Disorders of Purines Metabolism
Purine Metabolism

Purine nucleotides are essential cellular constituents.

- Purine metabolism can be divided into three pathways:
  1. The biosynthetic pathway, often termed *de novo*, starts with the formation of phosphoribosyl pyrophosphate (PRPP) and leads to the synthesis of inosine monophosphate (IMP).
  - From IMP, adenosine monophosphate (AMP) and guanosine monophosphate (GMP) are formed.
  - Further metabolism leads to their di- and triphosphates, to their corresponding deoxyribonucleotides, and to RNA and DNA.
2- **The catabolic pathway** starts from GMP, IMP and AMP, and produces uric acid, a poorly soluble compound, which tends to crystallize once its plasma concentration surpasses 6.5–7 mg/dl (0.38–0.47 mmol/l).

3- **The salvage pathway** utilizes the purine bases, guanine, hypoxanthine and adenine, which are provided by food intake or the catabolic pathway, and reconverts them into, respectively, GMP, IMP and AMP.
Biological significance of nucleotide metabolism

• Nucleotides make up nucleic acids (DNA and RNA)
• Nucleotide triphosphates are the “energy carriers” in cells (primarily ATP)
• Many metabolic pathways are regulated by the level of the individual nucleotides
  – Example: cAMP regulation of glucose release
• Adenine nucleotides are components of many of the coenzymes
  – Examples: NAD\(^+\), NADP\(^+\), FAD, FMN, coenzyme A
Inborn Errors of Purine Metabolism

Inborn errors of purine metabolism comprise errors of:
- **purine nucleotide synthesis**: phosphoribosylpyrophosphate (PRPP) synthetase superactivity, adenylosuccinase (ADSL) deficiency, AICA-ribosiduria caused by ATIC deficiency;
- **purine catabolism**: the deficiencies of muscle AMP deaminase (AMP-DA, also termed myoadenylate deaminase), adenosine deaminase (ADA), purine nucleoside phosphorylase (PNP) and xanthine oxidase;
- **purine salvage**: the deficiencies of hypoxanthine-guanine phosphoribosyltransferase (HGPRT) and adenine phosphoribosyltransferase (APRT). The deficiency of deoxyguanosine kinase causes mitochondrial DNA depletion.
Lesch-Nyhan Syndrome

- Due to severe or complete deficiency of Hypoxanthine Guanine Phosphoribosyltransferase (HGPRT).
- Recessive X-linked genetic disease.
- This disorder is an example for loss of normal feedback inhibition.
- This disorder is characterized by:
  - Choreoathetosis (movement disorder).
  - Severe neurologic disease, characterized by self-mutilating behaviors such as lip and finger biting and/or head banging.
  - A severe gouty arthritis.
  - Early death may be due to kidney failure.
Lesch-Nyhan syndrome

• Hyperuricemia, up to 20 times the uric acid in the urine than in normal individuals. Uric acid crystals form in the urine.
• The rate of purine synthesis is increased about 200X
• The neurological abnormalities may result from change in brain purine levels produced by the disease because some purines are putative neurotransmitters.
• No successful treatment, and afflicted individuals die in early age. Treated with allopurinol, a competitive inhibitor of xanthine oxidase, reduces the uric acid formation and symptoms of gout.
Diagnosis

1- By hyperuricemia. Patients excrete excessive amounts of uric acid, ranging from 25 to 140 mg (0.15 to 0.85 mmol)/kg of body weight per 24 h, as compared to an upper limit of 18 mg (0.1 mmol)/kg per 24 h in normal children.

2- Determination of the ratio of uric acid to creatinine (mg/mg) in morning samples of urine provides a screening test. This ratio is increased in the patients.

3- Enzyme assay in red blood cells. Patients with the Lesch-Nyhan syndrome display nearly undetectable HGPRT activity.
Lesch-Nyhan syndrome
Partial deficiency of HGPRT

- Individual with residual activity of HGPRT up to 30% have only gout as a result of uric acid overproduction. No neurological finding.
- Recassive X-linked disorder.
- This accounts for less than 2% of all male patients with gout.
Adenosine Deaminase Deficiency
Severe combined immune deficiency (SCID)

• Autosomal recessive disease.
• Both B and T lymphocytes are affected.
• Patients are susceptible, often fatally, to infectious diseases because of an inability to mount an immune response to antigenic challenge.
• Infants with this deficiency have a high fatality rate due to infections.
ADA deficiency

- In the absence of ADA lymphocytes are destroyed
- Deoxyadenosine is not destroyed, is converted to dAMP and then into dATP
- There marked increase in cellular concentrations of dATP due to the lack of conversion of excess deoxyadenosine to deoxyinosine and hypoxanthine.
- dATP is a potent feedback inhibitor of deoxynucleotide biosynthesis and DNA replication
ADA deficiency

• treatment consists of administering pegylated ADA which can remain in the blood for 1 – 2 weeks
• more efficient is gene therapy: replacing the gene that is missing or defective
• gene therapy has been performed on selected patients
purine nuceloside phosphorylase deficiency

- A less severe immunodeficiency results from the lack of another purine degradative enzyme, purine nuceloside phosphorylase (PNP). Decreased activity of this enzyme leads to accumulation primarily of dGTP. This accumulation also affects DNA replication, but less severely than does excessive dATP.
- Autosomal recessive disease.
- The phosphorylase deficiency destroys only the T class of lymphocytes and not the B cells.
- There reduction in uric acid production, increased levels of guanosine deoxyguanosine, inosine and deoxyinosine.
GOUT

• A heterogenous disorder caused by genetic and environmental factors due to hyperuricemia.

• Increased ingestion of proteins, specially in old age, leading to hyperuricemia.

• Uric acid has a threshold in kidney, the excess will precipitate as mono-sodium urates in joints, tendons, and surrounding tissues.
Intracellular uric acid crystal under polarised light (left) and under non-polarised light (right)

With time, elevated levels of uric acid in the blood may lead to deposits around joints. Eventually, the uric acid may form needle-like crystals in joints, leading to acute gout attacks. Uric acid may also collect under the skin as tophi or in the urinary tract as kidney stones.
Pathophysiology

The greater the degree and duration of hyperuricemia, the greater is the likelihood of gout and the more severe are the symptoms. Urate levels can be elevated because of:

• Decreased excretion.
• Increased production.
• Increased purine intake.

• There are two types of gout:
  • 1- **Primary gout**.
  • 2- **Secondary gout**, due to other diseases eg cancer, renal dysfunction, Metabolic syndrome (the combination of hypertension, diabetes, dyslipidemia, truncal obesity, increased cardiovascular disease risk).
Primary gout

- It is caused by an excessive formation of uric acid due to overproduction of purines by de novo pathway.
- Molecular basis:
  - 1- Mutations in PRPP synthetase (hyperactive)
  - 2- Partial deficiency of HGORT enzyme.
  - 3- Glycogen storage disease type I.
  - 4- Hyperactivity of glutathione reductase.
Gout

• Mainly in adult males
• Rarely encountered in premenopausal women
• Symptoms are caused by deposition of crystals of monosodium urate monohydrate (can be seen under polarized light)
• Usually affect joints in the lower extremities (the big toe is the classic site)
- Gout usually presents as recurrent attacks of acute inflammatory arthritis (red tender, hot, swollen joint). The joint that is most commonly affected is the first metatarsalphalangeal joint at the base of the big toe and when this occurs it is known as podagra.
- Prolonged or acute elevation of blood urate leads to precipitation, as crystals of sodium urate, in the synovial fluid of joints. These precipitates cause inflammation, resulting in painful arthritis, which can lead to severe degeneration of the joints.
- People with long-standing hyperuricemia can tophi (uric acid crystal deposits) in tissues. These are usually hard, non-painful deposits.
- Extensive tophi that invade bone are associated with arthritis due to bone erosion.
- Elevated levels of urine uric acid can lead to uric-acid crystals precipitating in the kidneys which may form kidney stones and lead to urate nephrophathy.
Four Stages of Gout

1. Asymptomatic hyperuricemia
2. Acute gouty arthritic attacks
3. Asymptomatic intercritical period
4. Tophaceous gout (characterized by the formation of tophi in joints)
Metabolic Derangement

- The enzyme forms phosphoribosyl pyrophosphate (PRPP) from ribose-5-phosphate and ATP.
- PRPP is the first intermediate of the \textit{de novo synthesis of purine} nucleotides which leads to the formation of inosine monophosphate (IMP), from which the other purine compounds are derived.
- PRPP synthetase is highly regulated. Various genetic regulatory and catalytic defects lead to superactivity, resulting in increased generation of PRPP.
- Because PRPP amidotransferase, the rate-limiting enzyme of the \textit{de novo pathway}, is physiologically not saturated by PRPP, the synthesis of purine nucleotides increases, and hence the production of uric acid.
- PRPP synthetase superactivity is one of the few known examples of an hereditary anomaly of an enzyme which enhances its activity.
Gout
Diagnosis

• Clinical criteria.
• Measurement of blood uric acid.
• Synovial fluid analysis for detection of sodium urate crystals.
• Extensive kinetic studies of the enzyme, which are performed on erythrocytes and cultured fibroblasts.
Genetics

- The various forms of PRPP synthetase superactivity are inherited as X-linked traits.
- Heterozygous females have also been found with gout and/or hearing impairment.
- Studies of the gene in some families revealed a different single base change in each of them.
Treatment

- Treatment with allopurinol, which inhibits xanthine oxidase.
- Avoid purine rich foods:
  - red meat and organ meat (liver, kidneys)
  - shellfish, anchovies, mackerel, tuna
  - peas and beans, asparagus, lentils
- high fluid intake and, since uric acid and xanthine are more soluble at alkaline than at acid pH, administration of sodium bicarbonate, potassium citrate or citrate mixtures to bring urinary pH to 6.0-6.5.
- Anti-inflammatory drugs.
- Adequate control of the uricemia prevents gouty arthritis and urate nephropathy.