

Proteinuria. Analysis of urine

Seminar No. 11

- Chapter 12 -

Filtration of plasma

- glomerular membrane is a filter system
- **structural barrier** (collagen IV)
- **electrostatic barrier** (neg. charges of sialic acid in glycoproteins repulse anionic macromolecules - proteins)
- the basement membrane allows free movement of electrolytes, water and small molecules (urea, glucose, AA, creatinine ...)

The filtration of proteins strongly depends on their molecular mass

scheme on p. 69

- **Proteins with $M_r < 60\ 000$ (microproteins)**
pass easily into urine regardless of their charge
- **Proteins with $M_r\ 60\ 000 - 150\ 000$**
only very small amount is filtered into urine
- **Proteins with $M_r > 150\ 000$**
do not pass into urine regardless of their charge

Q.

How are proteins resorbed from primary urine?

Resorption of proteins

- microproteins ($M_r < 60\ 000$) are resorbed from the primary urine by pinocytosis
- after hydrolysis in tubular cells are returned into AA pool

Q.

Which enzyme is readily filtered into urine?

A.

- see p. 26
- amylase ($M_r \sim 50\,000$)
- catalyses the hydrolysis of starch into dextrans and maltose

Q.

What is normal daily excretion of

a) proteins

b) albumin?

A.

Daily excretion of proteins

total proteins: < 150 mg/day

albumin: < 20 mg/day

Q.

What are chemical tests for proteins in urine?

1. Test strips based on protein error of acid-base indicators

primarily specific for albumin

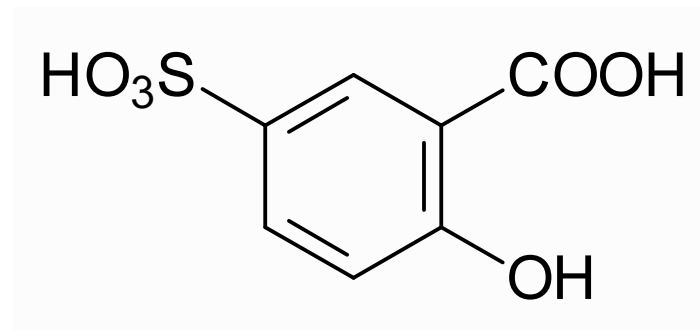
(albuPhan, Pliva-Lachema, Brno)

2. Precipitation tests

traditional test performed in a test tube

proteins are precipitated (denaturated) by 5-sulfosalicylic

acid as fine particles (turbidity)



see lab manual

Q.

What is microalbuminuria?

A.

- urine excretion of albumin in the range
of 20 – 300 mg/day
- an early indicator of diabetic nephropathy

Q.

Explain how microalbuminuria can be detected?

A.

- **immunochemical tests using antibody against human albumin**
- antibody can be **gold-labelled** \Rightarrow red coloured zone on a strip corresponds with albumin concentration in urine sample (test strips Micral)
- antibody **freely soluble** \Rightarrow nephelometry (p. 10)

see lab manual

Q.

What is SDS-PAGE?

A.

- sodium dodecyl sulfate polyacrylamide gel electrophoresis
- for principle see p. 7
- a modification of simple electrophoresis
- separation according to molecular mass to distinguish individual proteins \Rightarrow see scheme on p. 70

Glomerular proteinuria

- consequence of the loss of glomerular membrane integrity
- typical protein: albumin
- **selective glomerular proteinuria:**
proteins with M_r 60 000 – 100 000 pass into urine
- **non selective glomerular proteinuria:**
more severe glomerular lesions
proteins of all sizes $M_r > 60\ 000$ pass into urine

Tubular proteinuria

- tubules are unable to reabsorb proteins
- small proteins molecules (microproteins) $M_r < 60\ 000$
appear in the urine
- typical protein: β_2 -microglobulin

Urine sediment

- suspension material obtained by centrifugation of fresh urine sample under defined condition
- semiquantitative × quantitative
- **chemical sediment** – crystals of various compounds (salts)
- **biological sediment** – cells (RBC, WBC), casts, bacteria

Factors involved in renal stones formation

- increased concentrations of certain ions (Ca^{2+} , Mg^{2+} ...)
- extreme values of pH of urine
- low intake of fluid (low diuresis)

Insoluble compounds - Complete

Compound	Insolubility pH range		
	Acid	Neutral	Alkaline
Uric acid			
Ammonium hydrogen urate			
Calcium oxalate			
Calcium hydrogen phosphate			
Hydroxyapatite			
Ammonium magnesium phosphate			

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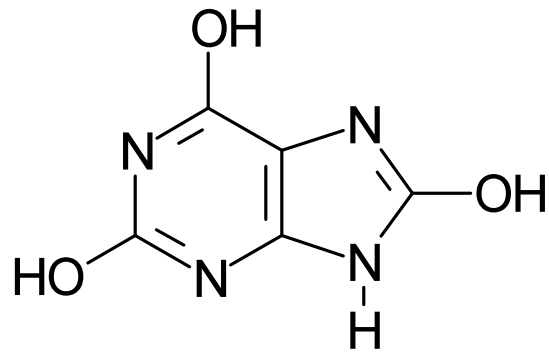
Q.

What is uric acid?

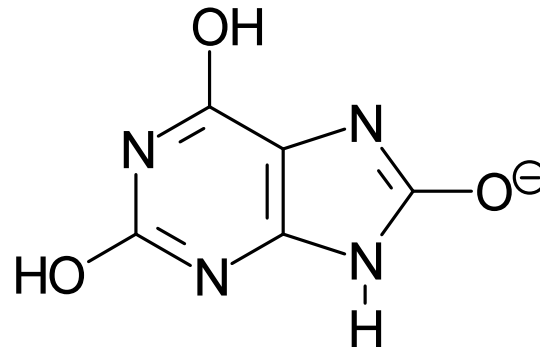
Uric acid in lactim form is a diprotic acid

$$pK_{A1} = 5.4$$

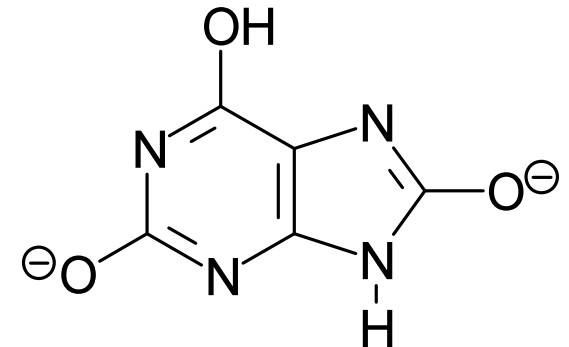
$$pK_{A2} = 10.3$$



uric acid (lactim)



hydrogenurate



urate

2,6,8-trihydroxypurine

Q.

Give the formulas of compounds:

Calcium oxalate

Calcium hydrogen phosphate

Hydroxyapatite

Ammonium magnesium phosphate

A.

Calcium oxalate



Calcium hydrogen phosphate



Hydroxyapatite



Ammonium magnesium phosphate



Some food components conduce to urolithiasis

Food	Commentary
Meat (excess)	cystein catabolized to $\text{SO}_4^{2-} + 2\text{H}^+ \Rightarrow$ urine acidification
Milk products	rich in Ca^{2+} and phosphates
Mineral waters	some of them are rich in Ca^{2+} and Mg^{2+}
Spinach	contains oxalic acid (up to 1 %)
Rhubarb	contains oxalic acid (up to 1 %)
Vitamin C (excess)	catabolized to oxalic acid

Rhubarb (*Rheum rhaponticum*)



- a herb with long reddish leaf-stalks, rich in oxalic acid
- rhubarb juicy stalks are edible when cooked and sweetened

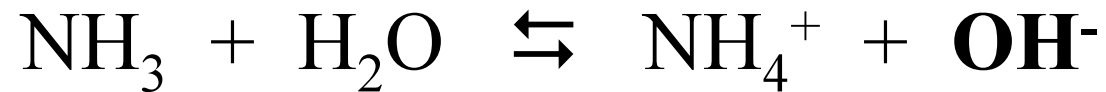
Q.

Why is urine alkaline in urinary infections?

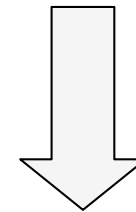
Bacterial urease catalyzes the hydrolysis of urea



ammonia



ammonium



alkaline urine \Rightarrow concrements of CaHPO_4

Q.

Which types of stones occur most frequently
in European population?

A.

- oxalates
- see the table in lab manual, chapter 11

Be sure to have the index

on the next Monday

May 29, 2006

Some students have absences

Student	Missed seminars
Botonaki F.	No. 9
Cheema S.S.	No. 4 + TEST , No. 5
Maloiso C.U.	No. 9
Nikolaides E.	No. 9
Sulaibeekh S.	No. 4 + TEST , No. 5
Tandail M.	No. 4 + TEST
Utulu D.I.	No. 5

they are strongly advised:

- to make them up in the written form (= answer questions from the corresponding chapter in seminar book)
- to write the 1st revision test, preferably on next Monday, after regular seminar (Cheema, Sulaibeekh, Tandail)