantitative methods for protein determinations

Task 1. Quantitative determinations of protein concentration by use of biuret method – the preparation of standard curve

Protocol: The preparation of standard curve
Prepare different dilutions of standard solution of protein (Casein) in accordance to table:

No of tube	Casein (1 mg/cm3)	H_2O dest	Concentration	Absorbance
0	–	1 cm^3	0,000	
1	1,0 cm^3	–	1%	
2	0,8 cm^3	0,2 cm^3	0,8%	
3	0,6 cm^3	0,4 cm^3	0,6%	
4	0,4 cm^3	0,6 cm^3	0,4%	
5	0,2 cm^3	0,8 cm^3	0,2%	

Add 4 cm^3 of copper reagent to each tube and incubate in room temperature for 25-30 min. Measure the absorbance against blank (tube 0) at wave length 545nm. Prepare the plot of dependencies between the concentration of casein and absorbance.

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Task 2. Quantitative determination of protein in unknown sample

The aim: quantitative determination of protein content in unknown sample by use of biuret method and previously prepared standard curve.

Protocol: Add 1 cm^3 of sample to 4 cm^3 of copper reagent. After 25-30 min of incubation in room temperature measure the absorbance twice at wave length 545 nm against blank (1cm^3 of H_2O dest. and 4 cm^3 of copper reagent). Calculate the concentration of protein in unknown sample by use of standard curve.

Measurement	Absorbance at 545 nm	Concentration of protein [%]
1		
2		
Mean	–	

Attention! Standard curve will be used again in further classes and semesters. Please keep it in your records

Lublin, 24.07.2017