NUCLEIC ACID METABOLISM

LUCIA DHIANTIKA WITASARI

Biosynthesis and Degradation of Nucleotides

synthesis of nucleotides :

- the **de novo pathways** → begins with their metabolic precursors: amino acids, ribose 5-phosphate, CO_2, and NH_3.
- **the salvage pathways** → recycle the free bases and nucleosides released from nucleic acid breakdown

SALVAGE PATHWAY

Activated ribose (PRPP) + base

Nucleotide

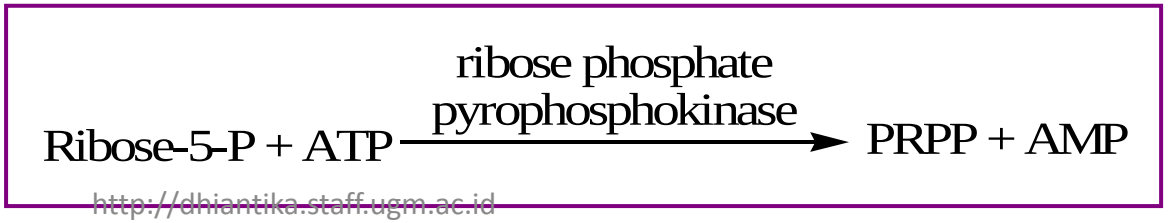
DE NOVO PATHWAY

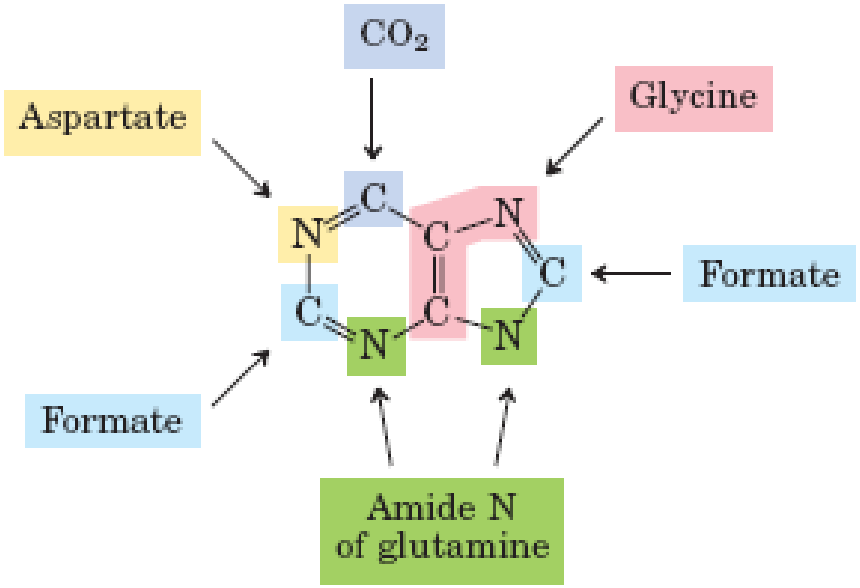
Activated ribose (PRPP) + amino acids + ATP + CO_2 + . . .

Nucleotide

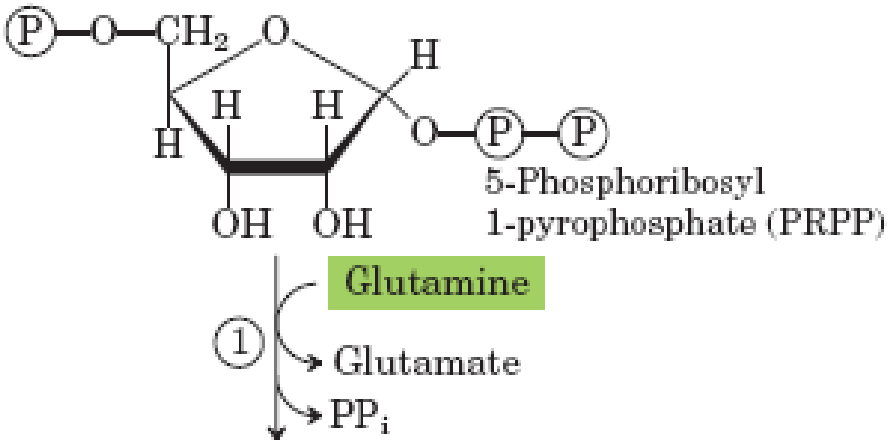
De novo pathways

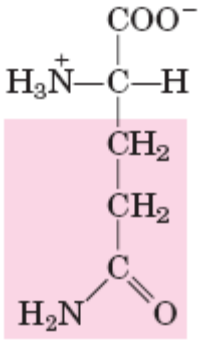
- Nearly identical in all living organisms
- Free bases are not intermediates
- Purine ring built up
- Pyrimidine ring synthesized as orotate
- Enzyme complexes
- Small cellular pools of nucleotides
- Important precursors:
  - 5-phosphoribosyl-1-pyrophosphate (PRPP)
  - Amino acids

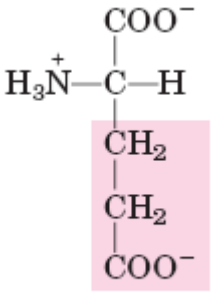


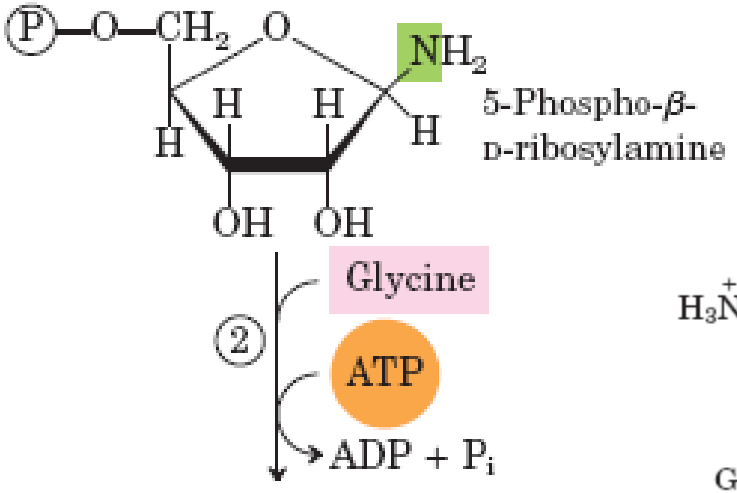


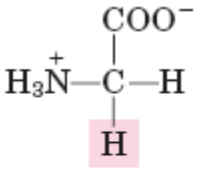
Origin of the ring atoms of purines.



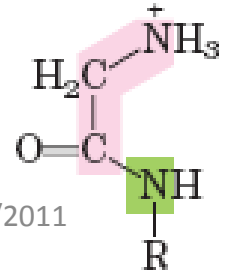


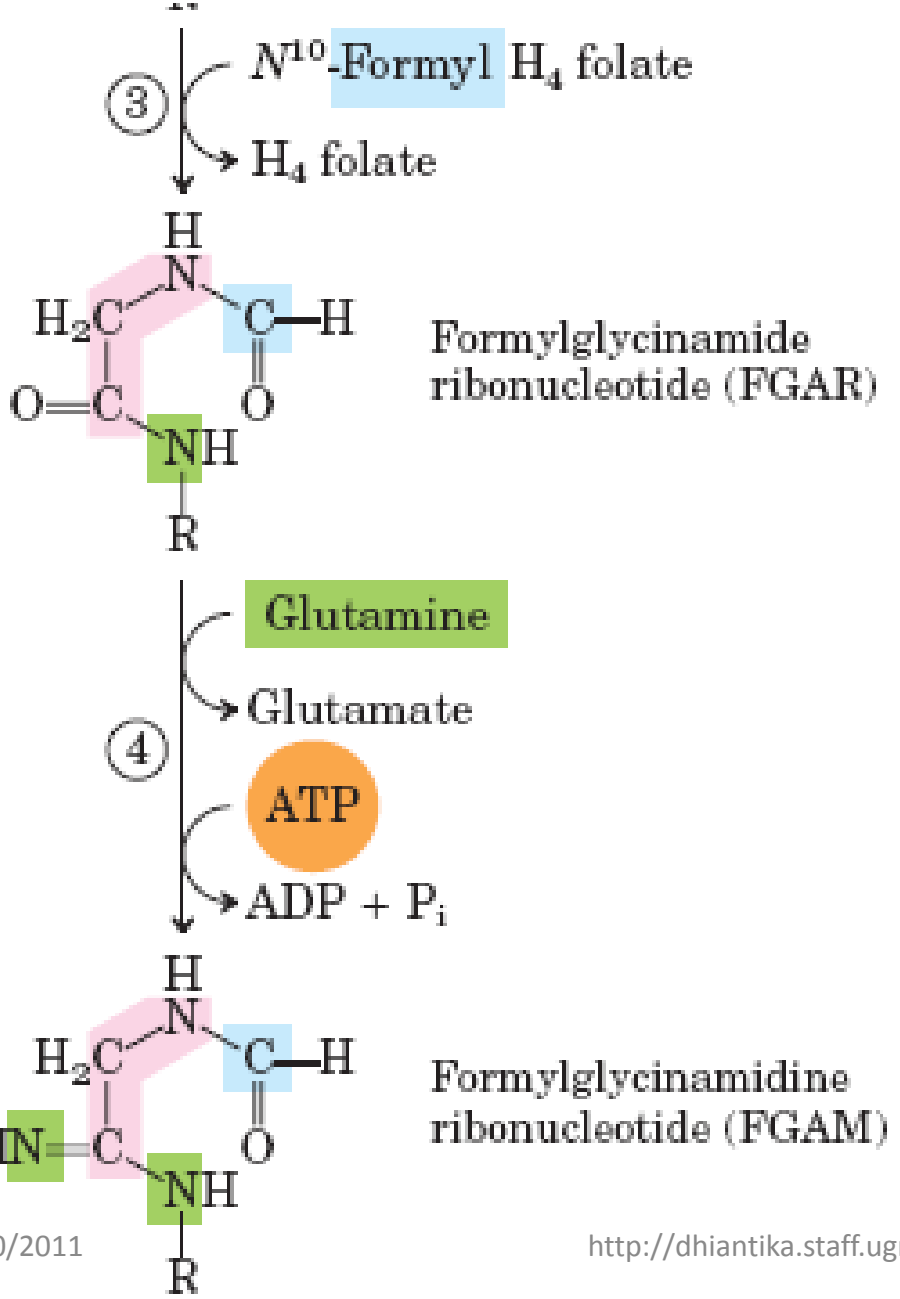


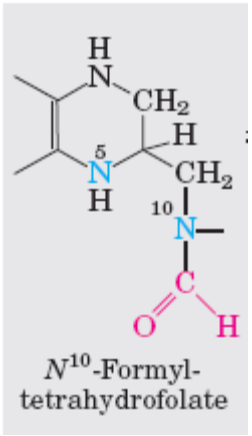


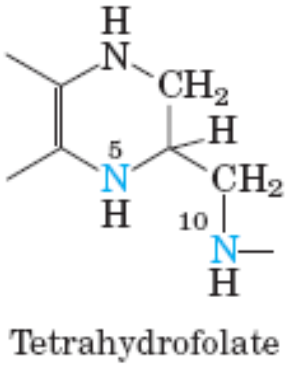


1. glutamine-PRPP amidotransferase
2. GAR synthetase
3. GAR transformylase
4. FGAR amidotransferase

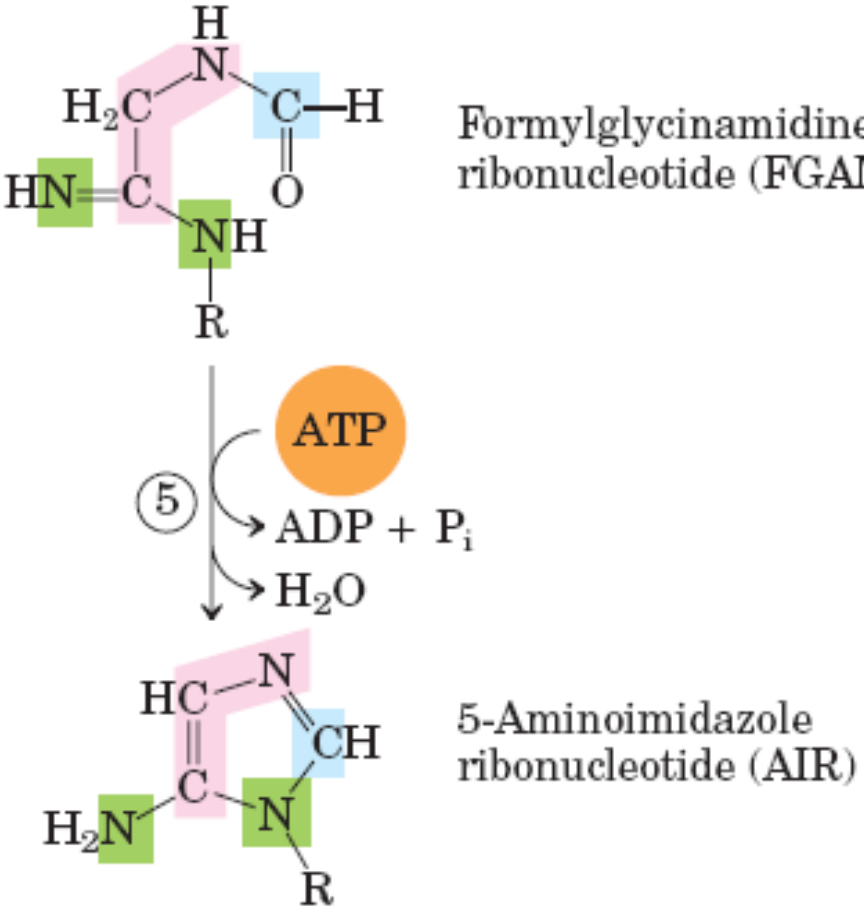




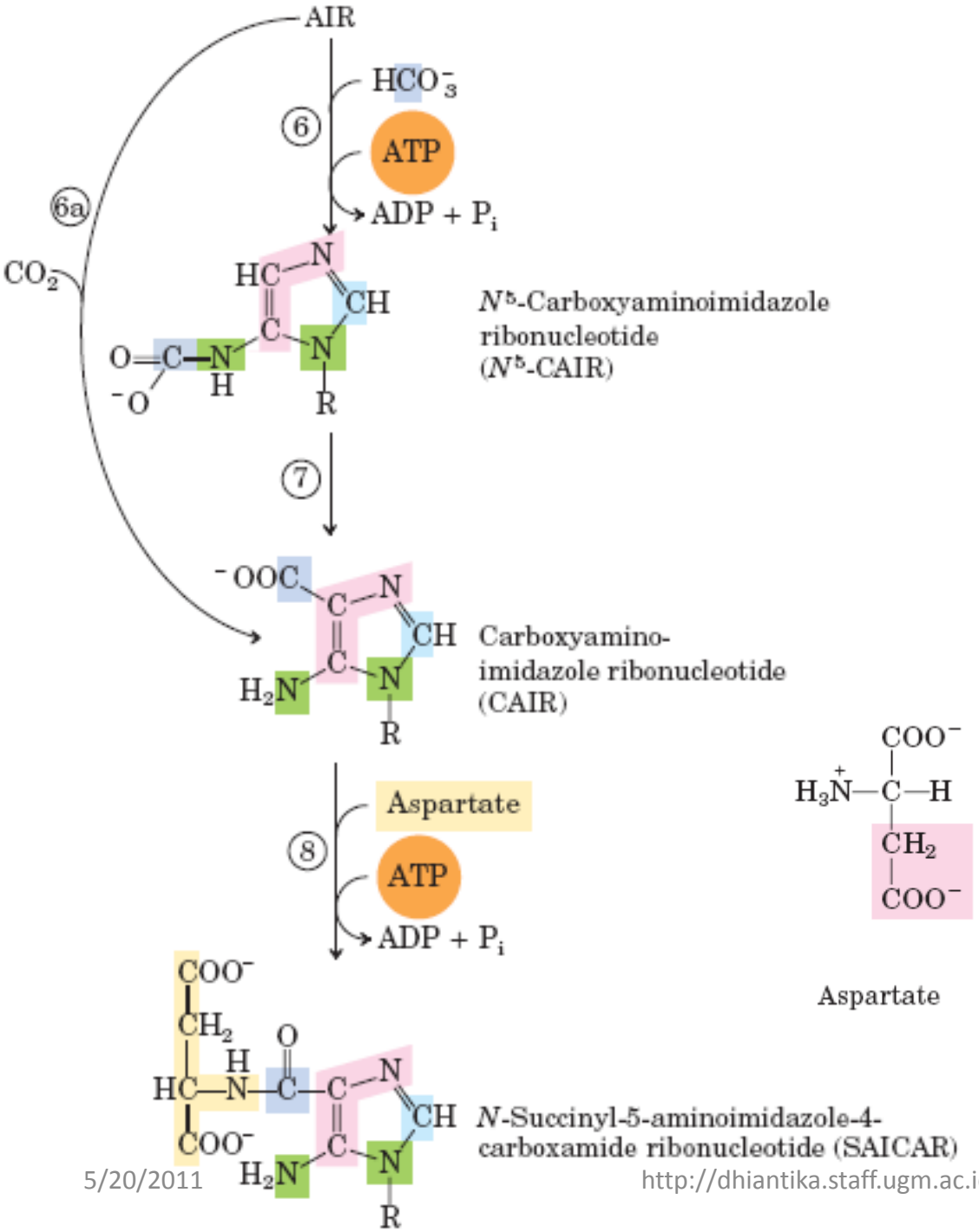




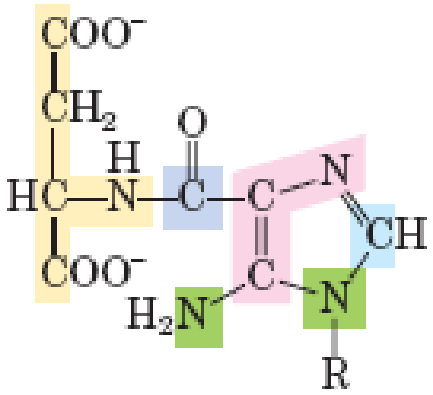
① glutamine-PRPP amidotransferase
② GAR synthetase
③ GAR transformylase
④ FGAR amidotransferase



(5) FGAM cyclase (AIR synthetase)
(6) N^5-CAIR synthetase
(6a) AIR carboxylase
(7) N^5-CAIR mutase
(8) SAICAR synthetase



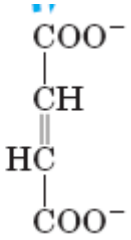
(5) FGAM cyclase (AIR synthetase)
(6) N^5-CAIR synthetase
(6a) AIR carboxylase
(7) N^5-CAIR mutase
(8) SAICAR synthetase

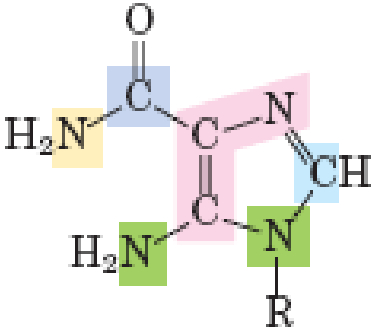


N-Succinyl-5-aminoimidazole-4-carboxamide ribonucleotide (SAICAR)

⑨ Fumarate

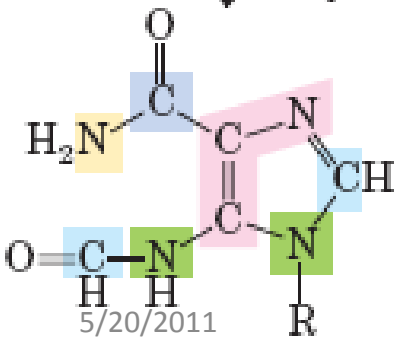
Fumarate





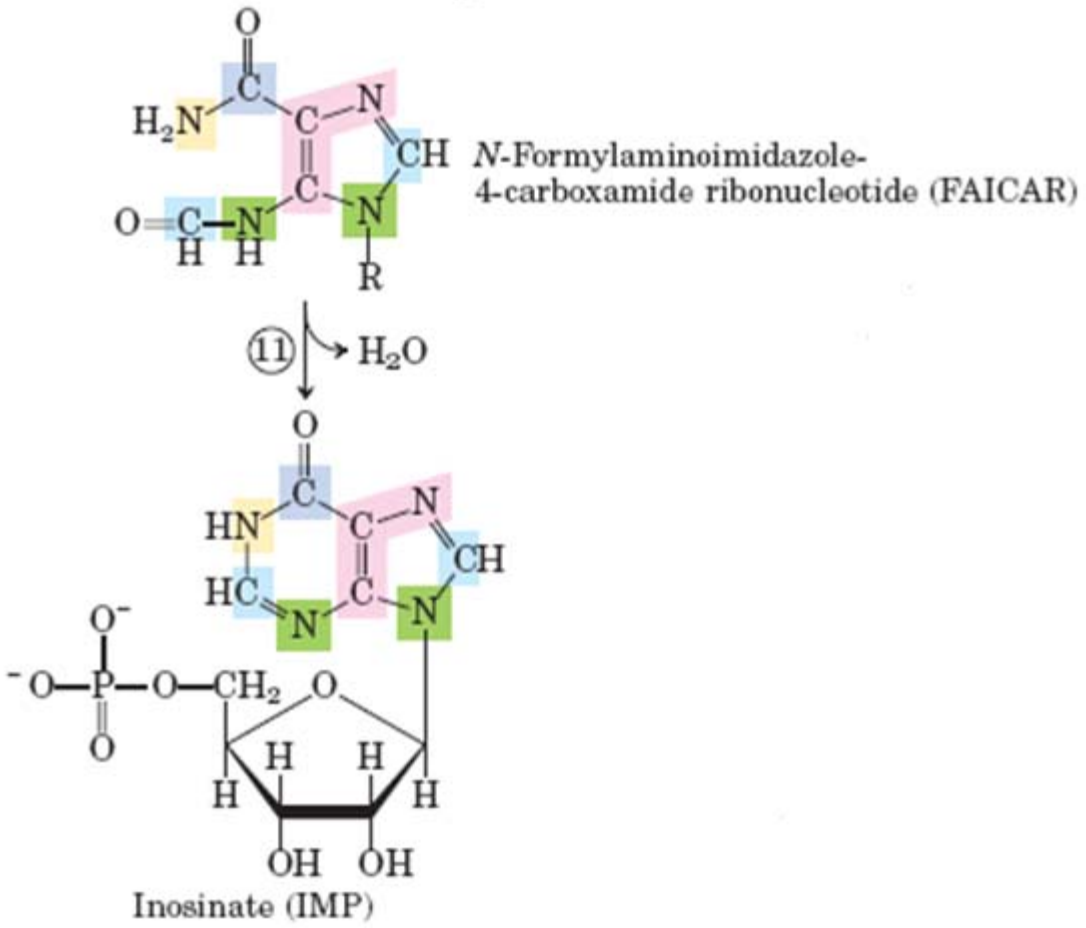
5-Aminoimidazole-4-carboxamide ribonucleotide (AICAR)

⑩ N^{10}-Formyl H_4 folate → H_4 folate



N-Formylaminoimidazole-4-carboxamide ribonucleotide (FAICAR)

⑨ SAICAR lyase
⑩ AICAR transformylase
⑪ IMP synthase



(9) SAICAR lyase
(10) AICAR transformylase
(11) IMP synthase

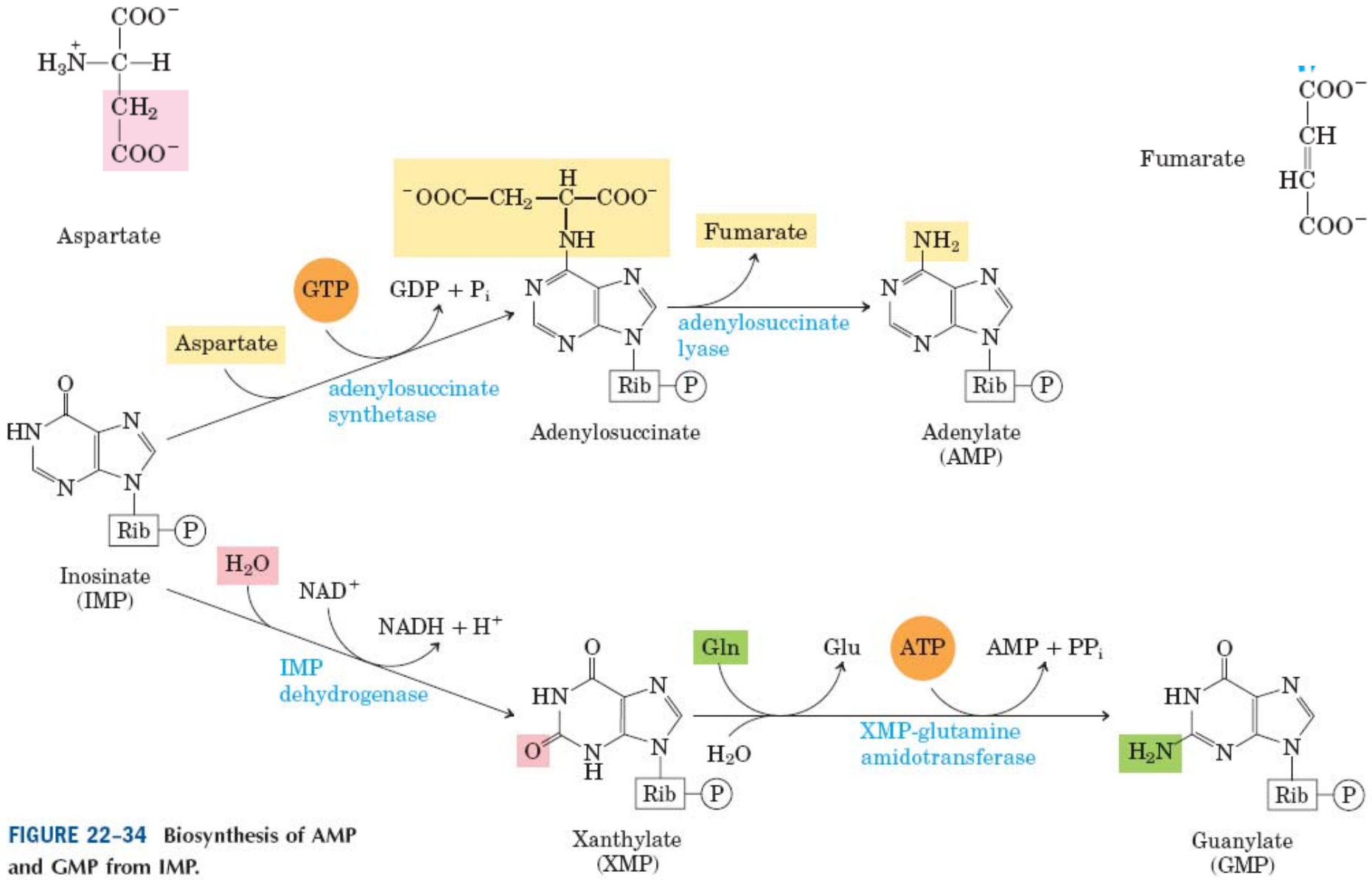
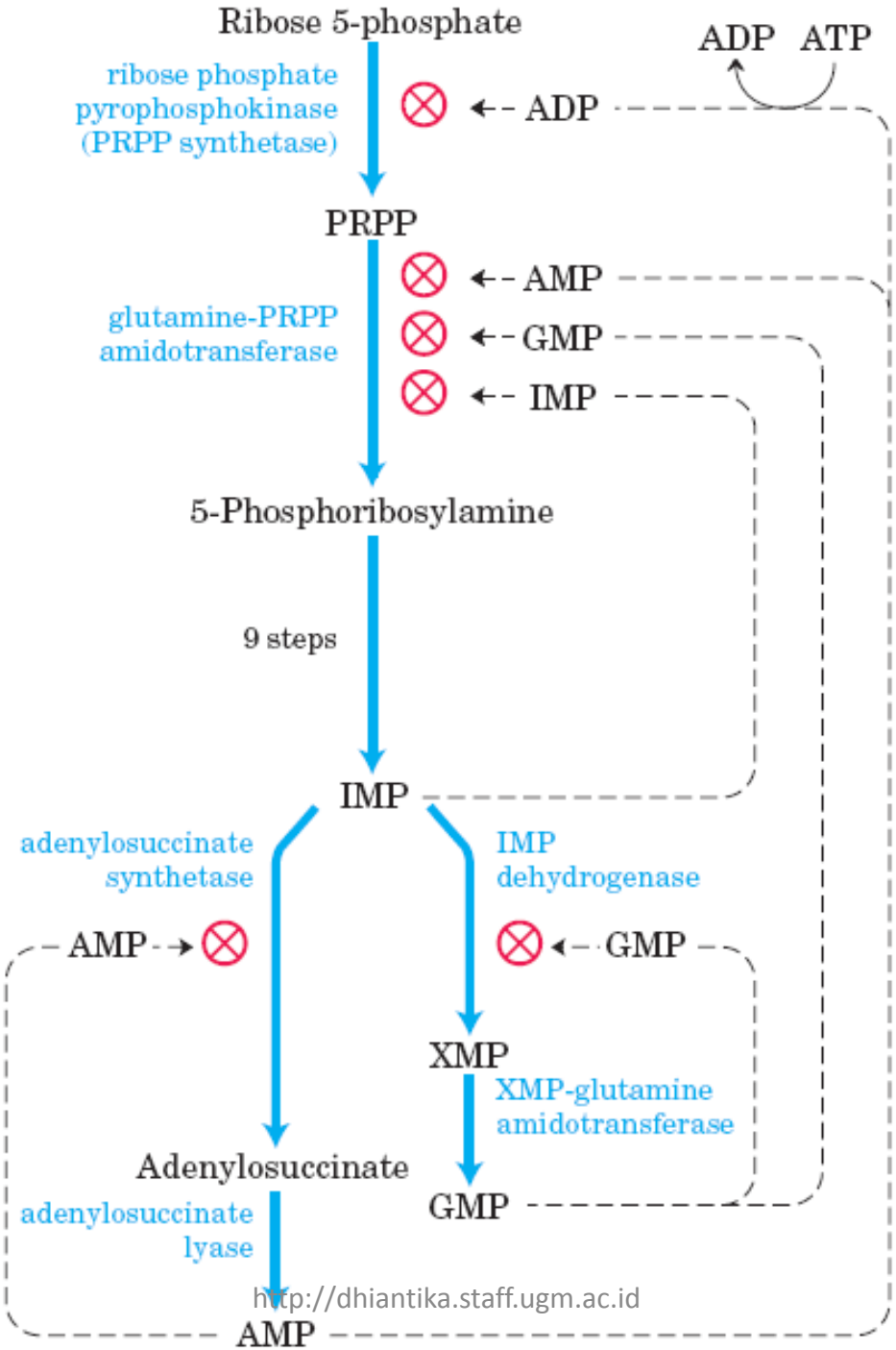
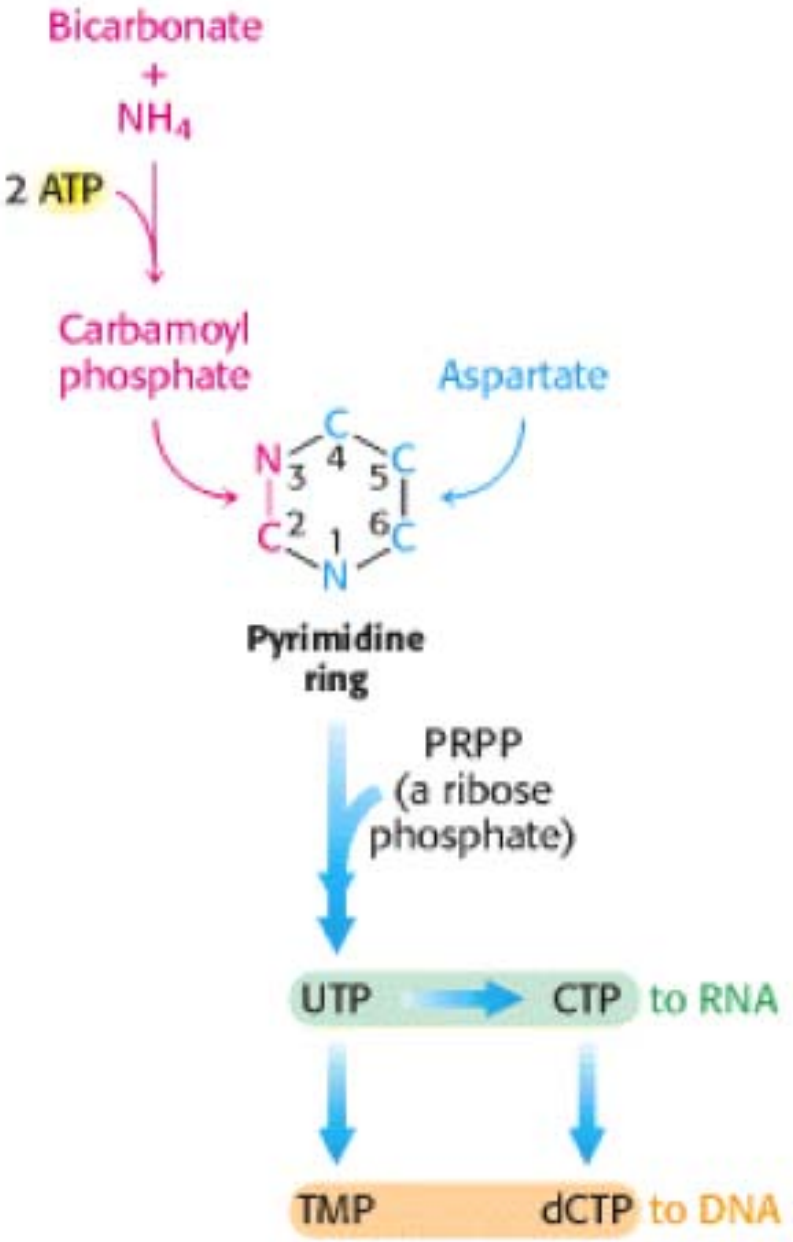


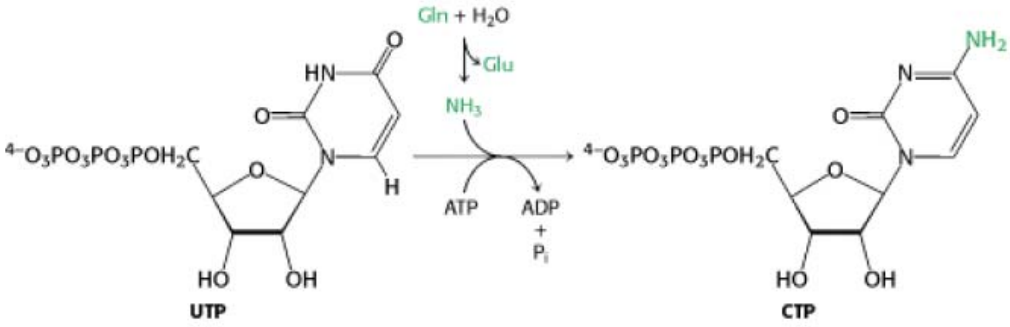
FIGURE 22–34 Biosynthesis of AMP and GMP from IMP.

Regulatory mechanisms in the biosynthesis of adenine and guanine nucleotides in *E. coli.*



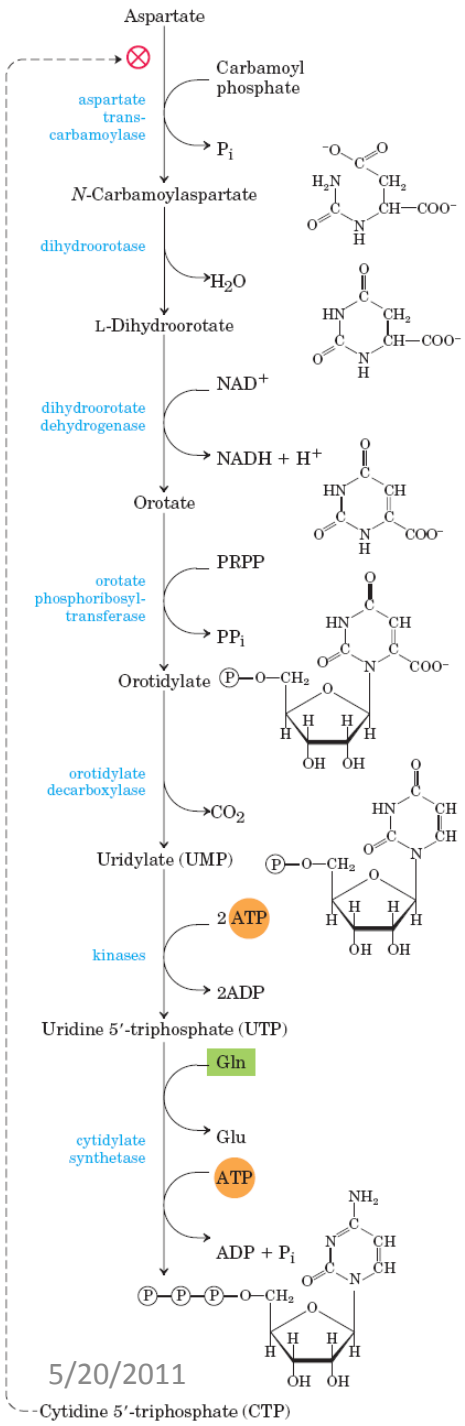
de Novo Pathway for Pyrimidine Nucleotide Synthesis

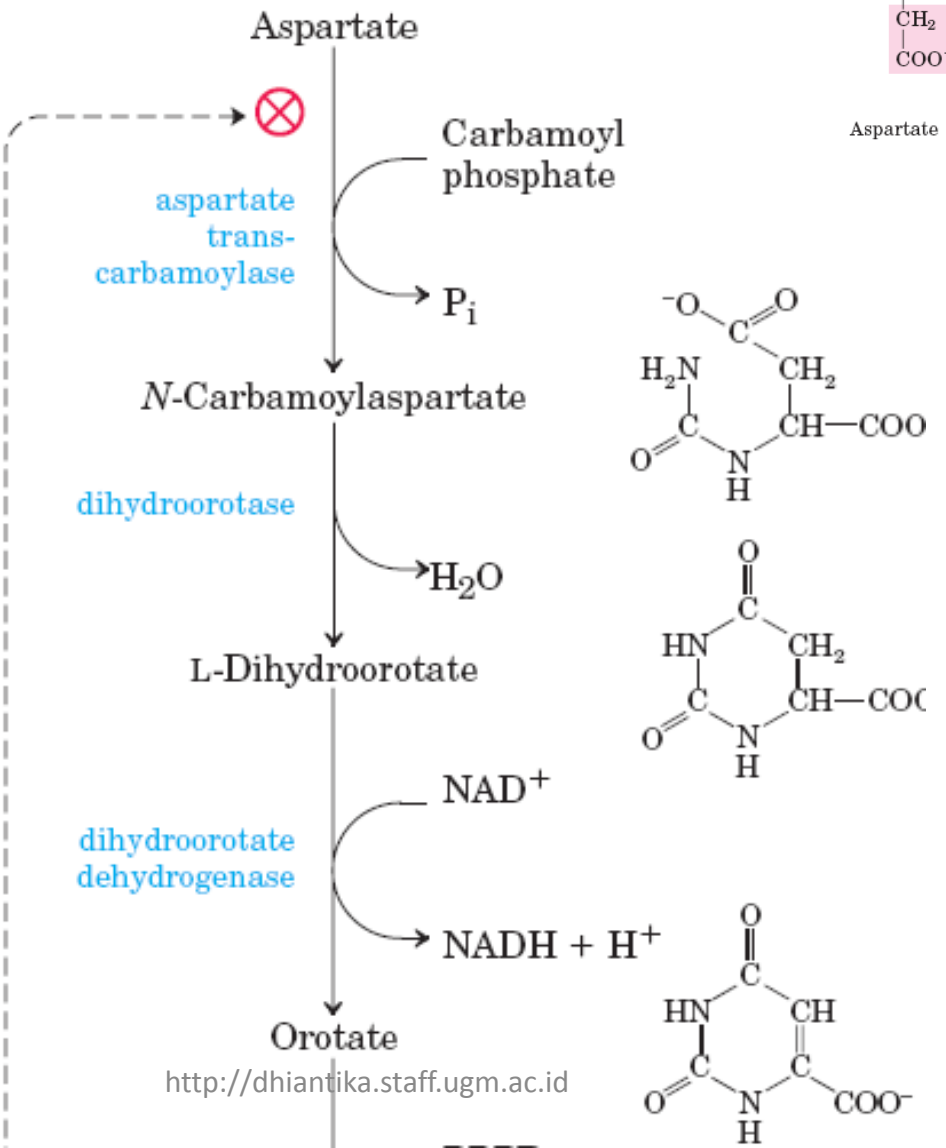


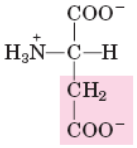


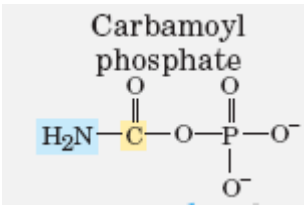
CTP Is Formed by Amination of UTP

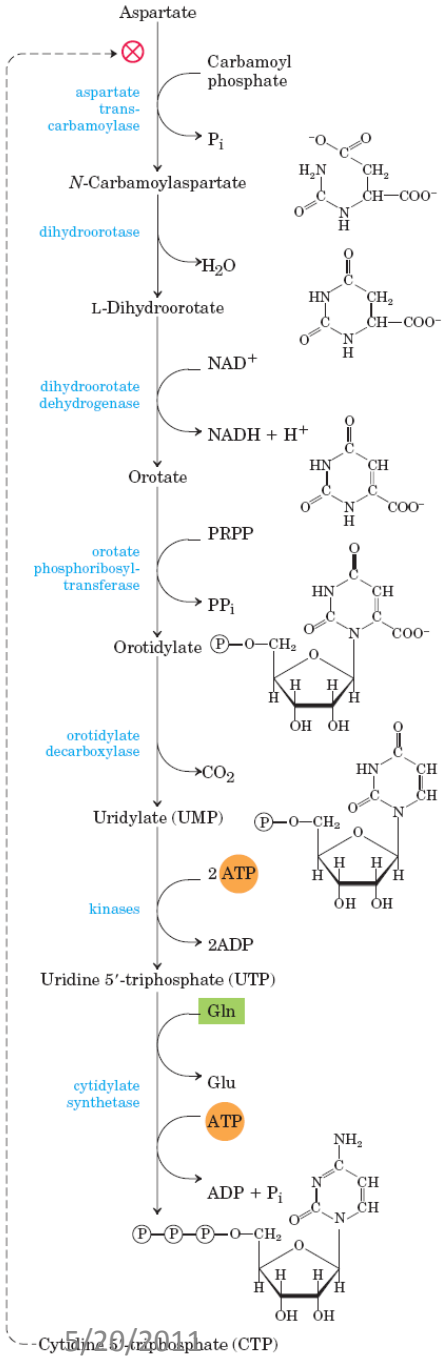
De novo synthesis of pyrimidine nucleotides: biosynthesis of UTP and CTP via orotidylate.

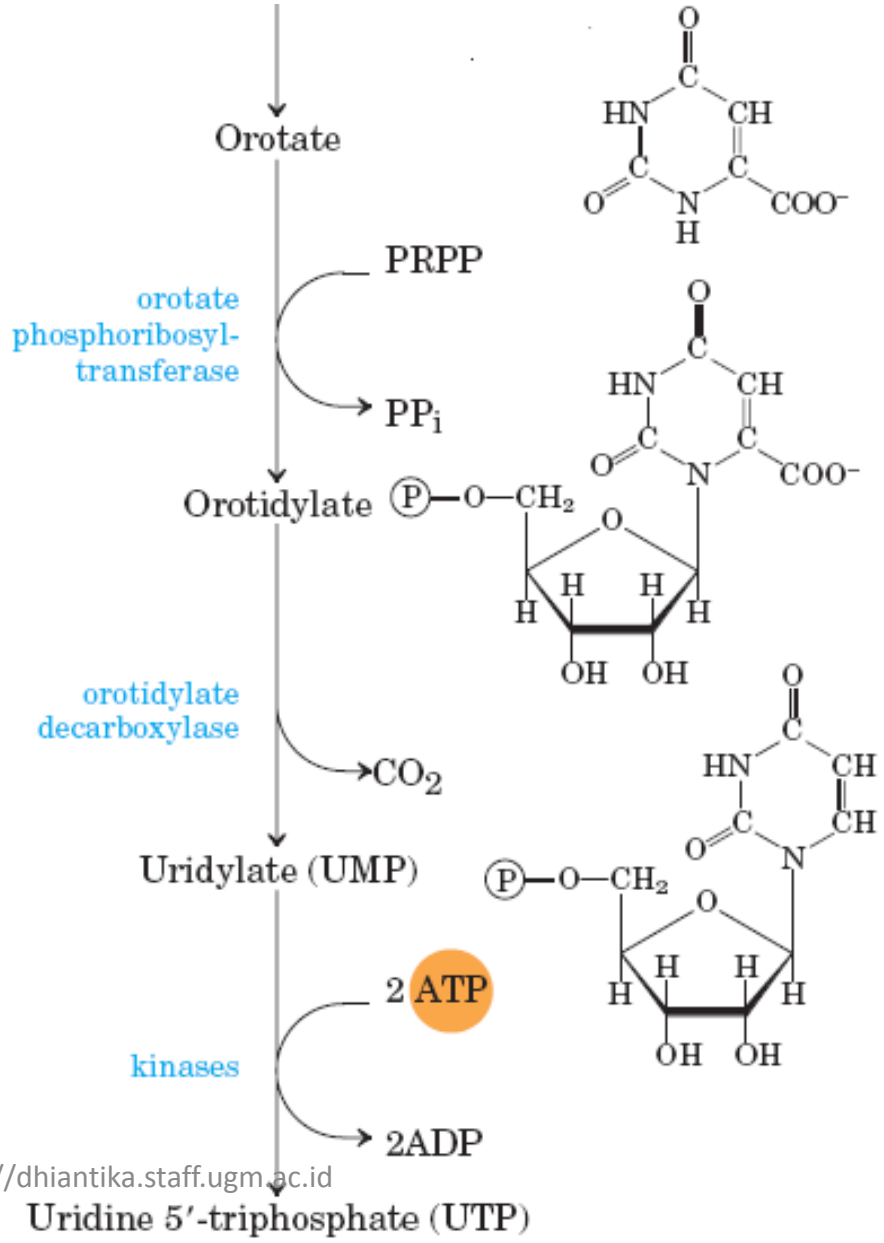


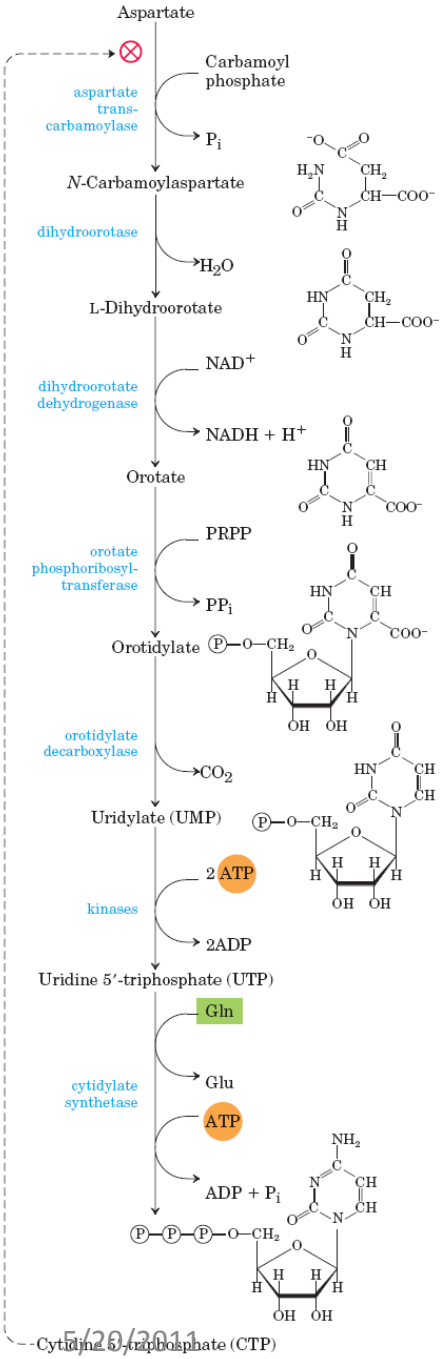


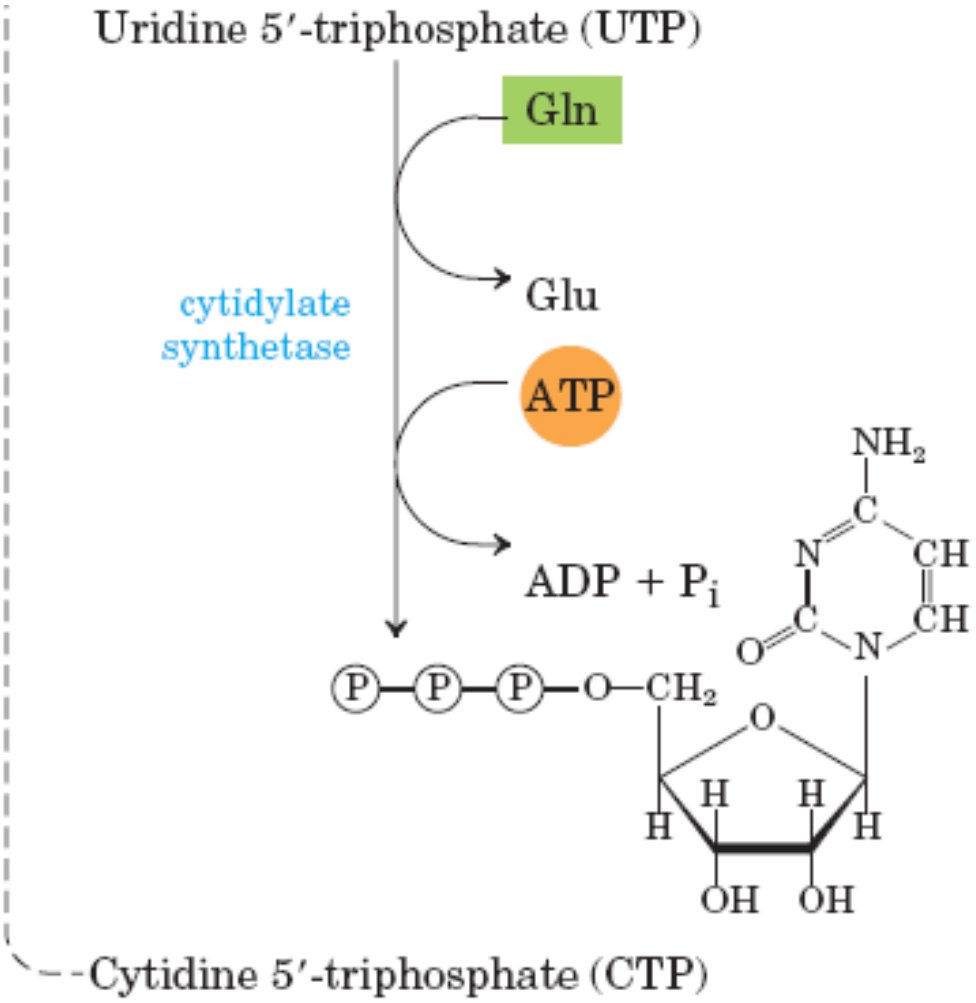




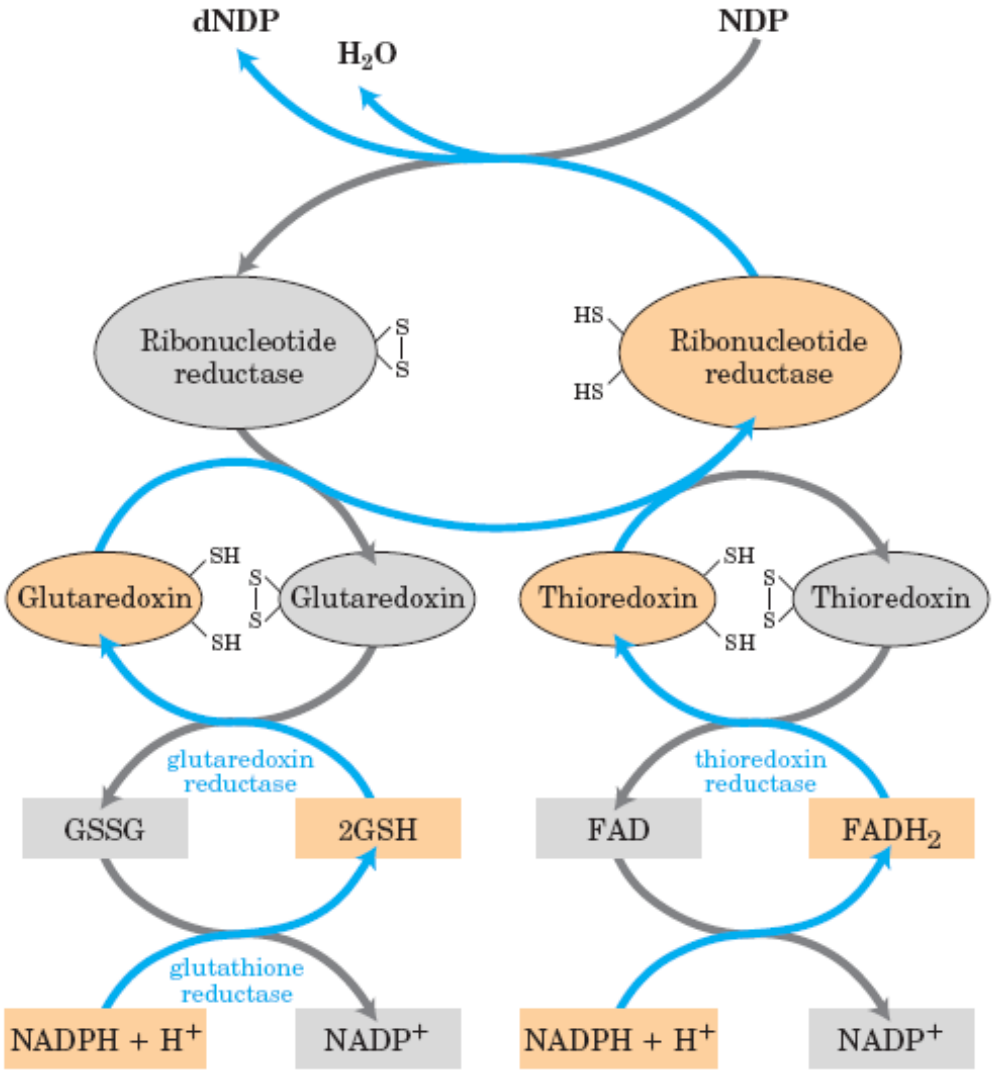





Aspartate
Carbamoyl phosphate
aspartate trans-carbamoylase
P_i
N-Carbamoylaspartate
dihydroorotase
H_2O
L-Dihydroorotate
NAD^+
dihydroorotate dehydrogenase
$NADH + H^+$
Orotate
PRPP
orotate phosphoribosyl-transferase
PP_i
Orotidylate
orotidylate decarboxylase
CO_2
Uridylate (UMP)
2 ATP
kinases
2ADP
Uridine 5′-triphosphate (UTP)
Gln
Glu
cytidylate synthetase
ATP
ADP + P_i
Cytidine 5′-triphosphate (CTP)

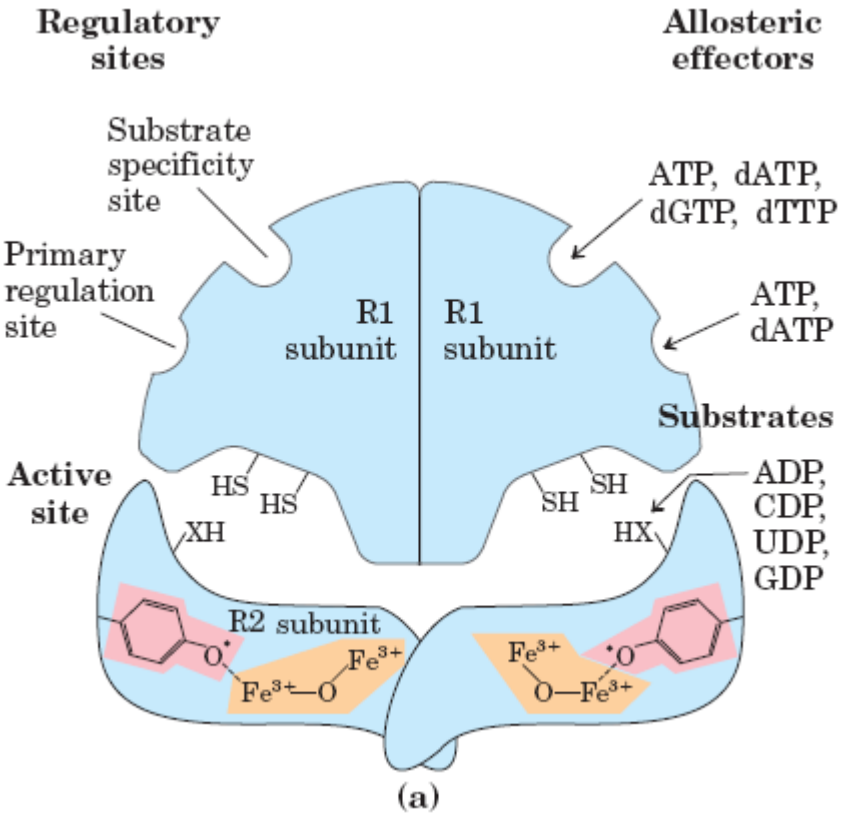

Uridine 5′-triphosphate (UTP)
Gln
Glu
cytidylate synthetase
ATP
ADP + P_i
Cytidine 5′-triphosphate (CTP)

Reduction of ribonucleotides to deoxyribonucleotides by ribonucleotide reductase.

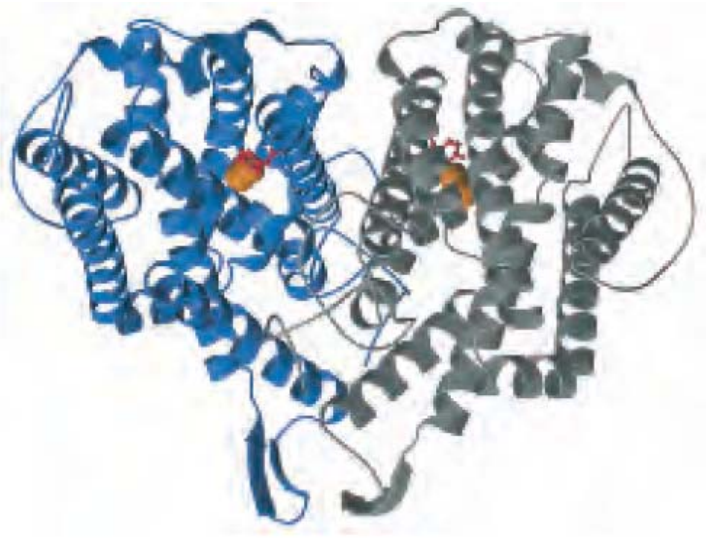


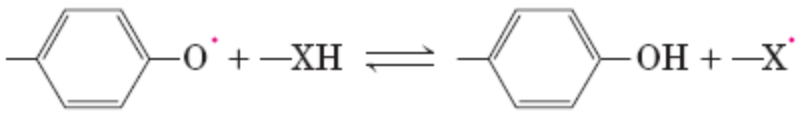
Electrons are transmitted (blue arrows) to the enzyme from NADPH by **(a) glutaredoxin or (b) thioredoxin.** The sulfide groups in glutaredoxin reductase are contributed by two molecules of bound glutathione (GSH; GSSG indicates oxidized glutathione). Note that thioredoxin reductase is a flavoenzyme, with FAD as prosthetic group.

Ribonucleotide reductase



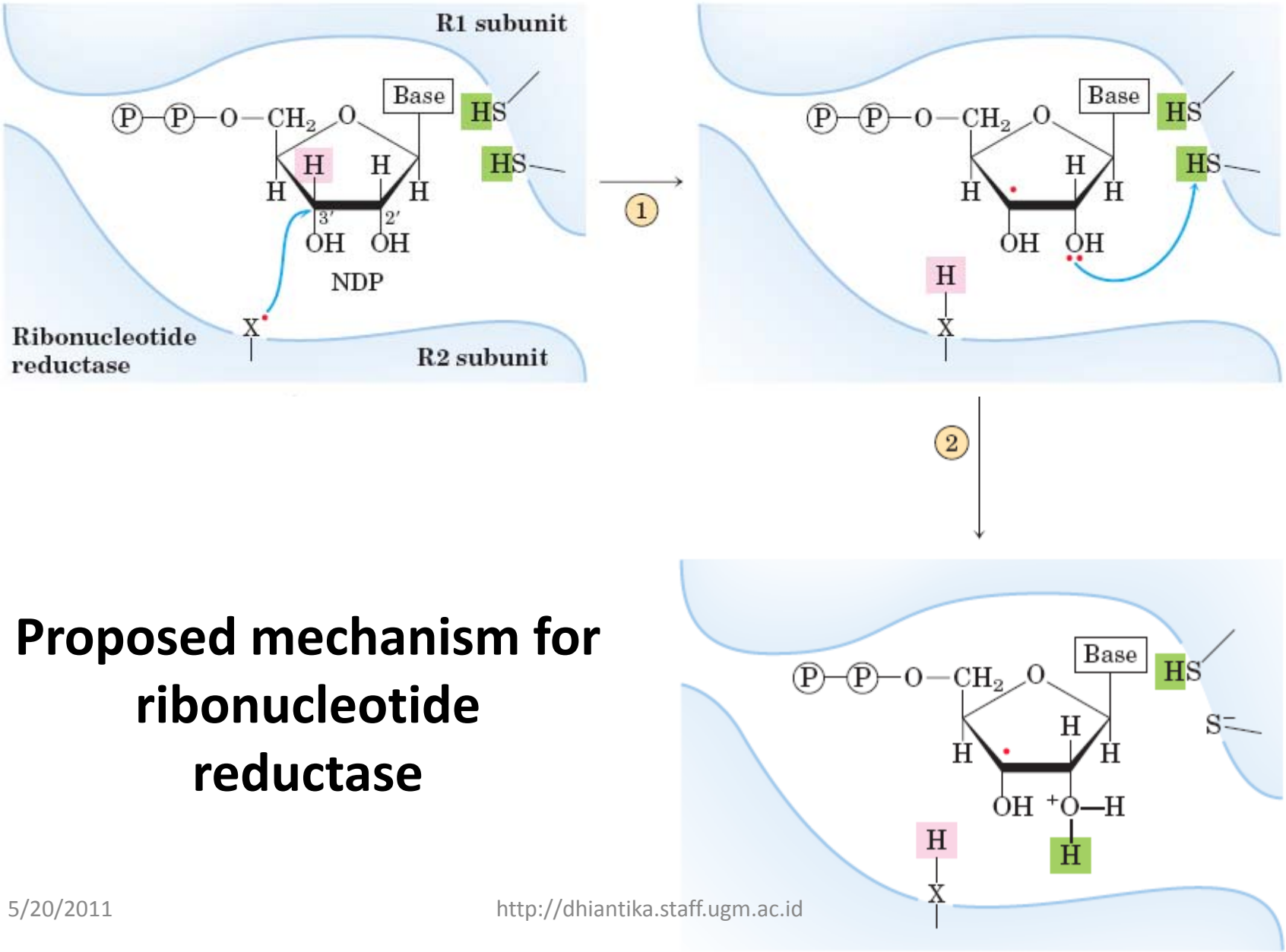
(a)

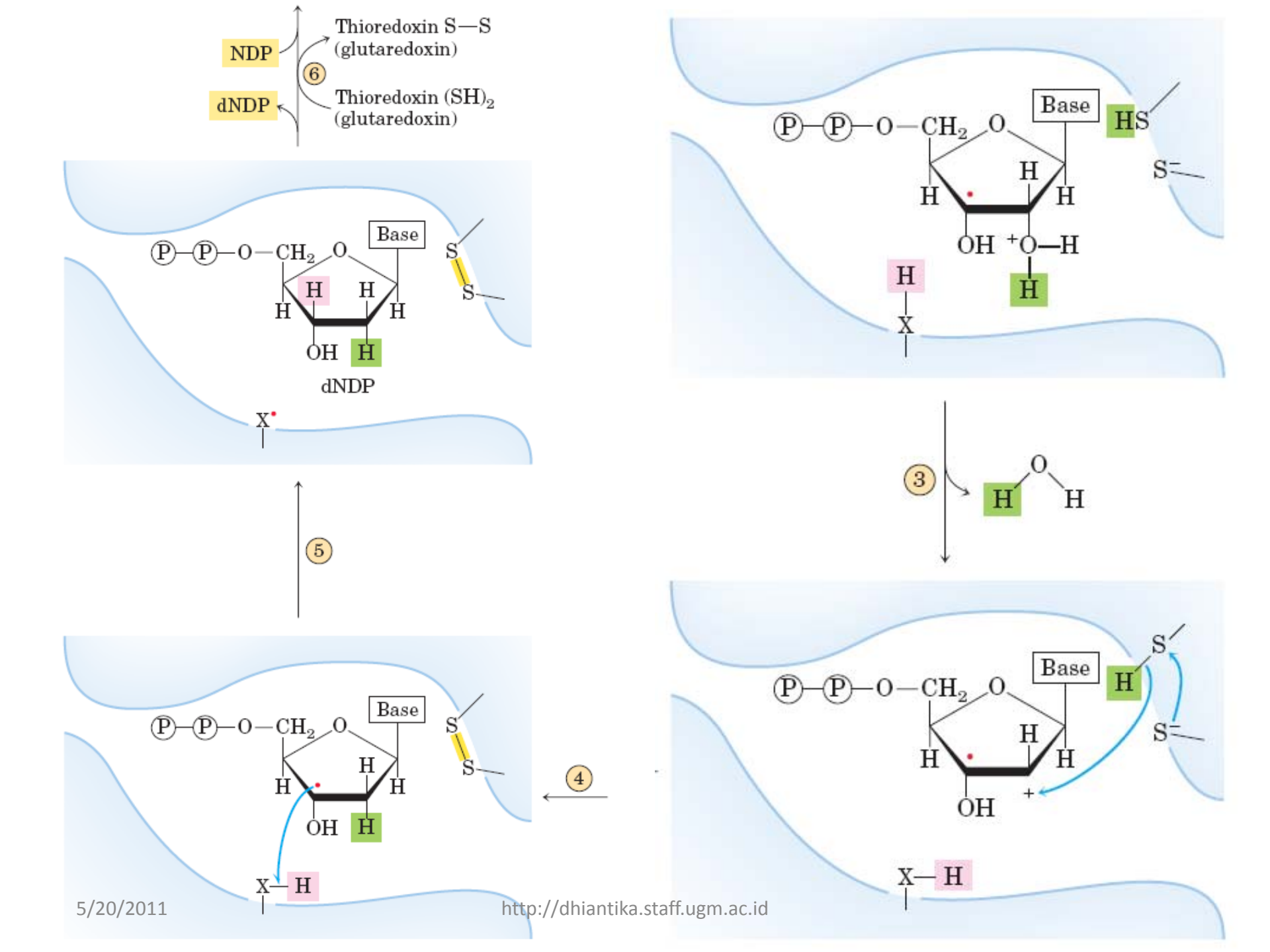

(b)



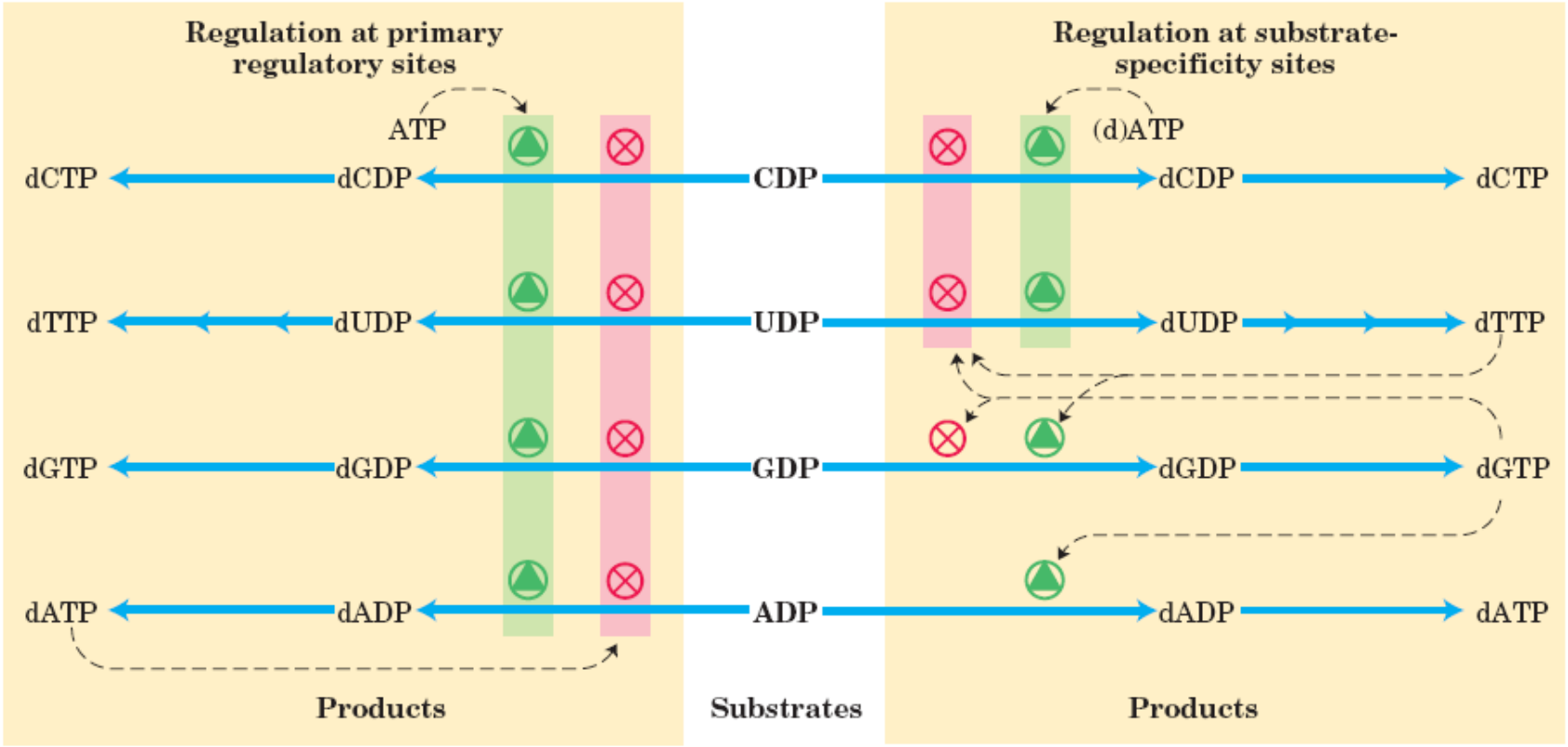
(c)

Proposed mechanism for ribonucleotide reductase

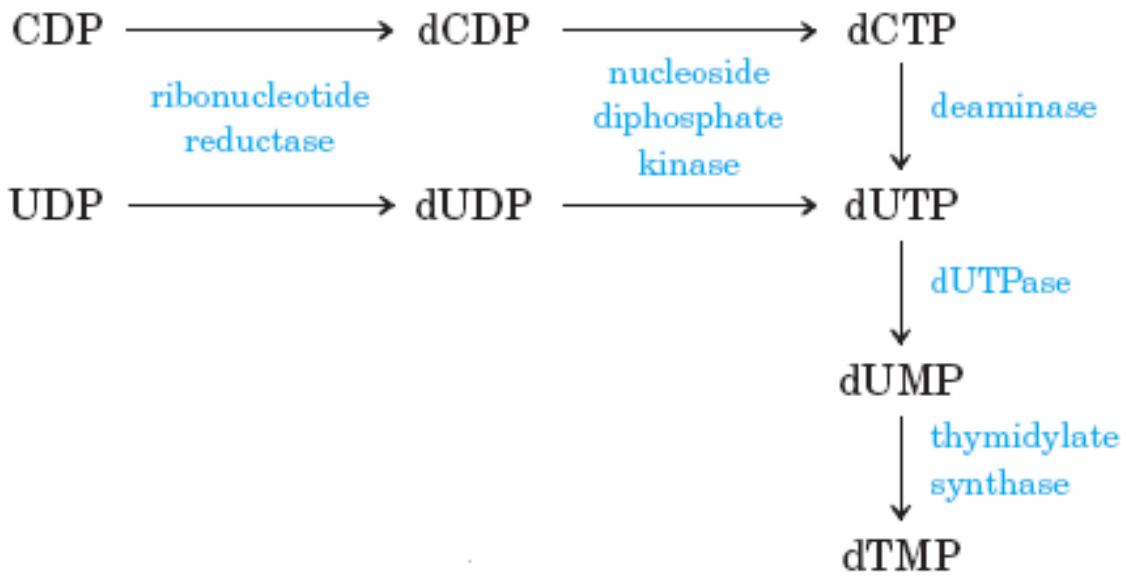



NDP
Thioredoxin S—S
(glutaredoxin)
6
dNDP
Thioredoxin (SH)2
(glutaredoxin)
Base
HS
S
H
OH
+O—H
H
X
3
Base
S
S
H
H
OH
H
dNDP
X•
5
Base
S
S
H
H
OH
H
X—H
4
Base
H
S
S
H
H
OH
+
X—H

Regulation of ribonucleotide reductase by deoxynucleoside triphosphates



Biosynthesis of thymidylate (dTMP)

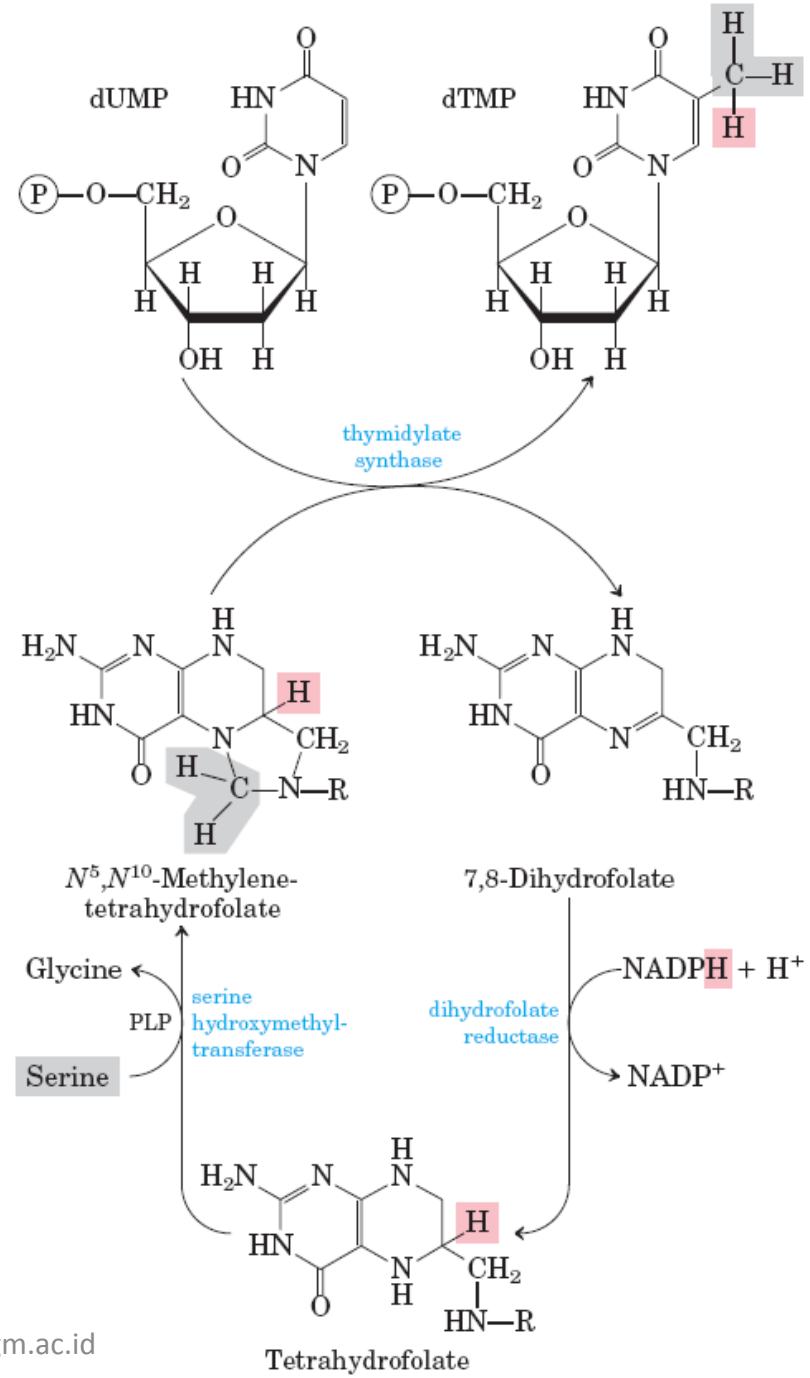


Thymidylate Is Formed by the Methylation of Deoxyuridylate

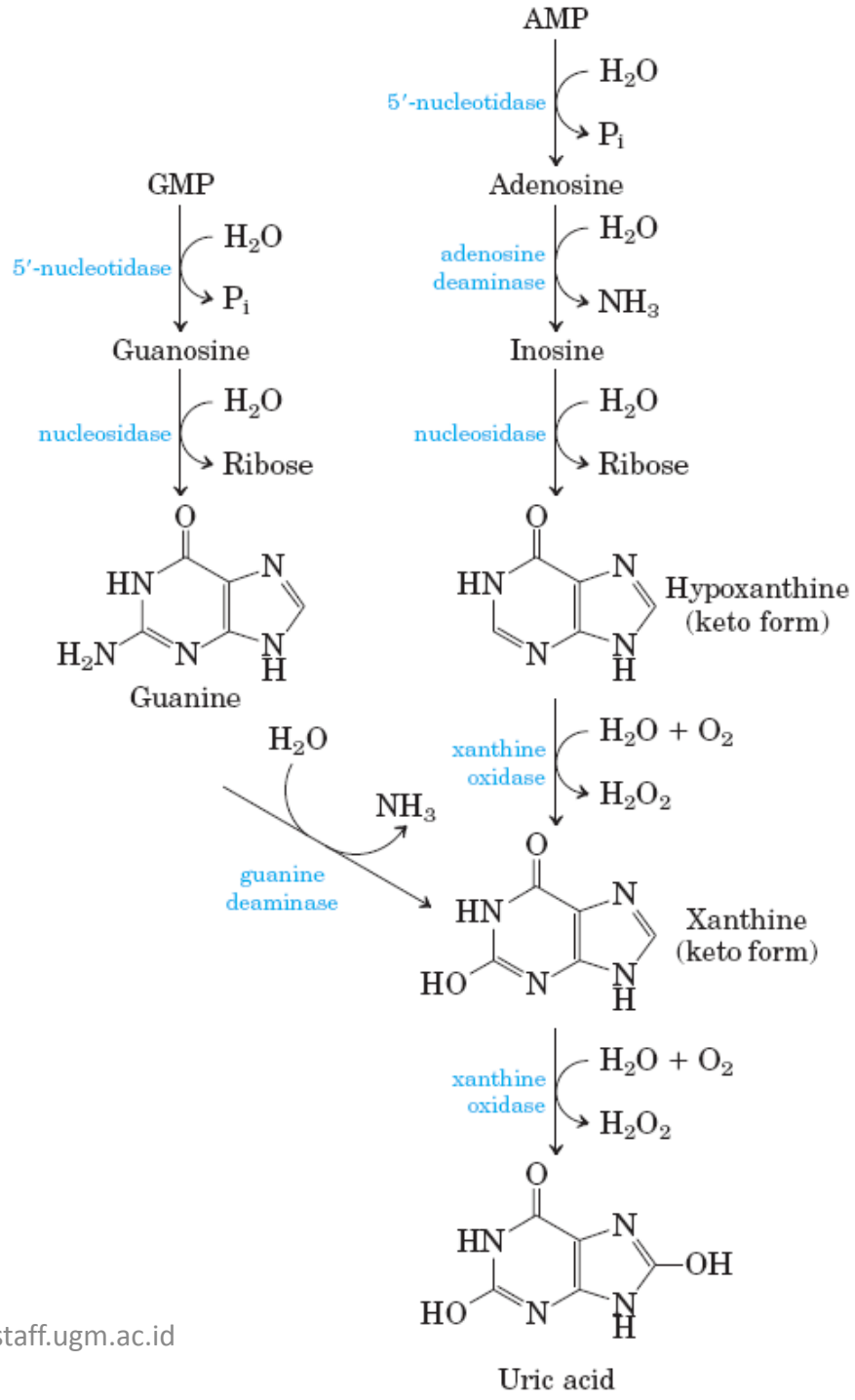
Enzyme : Thymidylate synthase

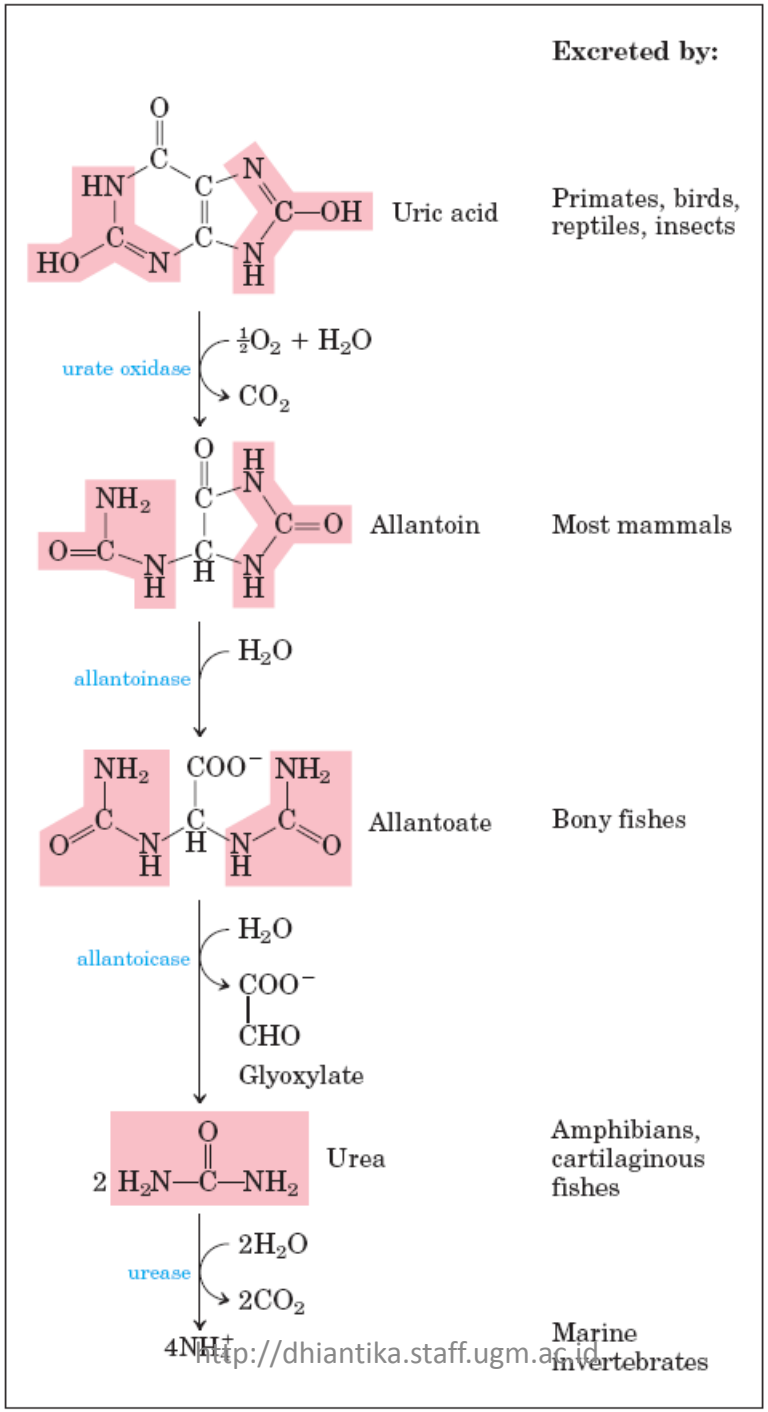
deoxyuridylate (dUMP) is methylated to thymidylate (TMP)

Conversion of dUMP to dTMP by thymidylate synthase and dihydrofolate reductase

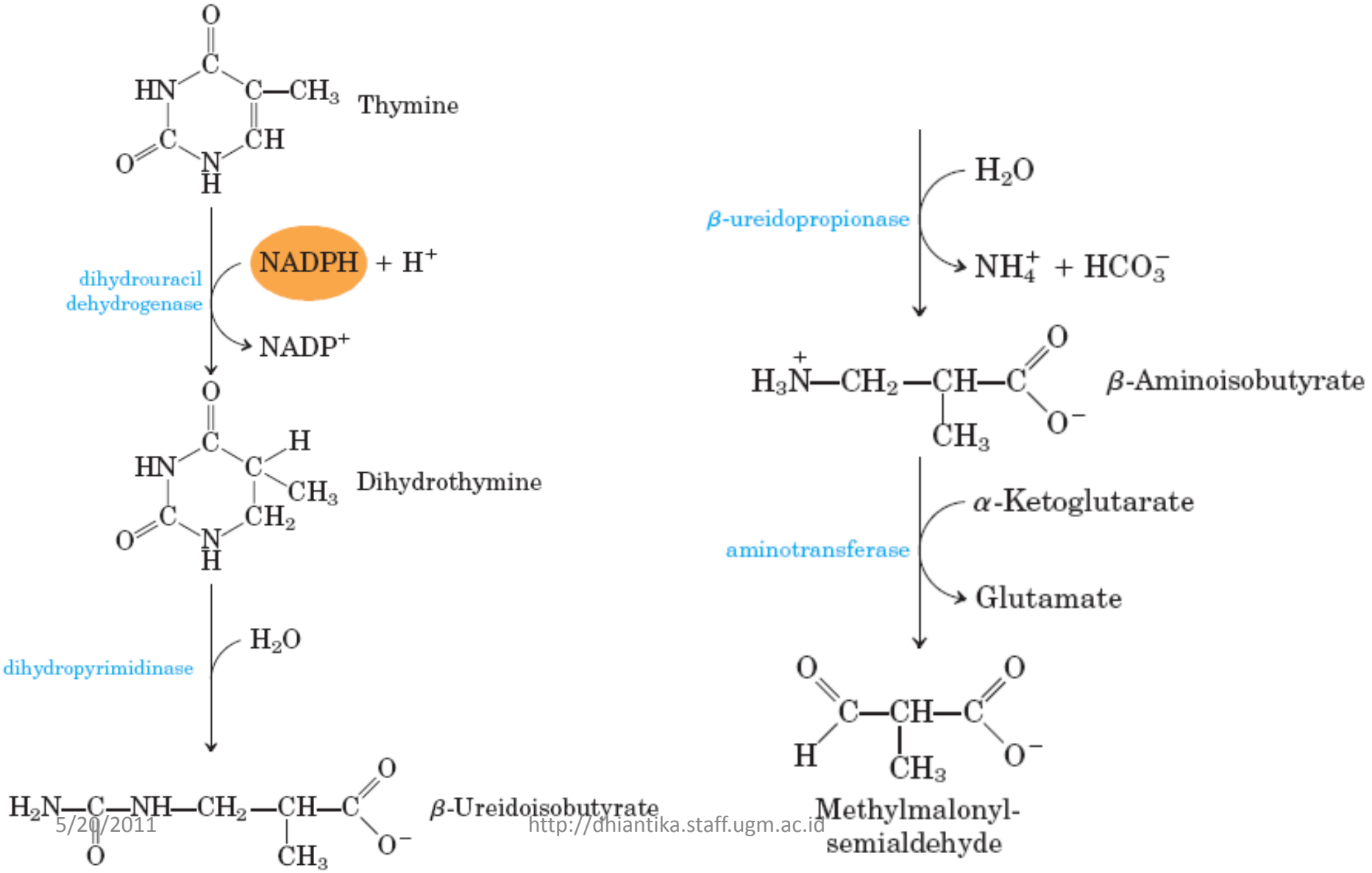


Catabolism of purine nucleotides




Excreted by:
Uric acid
Primates, birds, reptiles, insects
urate oxidase
½O2 + H2O
CO2
Allantoin
Most mammals
allantoinase
H2O
Allantoate
Bony fishes
allantoicase
H2O
COO−
CHO
Glyoxylate
Urea
Amphibians, cartilaginous fishes
urease
2H2O
2CO2
4NH4+
Marine invertebrates

Catabolism of a pyrimidine



- The pathways for degradation of pyrimidines generally lead to NH4 production and thus to urea synthesis.

Excess Uric Acid Causes Gout

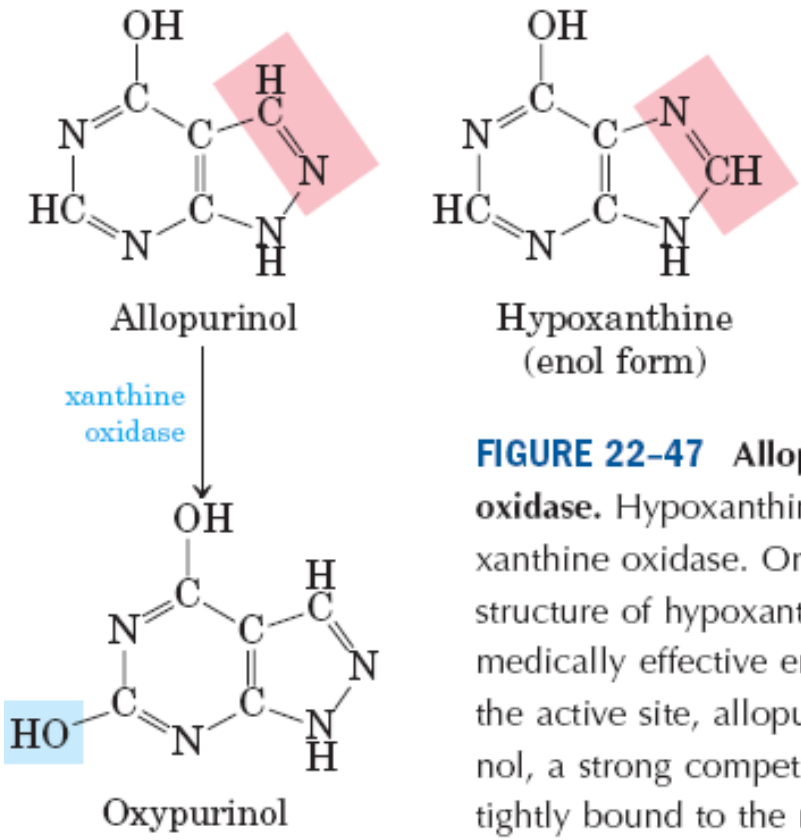


FIGURE 22–47 Allopurinol, an inhibitor of xanthine oxidase. Hypoxanthine is the normal substrate of xanthine oxidase. Only a slight alteration in the structure of hypoxanthine (shaded pink) yields the medically effective enzyme inhibitor allopurinol. At the active site, allopurinol is converted to oxypurinol, a strong competitive inhibitor that remains tightly bound to the reduced form of the enzyme.

Oxypurinol inactivates the reduced form of the enzyme by remaining tightly bound in its active site. When xanthine oxidase is inhibited, the excreted products of purine metabolism are xanthine and hypoxanthine, which are more watersoluble than uric acid and less likely to form crystalline deposits.

Many Chemotherapeutic Agents Target Enzymes in the Nucleotide Biosynthetic Pathways

- Cancer cells have greater requirements for nucleotides as precursors of DNA and RNA, and consequently are generally more sensitive than normal cells to inhibitors of nucleotide biosynthesis

Inhibition glutamine amidotransferases by glutamine analogs

Glutamine is a nitrogen donor in at least half a dozen separate reactions in nucleotide biosynthesis

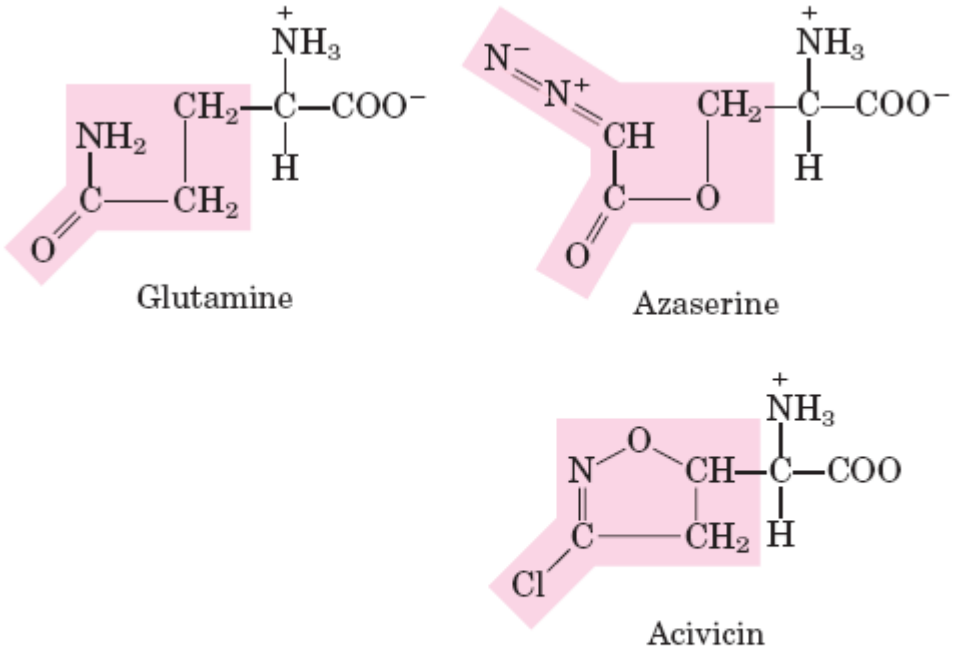
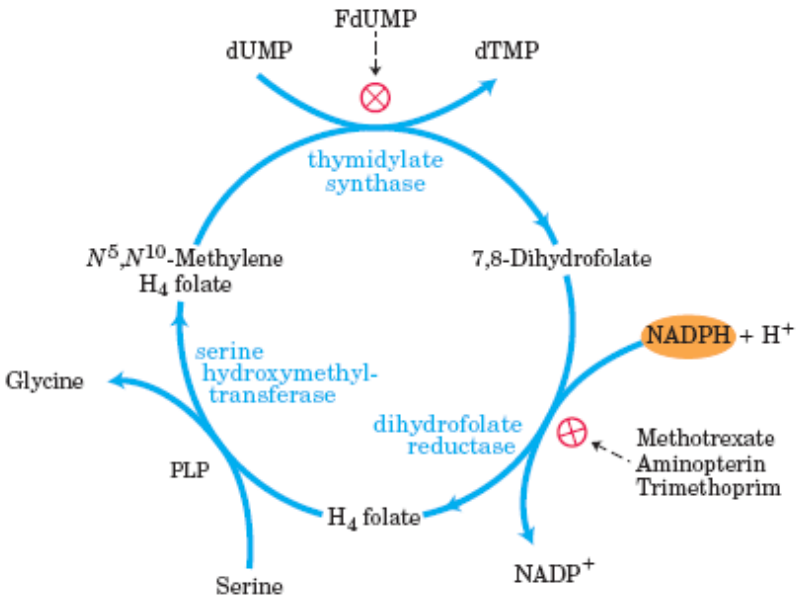


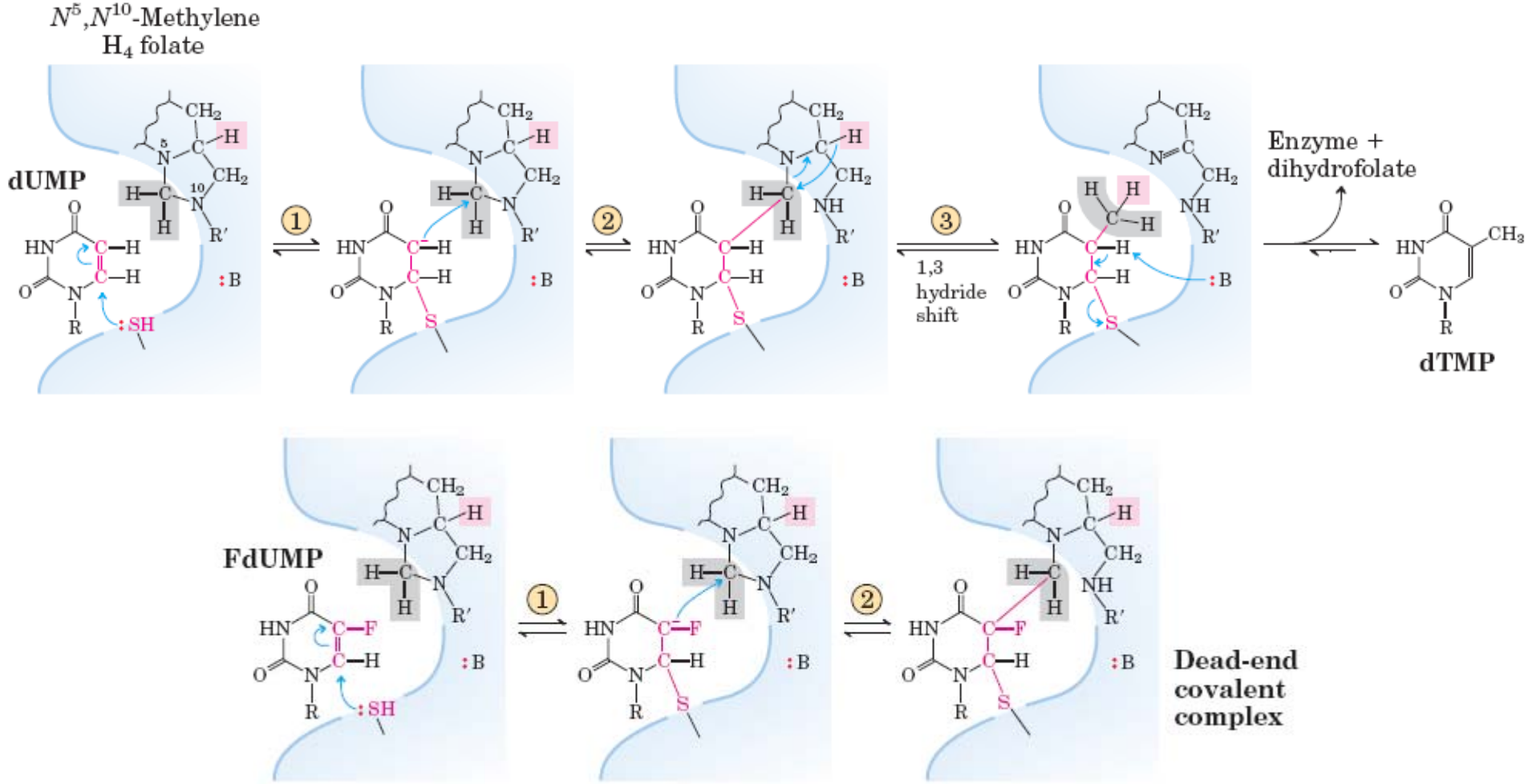
FIGURE 22–48 Azaserine and acivicin, inhibitors of glutamine amidotransferases. These analogs of glutamine interfere in a number of amino acid and nucleotide biosynthetic pathways.

Inhibition thymidylate synthase by **fluorouracil**



- This enzyme→ the only cellular pathway for thymine synthesis
- Fluorouracil itself is not the enzyme inhibitor.
- In the cell, salvage pathways convert it to the deoxynucleoside monophosphate FdUMP, which binds to and inactivates the enzyme

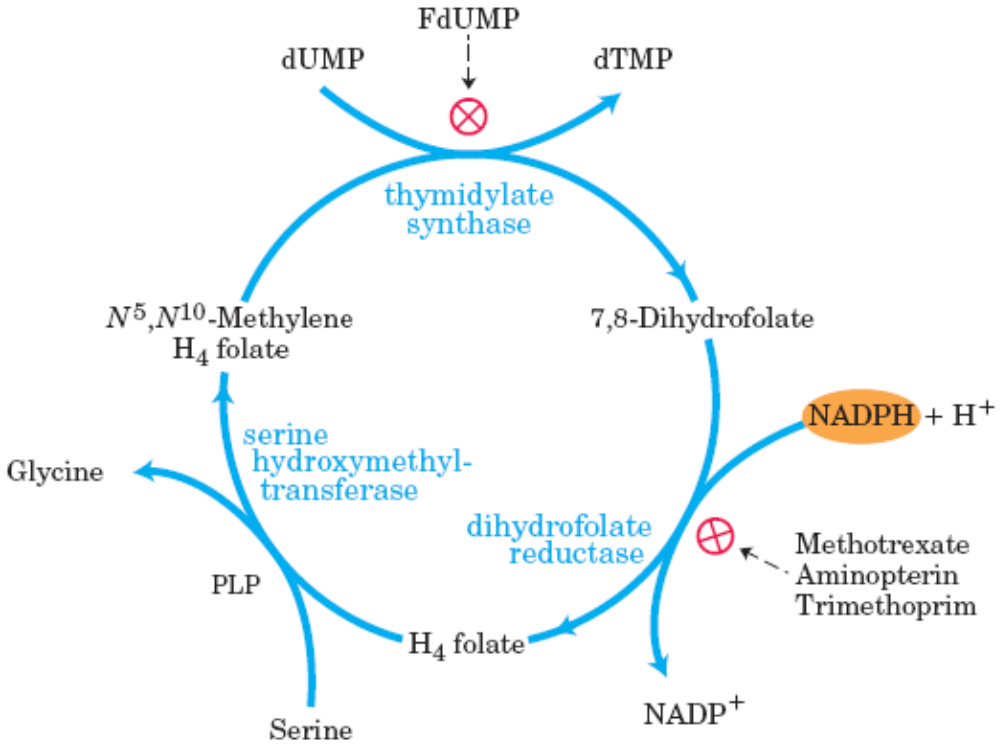
Conversion of dUMP to dTMP and its inhibition by FdUMP

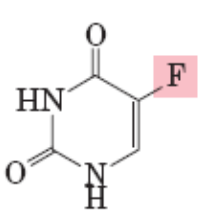


Inhibition dihydrofolate reductase by **methotrexate**

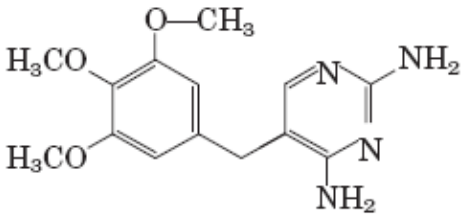
- This folate analog acts as a competitive inhibitor
- the enzyme binds methotrexate with about 100 times higher affinity than dihydrofolate.

Thymidylate synthesis and folate metabolism as targets of chemotherapy

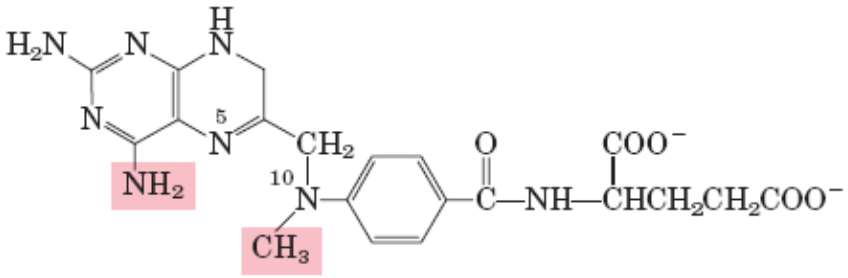




Fluorouracil



Trimethoprim



Methotrexate

(b)

Antibiotic agent

- **Trimethoprim,** binds to bacterial dihydrofolate reductase nearly 100,000 times better than to the mammalian enzyme.
- It is used to treat certain urinary and middle ear bacterial infections.

adenosine deaminase (ADA) deficiency

- severe immunodeficiency disease
- in which T lymphocytes and B lymphocytes do not develop properly
- Lack of ADA leads to a 100-fold increase in the cellular concentration of dATP, a strong inhibitor of ribonucleotide reductase
- High levels of dATP produce a general deficiency of other dNTPs in T lymphocytes
- Individuals with ADA deficiency lack an effective
- immune system and do not survive unless isolated in a sterile “bubble” environment

Salvage Pathways
Specific Enzymes

Adenosine phosphoribosyltransferase

$$\text{Adenine} + \text{PRPP} \longrightarrow \text{adenylate} + PP_i$$

Hypoxanthine-guanine phosphotransferase (HGPRT)

$$\text{Hypoxanthine} + \text{PRPP} \longrightarrow \text{inosinate} + PP_i$$
$$\text{Guanine} + \text{PRPP} \longrightarrow \text{guanylate} + PP_i$$

inosinate (inosine monophosphate, IMP), a precursor of guanylate and adenylate

Lesch-Nyhan syndrome : Mutations in a Salvage-Pathway Enzyme

- A genetic lack of hypoxanthine-guanine phosphoribosyltransferase activity
- Children with this genetic disorder, which becomes manifest by the age of 2 years, are sometimes poorly coordinated and mentally retarded.
- In addition, they are extremely hostile and show compulsive self-destructive tendencies: they mutilate themselves by biting off their fingers, toes, and lips.

- Hypoxanthine and guanine arise constantly from the breakdown of nucleic acids.
- In the absence of hypoxanthineguanine phosphoribosyltransferase, PRPP levels rise and purines are overproduced by the de novo pathway, resulting in high levels of uric acid production and goutlike damage to tissue
- The brain is especially dependent on the salvage pathways, and this may account for the central nervous system damage in children with Lesch-Nyhan syndrome.

Homework

- **Relationship between Folic Acid Deficiency and Anemia Folic acid deficiency,** believed to be the most common vitamin deficiency, causes a type of anemia in which hemoglobin synthesis is impaired and erythrocytes do not mature properly.
- What is the metabolic relationship between hemoglobin synthesis and folic acid deficiency?