جامعة الملك سعود
King Saud University

# ESTIMATION OF URIC ACID IN SERUM

BCH 472 [Practical]

# Uric acid production

1. **Uric acid** is the product of catabolism of the purine (adenosine and guanine) that result from the break down of ingested nucleic acid (*exogenous*) or from tissue destruction (*endogenous*).
2. **Uric acid** is transported by the plasma from the **liver** to the **kidney**, where it is filtered and where about 70% is excreted. The remainder of uric acid is excreted into the GI tract and degraded.

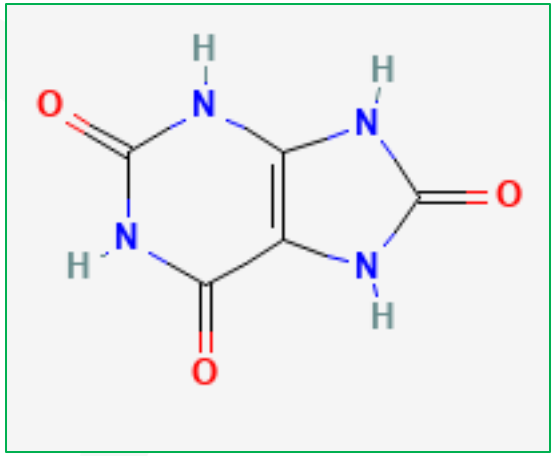


Figure 1. Chemical structure of Uric acid

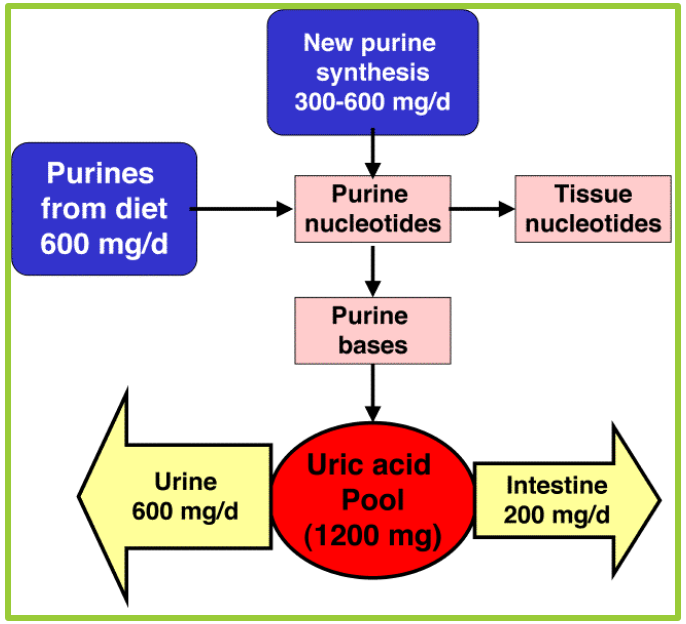


Figure 2. uric acid production and excretion

# Uric acid excretion

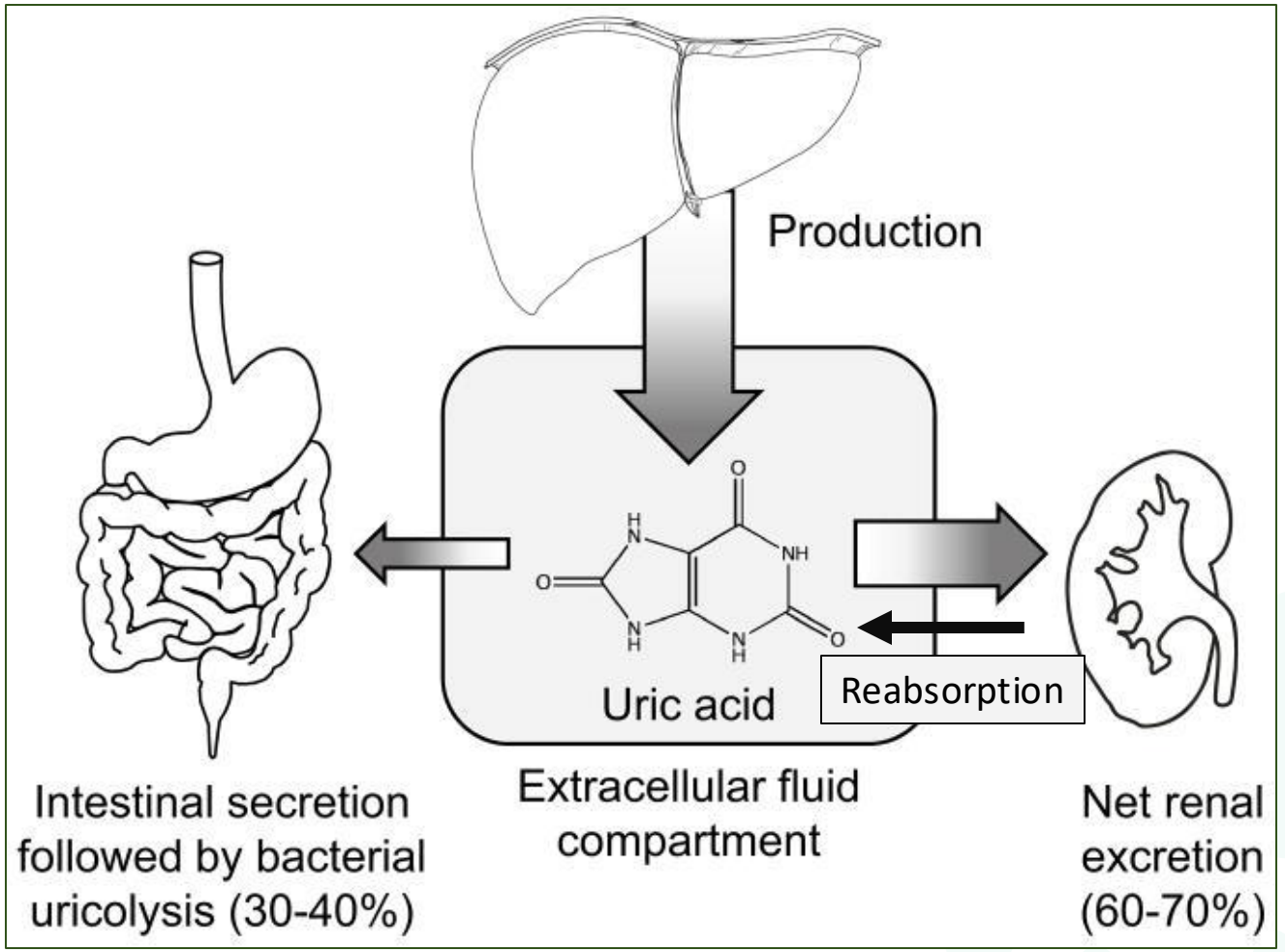


Figure 3. Uric acid excretion

# Purine metabolism

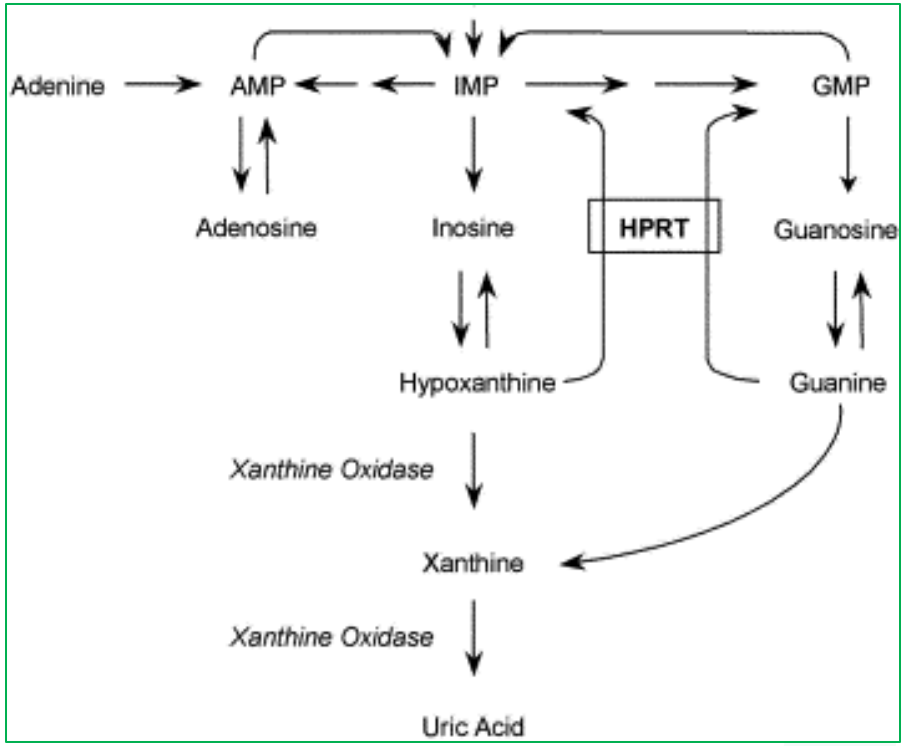


Figure 4. Pathways of purine metabolism, Source: sciencedirect.com

# Clinical Application

1. Uric acid is measured to assess inherited disorders of **purine metabolism**.
2. To conform diagnosis and monitor treatment of **gout. ??**
3. To assist the diagnosis of **renal calculi (**uric acid kidney stones).
4. To detect **kidney dysfunction**.
5. Evaluation of **leukemia.**

# Serum Uric Acid

| Case | Cause |
| --- | --- |
| **Increased** uric acid serum (Elevated uric acid levels (**hyperuricemia**) | • **Gout** (the amount of increase is <u>not</u> directly related to the severity of the disease). |
| | • **Renal diseases** and **renal failure** (decreased excretion of uric acid) |
| | • Leukemia, multiple myeloma, lymphoma. |
| | • Lesch-Nyhan syndrome (rare hereditary gout result from an enzyme defiance hypoxanthine-guanine phosphoribosyltransferase (HGPRT)). |
| **Decreased** uric acid serum (**hypouricemia** ) | • Liver disease (Decreased Production). |
| | • Fanconi syndrome (Increased excretion). |

# Urine Uric acid

- This test evaluates uric acid metabolism in gout and renal calculus formation.
- The uric acid urine test measured in a sample of urine collected over 24 hours.
- A **high level of uric acid** in the urine means that the patient is **more** likely to develop uric acid kidney stones.

| Case | Cause |
|---|---|
| **Increased** urine uric acid (uricosuria) | Tubular reabsorption defect (Fanconi syndrome) |
| | Multiple myeloma, lymphoma |
| | Lesch-Nayhan syndrome |
| **Decreased** urine uric acid | Kidney disease |

# Gout

- Excess monosodium urate crystallizes and deposits (*needle like crystals*) in the joints, soft tissues, and organs.
- This will lead to **inflammation** of tissues ➔This inflammation is responsible for the crisis symptoms acute gouty arthritis.

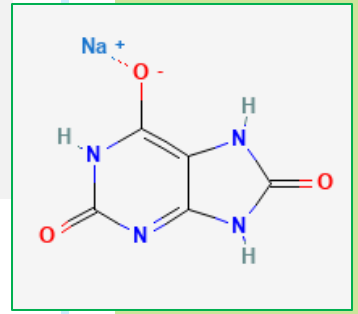


Figure 5. Chemical structure of monosodium urate

Notes:

- Hyperuricemia **does not** always lead to gout. Less than 20% of cases develop into arthritic gout disease.
- Uric acid level is just one of several criteria necessary for diagnosis.
- Blood test results can be misleading, though ➔ Some people have high uric acid levels, but never experience gout, and some people have signs and symptoms of gout, but don't have unusual levels of uric acid in their blood.

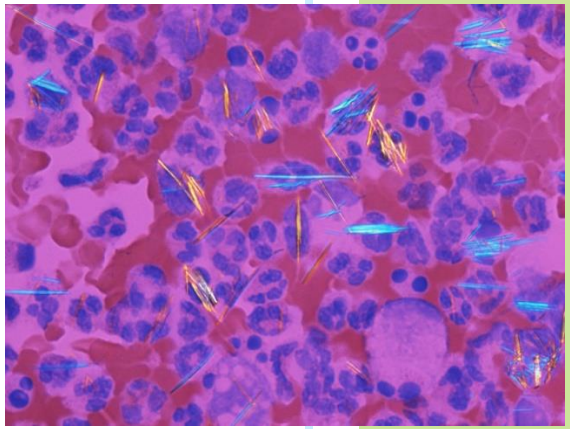

Figure 6. monosodium urate crystals

# Practical Part

## Objectives:

- To estimate the amount of uric acid in blood by using uric acid liquicolor kit

## Principle (*of the used kit*):

1. Kit contains:

The enzyme reagent used includes: buffer, uricase, peroxidase, 4-Aminophenazone and 3,5-Dichloro-2-hydroxybenzene-sulfonic acid (DCHBS) .

1. Uric acid in the sample oxidized by **uricase** to allantoin and hydrogen peroxide.

$$\text{Uric acid} + O_2 + 2H_2O \xrightarrow{\textit{Uricase}} \text{Allantoin} + CO_2 + H_2O_2$$

2. Hydrogen peroxide reacts with 3,5-Dichloro-2-hydroxybenzene-sulfonic acid (DCHBS) and 4-aminophenazone (PAP) in the presence of **peroxidase** (Hydrogen peroxide oxidoreductase ) to yield a red-violet quinoneimine dye (chromogen).

$$2H_2O_2 + \text{DCHBS} + \text{PAP} \xrightarrow{\textit{Peroxidase}} \textbf{Quinonimine} + HCl + 4H_2O$$

- The intensity of the dye is measured at 520nm and it is **directly proportional** to the concentration of uric acid present in the sample.

## Materials:

Uric acid liquid (QCA) kit.

## Method:

| | Blank | Standard | Test |
|---|---|---|---|
| Buffer | 1ml | 1ml | 1ml |
| Standard | — | 0.02 ml | — |
| Sample | — | — | 0.02 ml |

1. Water bath at 37ºC for 5 min.
2. Read absorbance at 520 nm.

## Calculations of the Results:

$$\text{-Serum uric acid} = \frac{\text{A (Sample)}}{\text{A (Standard)}} \times \text{Std. Conc. (8 mg/dl)}$$

**Reference value in serum:**

Serum: 2.5–7.0 mg/dL

Urine: 250 –750 mg/24 hour

## Homework

- Why does uric acid increase in leukemia?
- Why does Lesch-Nyhan syndrome cause hyperuricemia?
- What is the mechanism of action of allopurinol in managing gout?

## References


- Bobulescu, I. A., & Moe, O. W. (2012). Renal Transport of Uric Acid: Evolving Concepts and Uncertainties. *Advances in Chronic Kidney Disease*, *19*(6), 358–371. http://doi.org/10.1053/j.ackd.2012.07.009.
- A Manual of Laboratory and Diagnostic Tests 9th edition (January , 2014), Frances T Fischbach RN, BSN, MSN By Lippincott Williams & Wilkins Publishers.