

Comenius University in Bratislava, Faculty of Medicine
Institute of Medical Chemistry, Biochemistry and Clinical Biochemistry

LABORATORY PROTOCOL GM-WS - 11th seminar

Determination of urea concentration in serum and in urine

Name, group:	Date:
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Principle:

Urea reacts with diacetyl monoxime in highly acidic environment and forms red-colored complex in the presence of thiosemicarbazide and iron (III) ions. Amount of urea can be determined spectrophotometrically at 525 nm.

Procedure:

	S ₁	S ₂	U ₁	U ₂	standard	ref. sample
serum 1	0.1 ml	---	---	---	---	---
serum 2	---	0.1 ml	---	---	---	---
urine 1 (1:100)	---	---	0.1 ml	---	---	---
urine 2 (1:100)	---	---	---	0.1 ml	---	---
standard	---	---	---	---	0.1 ml	---
water	---	---	---	---	---	0.1 ml
reagent	2.0 ml	2.0 ml	2.0 ml	2.0 ml	2.0 ml	2.0 ml

We mix the samples and let them stand for 15 minutes and measure absorbance at 525 nm.

Calculation:

	S ₁	S ₂	U ₁	U ₂	standard
absorbance					
concentration					5 mmol/l
correction for dilution	---	---			---

Reference values:

	in serum	in urine
urea	2.5 – 8.4 mmol/l	320 – 568 mmol/24 hours

Diuresis: Patient 1: 1.5 l/24 hours Patient 2: 0.5 l/24 hours

Assessment of oliguria:

If a patient develops oliguria (patient produces 500 ml or less of final urine in 24 hours), **we can use a ratio of concentration of urea in the urine and concentration of urea in the serum to determine probable cause of oliguria in a patient.** If this ratio is lower than 10, the oliguria is probably due to kidney malfunction. If the ratio is greater than 10, the oliguria is probably due to other causes than renal (extrarenal causes e.g. severe dehydration).

Conclusion:

Literature for next week:

Role of kidneys, synthesis of creatine.

- Practical exercises in biochemistry (Asklepios, 1993) – Chapter 12

- Lippincott's: Chapter 21, Part III, letter D: Creatine, formulas: Fig. 21.19.