BIOCHEMISTRY

Nucleic acid metabolism

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Chapter Description

- Overview

  This chapter is introduces the complex metabolism of nucleic acid.

- Expected Outcomes

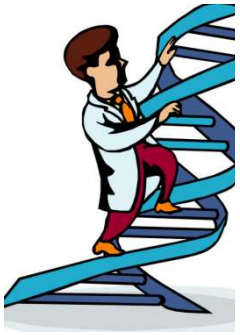
  You should be able to understand importance of nucleic acid other than for genetics, its anabolism and catabolism, compare between salvage and de novo pathways etc.

- Other related Information

  Some relevant questions been provided for improving your understanding of the topic. You are expected to search for external sources for information to adequately answer the questions. All pictures and figures within this chapter categorized as creative commons for the purpose of education only.

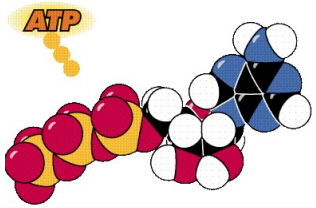
Nucleotide Biosynthesis

- An ample supply of nucleotides is essential for many life processes. Why?



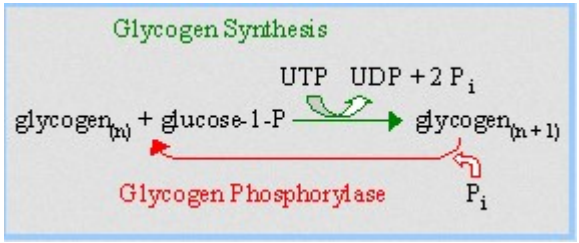
- First, nucleotides are the activated precursors of nucleic acids. As such, they are necessary for the **replication of the genome** and the transcription of the **genetic information** into RNA.

- Second, an adenine nucleotide, ATP, is the **universal currency** of energy.

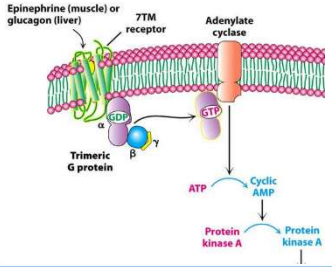


Nucleotide Biosynthesis

- Third, nucleotide derivatives such as UDP-glucose participate in biosynthetic processes such as the **formation of glycogen**.



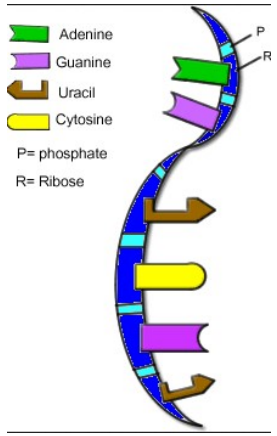
- Fourth, nucleotides are essential components of signal-transduction pathways. Cyclic nucleotides such as cyclic AMP and cyclic GMP are secondary messengers that **transmit signals** both within and between cells.



Nomenclature of bases, nucleosides, and nucleotide:

RNA

Base	Ribonucleoside	Ribonucleotide
Adenine (A)	Adenosine	Adenylate (AMP)
Guanine (G)	Guanosine	Guanylate (GMP)
Uracil (U)	Uridine	Uridylate (UMP)
Cytosine (C)	Cytidine	Cytidylate (CMP)



DNA

Base	Deoxyribonucleoside	Deoxyribonucleotide
Cytosine (C)	Deoxycytidine	Deoxycytidylate (dCMP)
Adenine (A)	Deoxyadenosine	Deoxyadenylate (dAMP)
Guanine (G)	Deoxyguanosine	Deoxyguanylate (dGMP)
Thymine (T)	Thymidine	Thymidylate (TMP)

Overview

- The pathways for the biosynthesis of nucleotides fall into two classes: ***de novo pathways*** and ***salvage pathways*** . In *de novo* (from scratch) pathways, the nucleotide bases are assembled from simpler compounds.

- The framework for a ***pyrimidine*** *base is assembled first and then attached to ribose. In contrast, the framework for a* ***purine*** *base is* synthesized piece by piece directly onto a ribose-based structure.

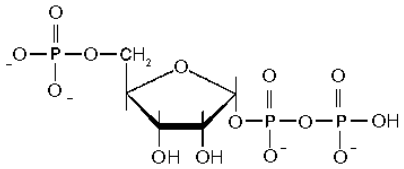
Overview

- Both *de novo* and salvage pathways lead to the synthesis of ***ribonucleotides***.

- *However, DNA is built from deoxyribonucleotides. Consistent with the notion that RNA preceded DNA in the course of evolution,* ***all deoxyribonucleotides are synthesized from*** *the corresponding* ***ribonucleotides***.

- The deoxyribose sugar is generated by the reduction of ribose within a fully formed nucleotide. Furthermore, the methyl group that distinguishes the **thymine** of DNA from the **uracil** of RNA is added at the last step in the pathway.

- Salvage and *de novo* Pathways: In a salvage pathway, a base is reattached to a ribose, activated in the form of 5-phosphoribosyl-1-pyrophosphate (PRPP).



PRPP

SALVAGE PATHWAY

Activated ribose (PRPP) + base

↓

Nucleotide

- In *de novo* synthesis, the base itself is synthesized from simpler starting materials, including amino acids. ATP hydrolysis is required for de novo synthesis.

DE NOVO PATHWAY

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

↓

Nucleotide

de novo Pyrimidine (ring) biosynthesis

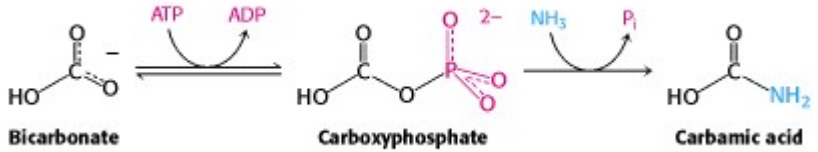
- In *de novo* synthesis of pyrimidines, the **ring** is **synthesized first** and **then** it is **attached to ribose** to form a *pyrimidine nucleotide.*

- *Pyrimidine rings are assembled from* **bicarbonate, aspartic acid, and ammonia.** *Although* ammonia can be used directly, it is usually produced from the hydrolysis of the side chain of **glutamine**.

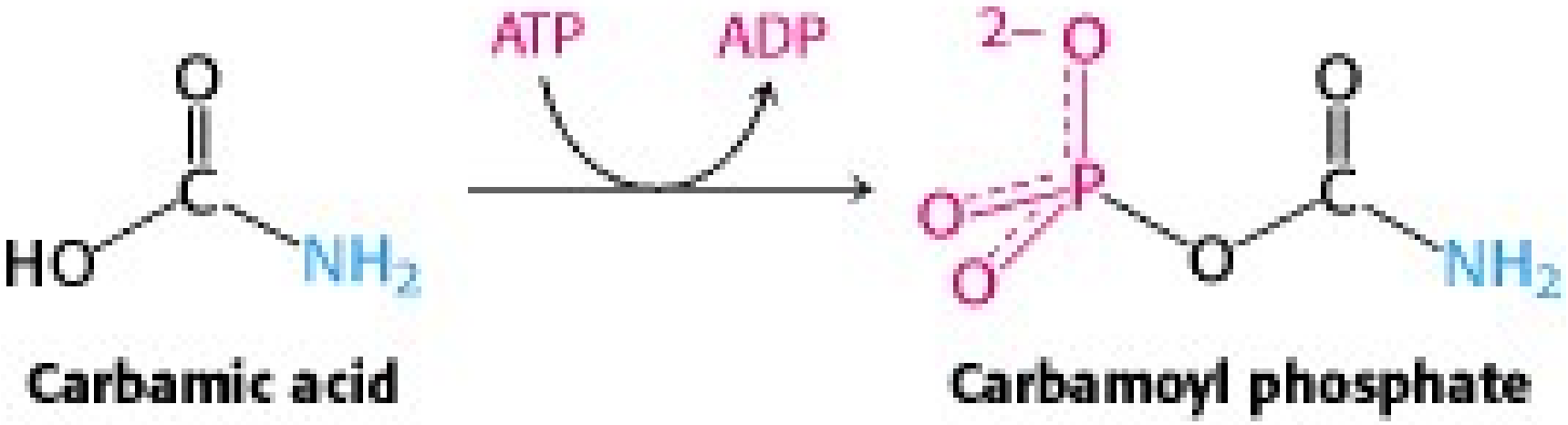
- The first step in *de novo* pyrimidine biosynthesis is the synthesis of ***carbamoyl phosphate*** *from* ***bicarbonate*** *and* **ammonia** in a multistep process, requiring the cleavage of two molecules of ATP.

- This reaction is catalyzed by ***carbamoyl phosphate synthetase (CPS).*** Analysis of the structure of CPS reveals two homologous domains, each of which catalyzes an ATP-dependent step.

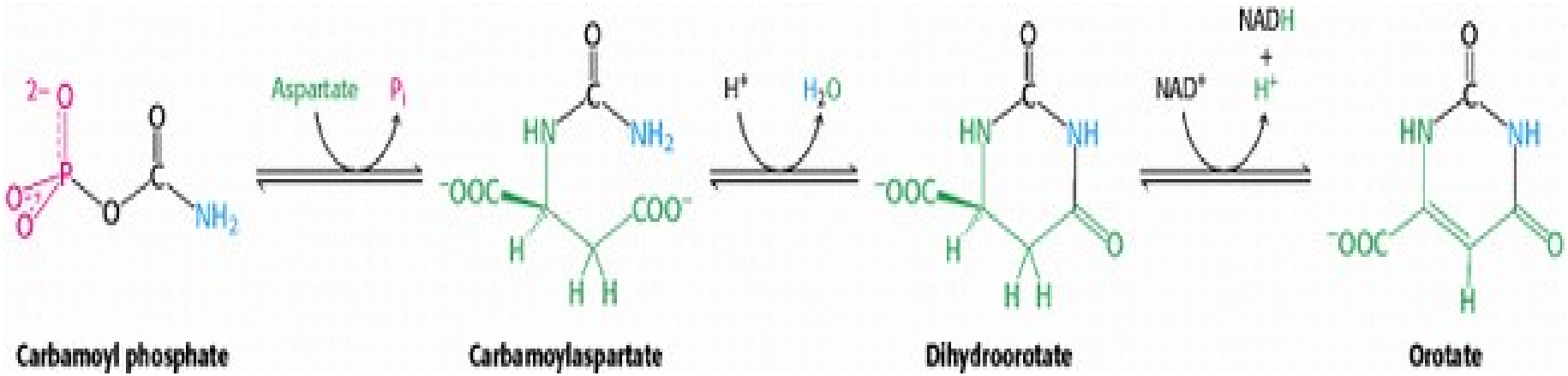
1. In the first step of the carbamoyl phosphate synthesis pathway, bicarbonate is phosphorylated by ATP to form carboxyphosphate and ADP. Ammonia then reacts with carboxyphosphate to form carbamic acid and inorganic phosphate.



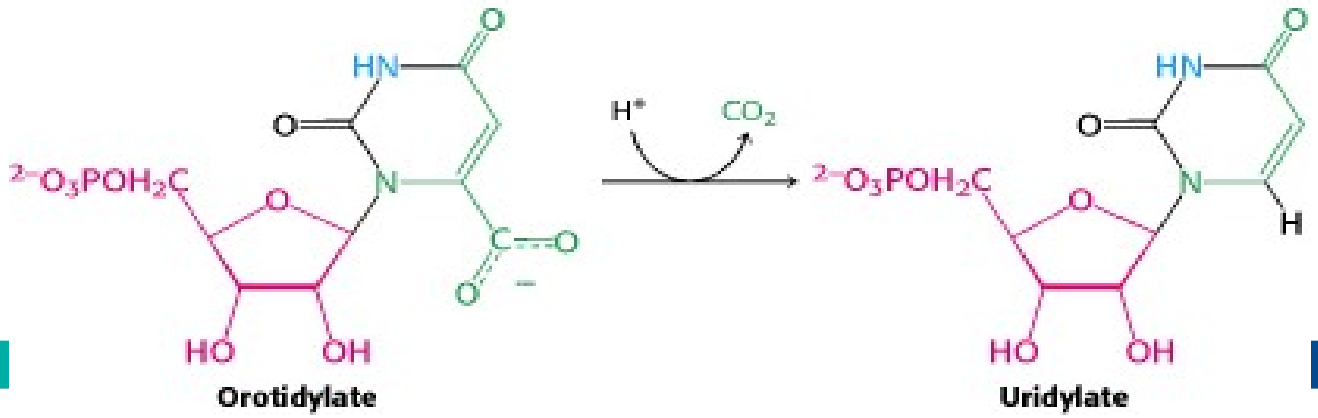
2. In the final step catalyzed by carbamoyl phosphate synthetase, carbamic acid is phosphorylated by another molecule of ATP to form carbamoyl phosphate. This reaction takes place in a second ATP-grasp domain



- **Carbamoyl phosphate** reacts with **aspartate** to form **carbamoylaspartate** in a reaction catalyzed by *aspartate transcarbamoylase*. **Carbamoylaspartate** *then cyclizes to form* **dihydroorotate** *which is then oxidized by* NAD+ to form **orotate**.



- At this stage, **orotate couples to ribose**, in the form of *5-phosphoribosyl-1-pyrophosphate (PRPP), a form of ribose* activated to accept nucleotide bases.
- **PRPP** is synthesized from **ribose-5-phosphate**, formed by the **pentose phosphate pathway**, by the addition of pyrophosphate from ATP.
- **Orotate** reacts with PRPP to form **orotidylate**, *a pyrimidine* nucleotide. This reaction is driven by the hydrolysis of pyrophosphate. The enzyme that catalyzes this addition is *pyrimidine phosphoribosyl transferase.*
- **Orotidylate** is then decarboxylated to form **uridylate** *(UMP), a major* pyrimidine nucleotide that is a precursor to RNA. This reaction is catalyzed by *orotidylate decarboxylase.*

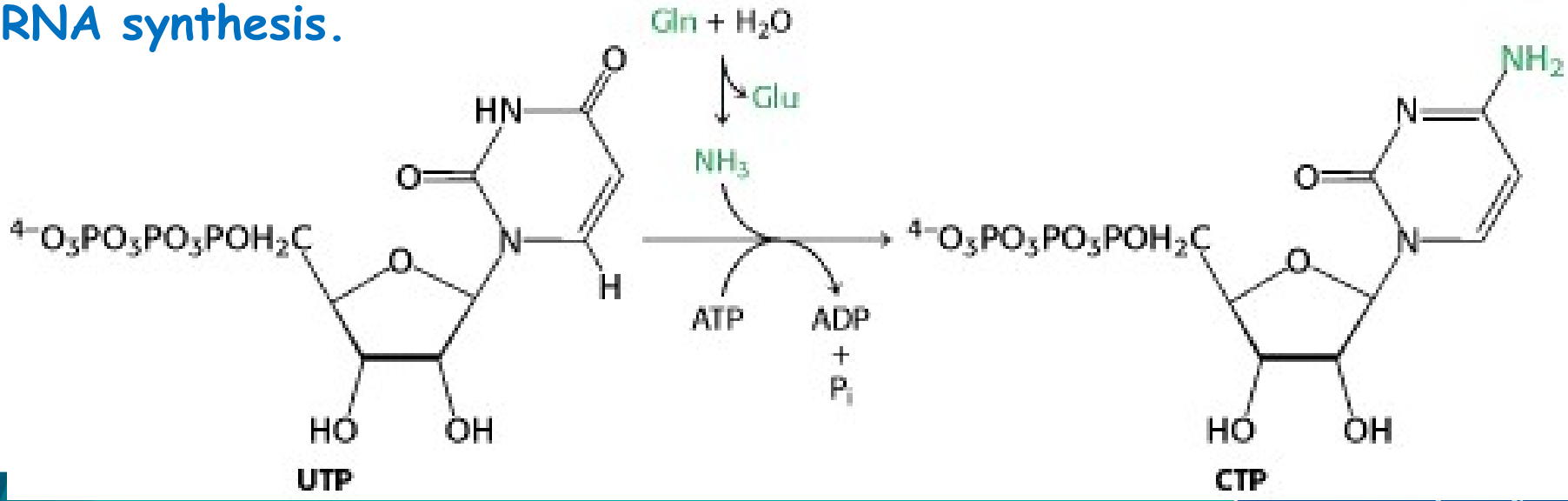


Orotidylate Uridylate

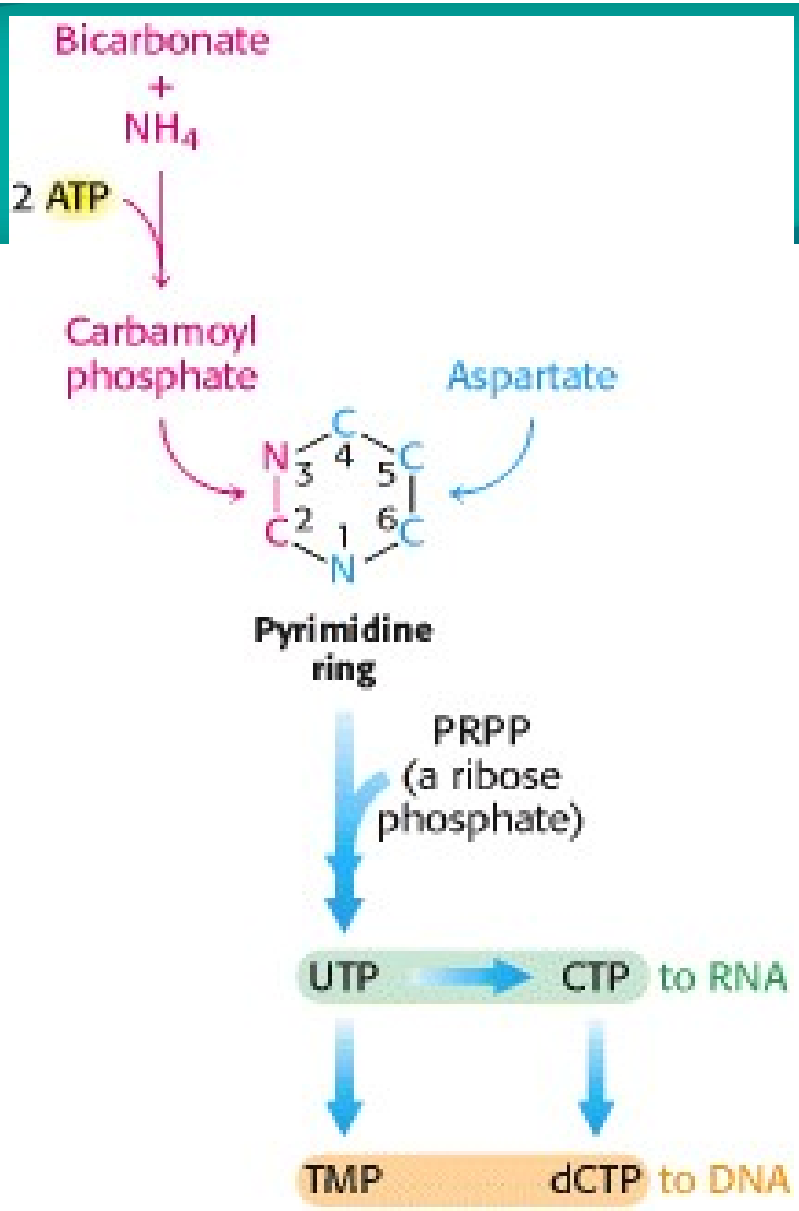
- After **uridine triphosphate** has been formed, it can be transformed into **cytidine triphosphate** by the replacement of a carbonyl group by an amino group.

- Like the synthesis of carbamoyl phosphate, this reaction **requires ATP and uses glutamine** as the source of the amino group.

- CTP can then be used in many biochemical processes, including RNA synthesis.



Summary:


Bicarbonate
+
NH_4
2 ATP
Carbamoyl phosphate
Aspartate
Pyrimidine ring
PRPP (a ribose phosphate)
UTP
CTP
to RNA
TMP
dCTP
to DNA

Purine biosynthesis

- Purine nucleotides can be synthesized in **two distinct pathways**. First, purines are synthesized **de novo**, beginning with simple starting materials such as amino acids and bicarbonate .

- Unlike the case for pyrimidines, the purine bases are **assembled** already attached **to the ribose ring from the start**.

- Alternatively, purine bases, released by the hydrolytic degradation of nucleic acids and nucleotides, can be **salvaged** and **recycled**. Purine salvage pathways are especially noted for the **energy** that they **save** .

Salvage Pathways Economize Intracellular Energy Expenditure

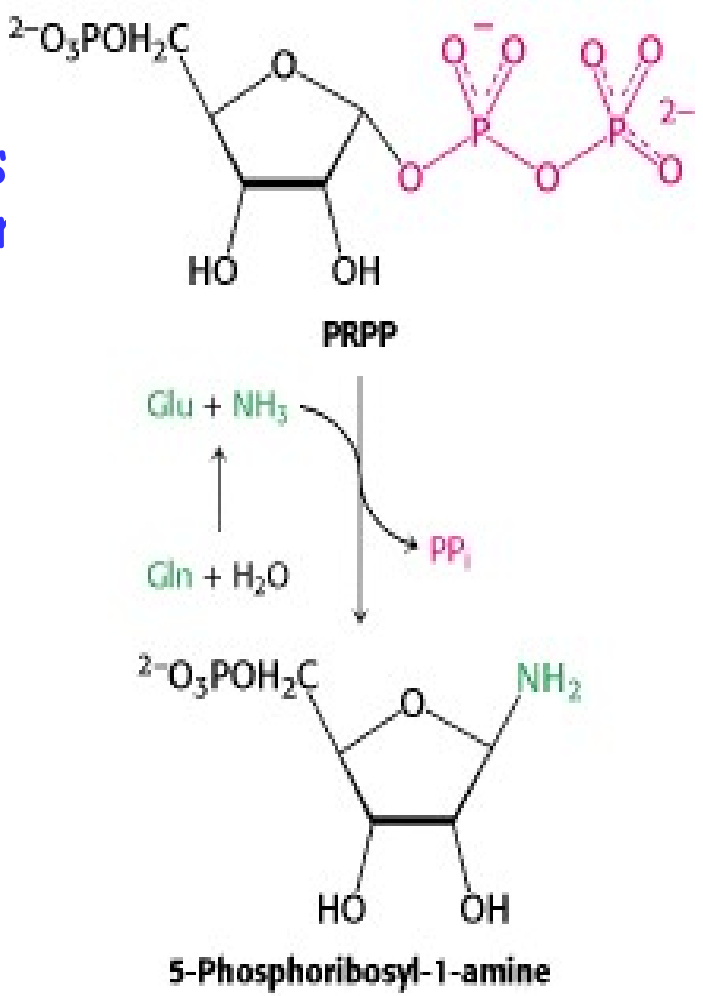
- Free purine bases, derived from the **turnover of nucleotides or from the diet**, can be attached to PRPP to form purine nucleoside monophosphates, in a reaction analogous to the formation of orotidylate.

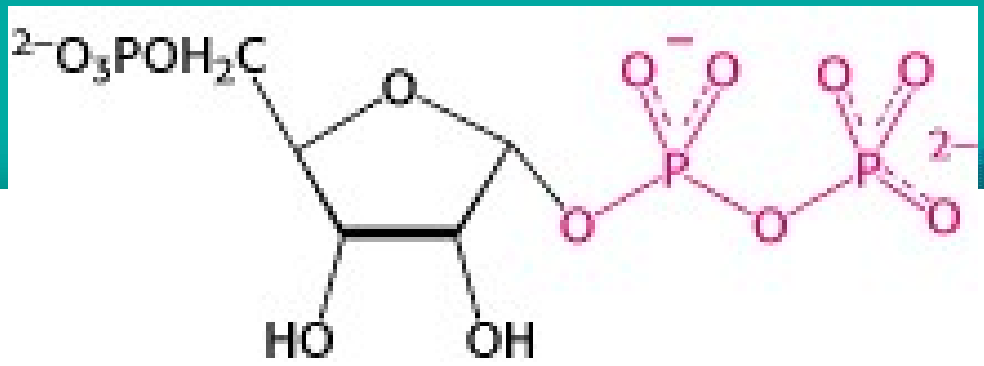
- Salvage enzymes with **different specificities** recover purine bases. **Adenine phosphoribosyl transferase** catalyzes the formation of adenylate

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

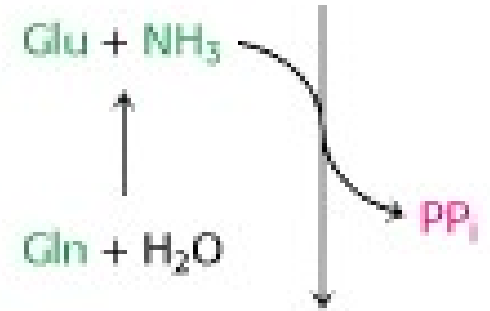
Purine *de novo* biosynthesis

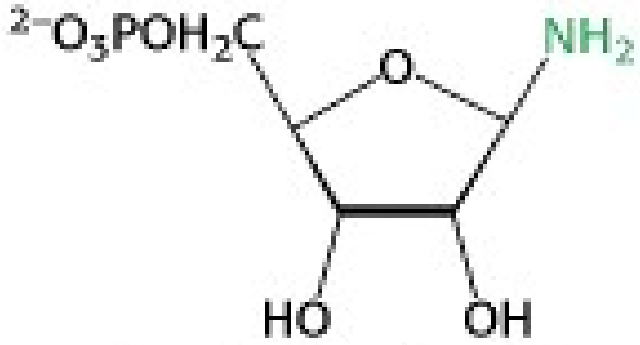
- Purine *de novo* biosynthesis, like pyrimidine biosynthesis, requires PRPP, but for purines, **PRPP** provides the foundation on which the bases ar **constructed** step by step **from** the **beginning**.
- The initial committed step is the **displacement of pyrophosphate by ammonia**, rather than by a pre-assembled base, to produce 5-phosphoribosyl-1-amine, *with the amine in the* β configuration.
- Glutamine phosphoribosylamido transferase catalyzes this reaction.





PRPP

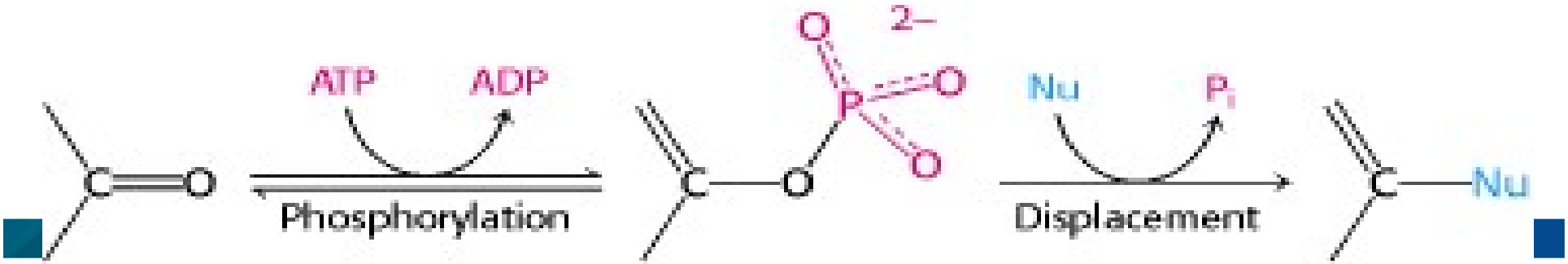




5-Phosphoribosyl-1-amine

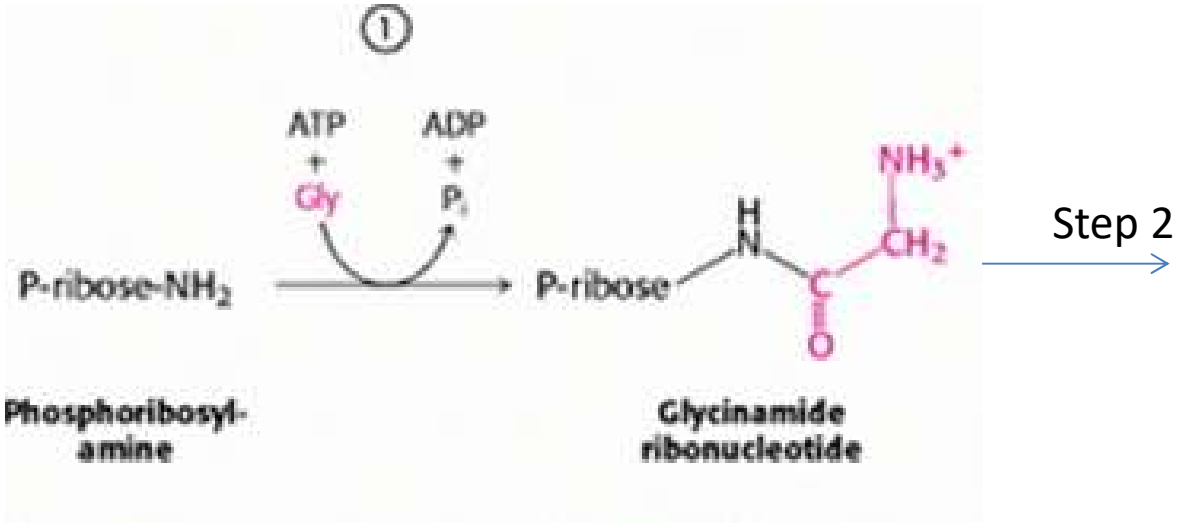
The Purine Ring Is Assembled by Successive Steps of Activation by Phosphorylation Followed by Displacement

- **Nine additional steps** are required to assemble the purine ring. Remarkably, the **first six steps** are **analogous reactions**.
- Most of these steps are catalyzed by enzymes with ATP-grasp domains that are homologous to those in carbamoyl phosphate synthetase.
- Each step consists of the **activation of a carbon-bound oxygen** atom (typically a carbonyl oxygen atom) by **phosphorylation**, followed by the **displacement of a phosphoryl group** by **ammonia** or **an amine group** acting as a **nucleophile (Nu)**.

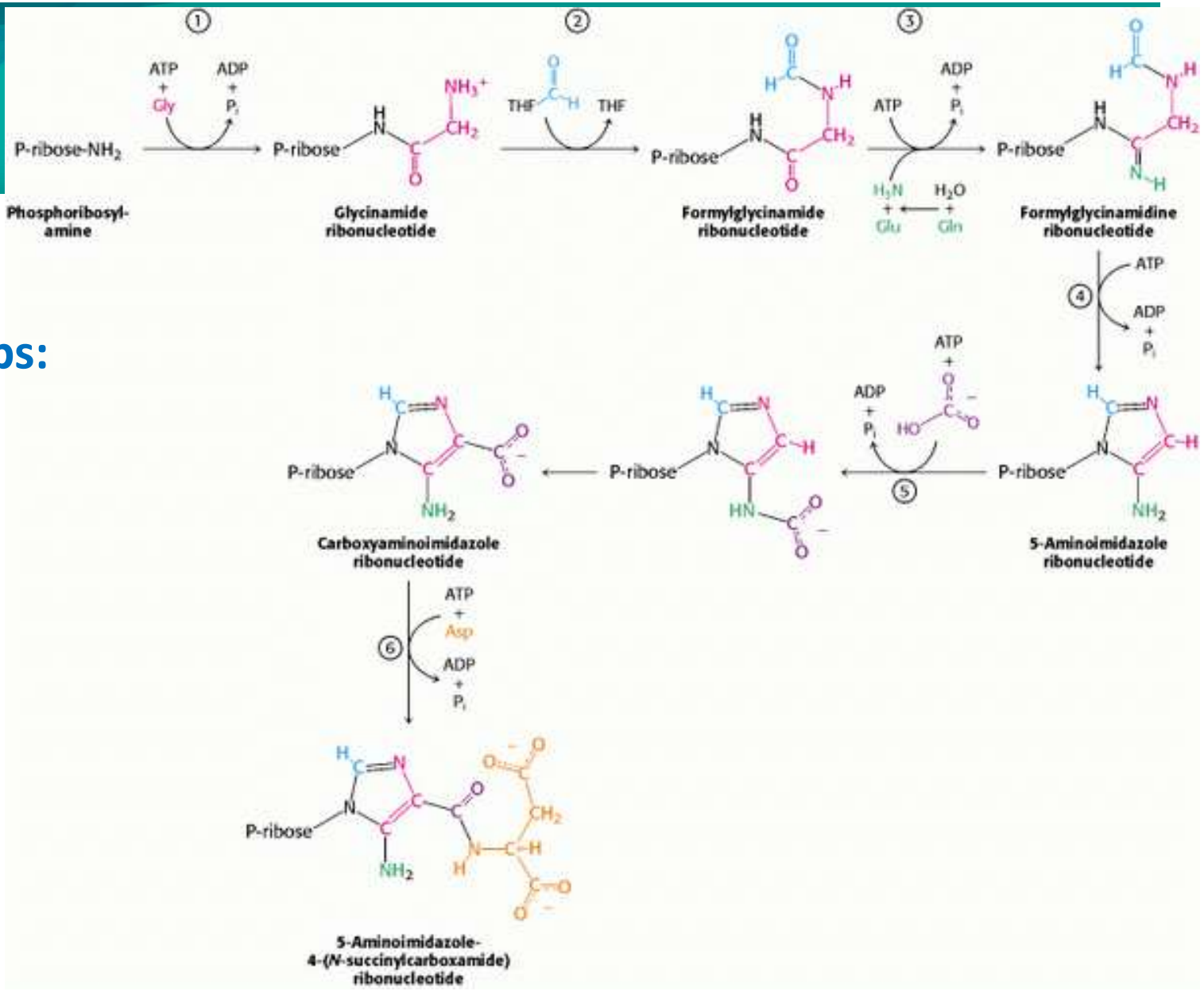


- **For example,** in the next step following the formation of phosphoribosyl amine, the carboxylate group of a **glycine** residue is **activated by phosphorylation** and then coupled to the amino group of phosphoribosylamine.

- ***A new amide bond*** is formed while the amino group of **glycine** is free to **act as a nucleophile** in the next step.



Further steps:

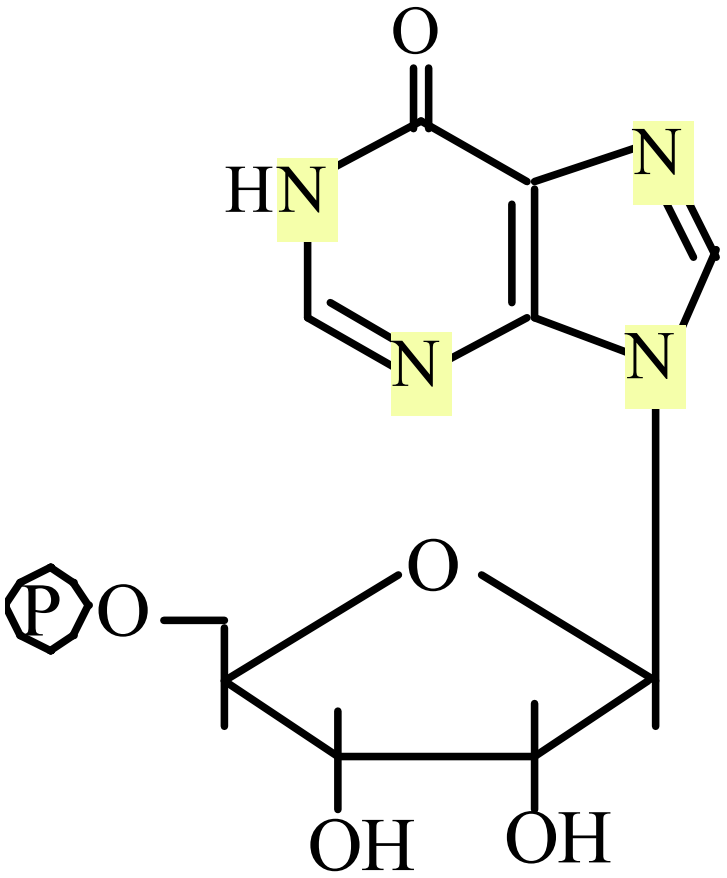


11 steps (Summary):

- Step 1: Activation of the anomeric carbon of ribose
- Step 2: Formation of the N-glycosidic bond
- Steps 3, 4 , 6: Building and closing the imidazole ring
- Steps 5 and 7 - 11: Building and closing the pyrimidine ring

The First Purine formed: Inosine Monophosphate (IMP)

(Note: the base is hypoxanthine)



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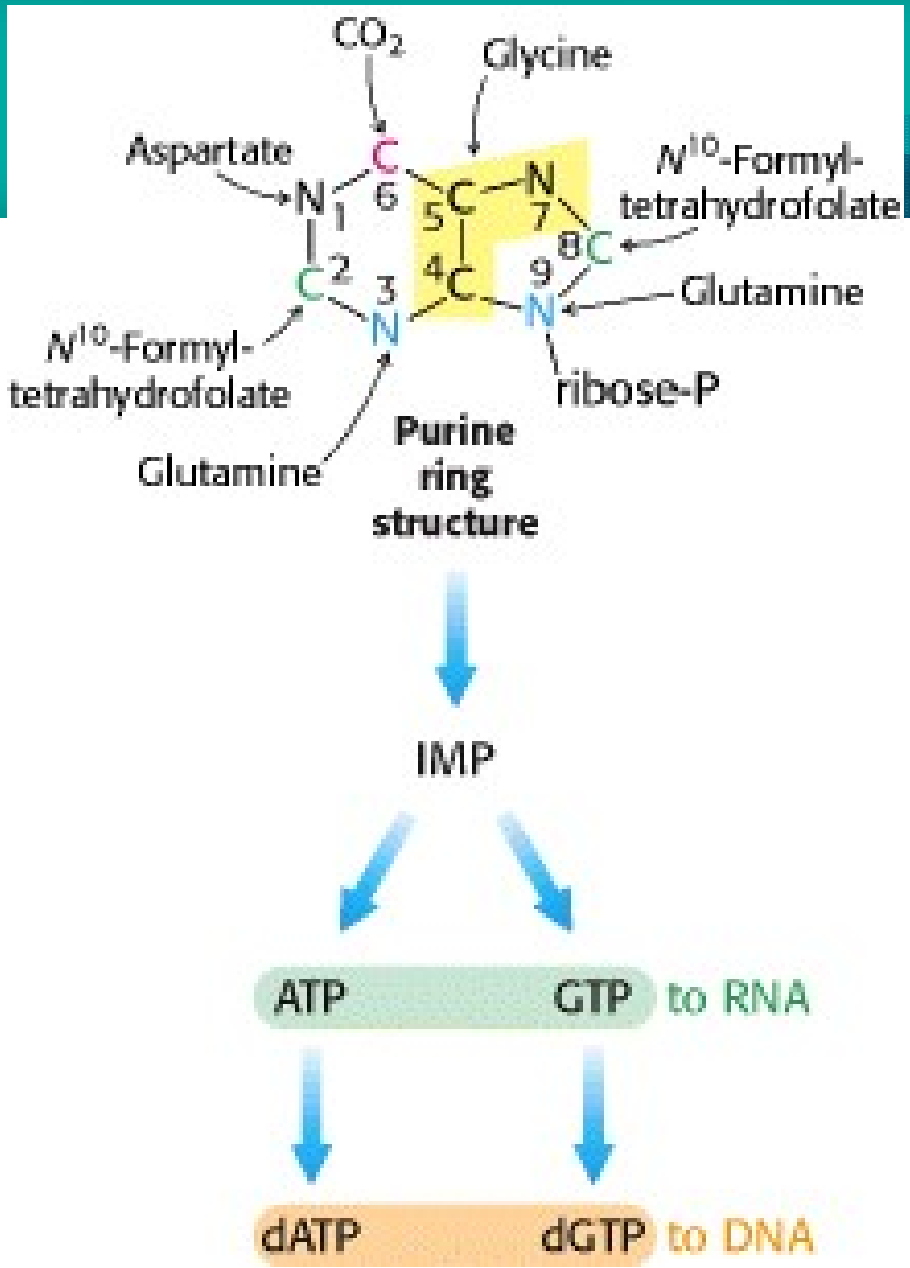
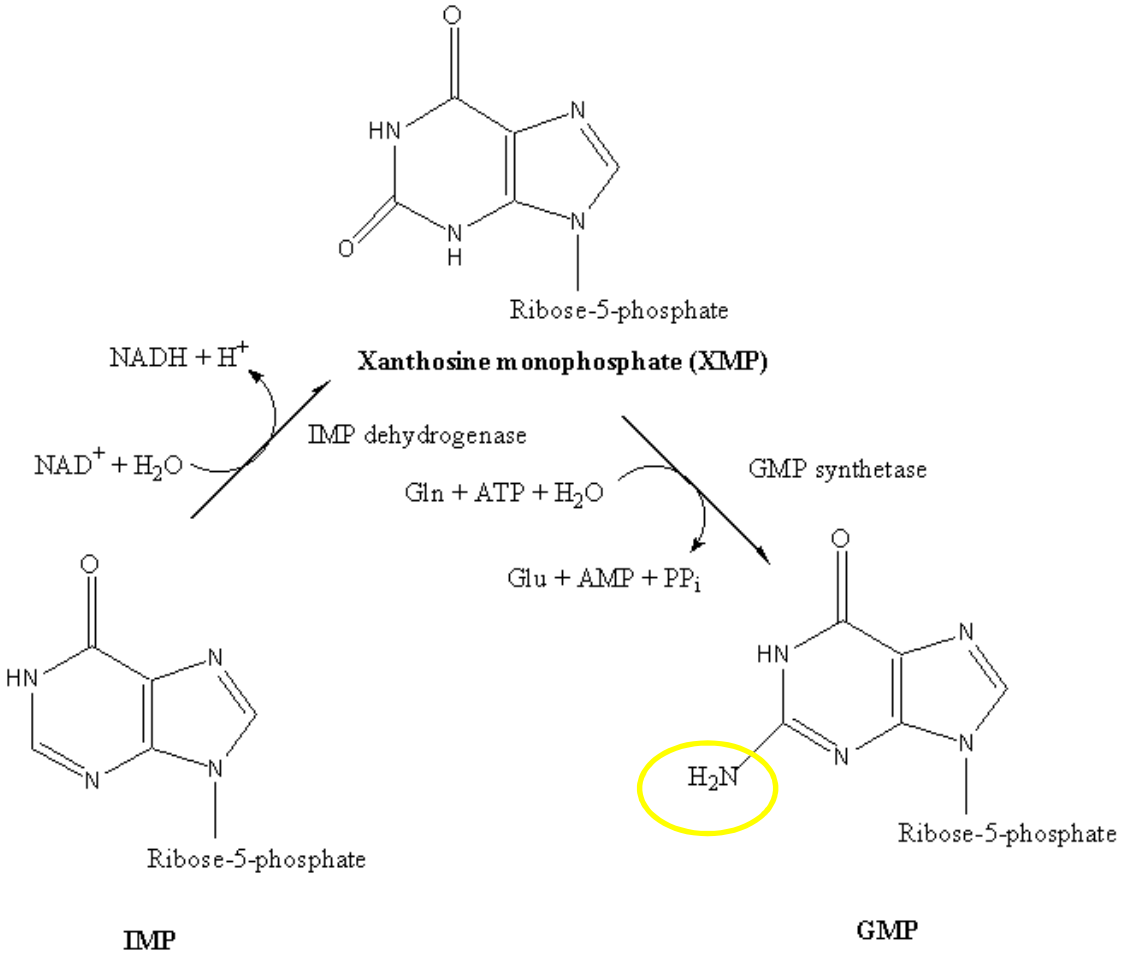
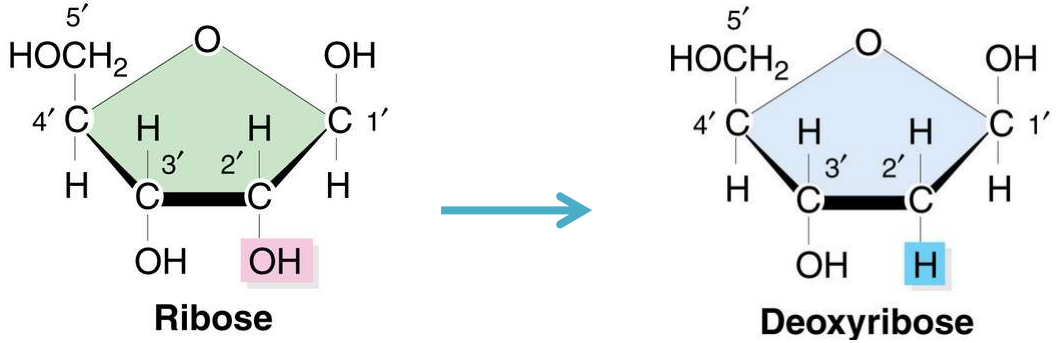

CO_2
Glycine
Aspartate
N^{10}-Formyl-tetrahydrofolate
Glutamine
N^{10}-Formyl-tetrahydrofolate
ribose-P
Glutamine
Purine ring structure
IMP
ATP
GTP
to RNA
dATP
dGTP
to DNA

AMP and GMP are formed from IMP



- We turn now to the **synthesis of deoxyribonucleotides**. These precursors of DNA are formed by the **reduction of ribonucleotides** specifically, the **2 -hydroxyl group** on the ribose moiety is replaced by a hydrogen atom.



- The substrates are ribonucleoside diphosphates or triphosphates, and the ultimate reductant is **NADPH**.

Biosynthesis of thymidylate

- Uracil, produced by the pyrimidine synthesis pathway, is not a component of DNA. Rather, **DNA contains thymine**, *a* methylated analogue of uracil.

- Another step is required to **generate thymidylate from uracil**. *Thymidylate synthetase* performs this and deoxyuridylate (dUMP) is methylated to thymidylate (TMP).

NUCLEIC ACID CATABOLISM

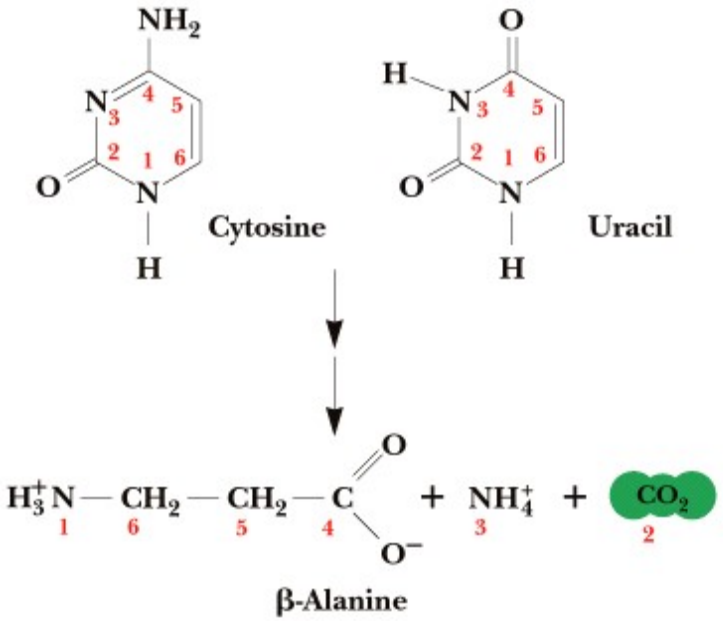
Catabolism of Pyrimidine

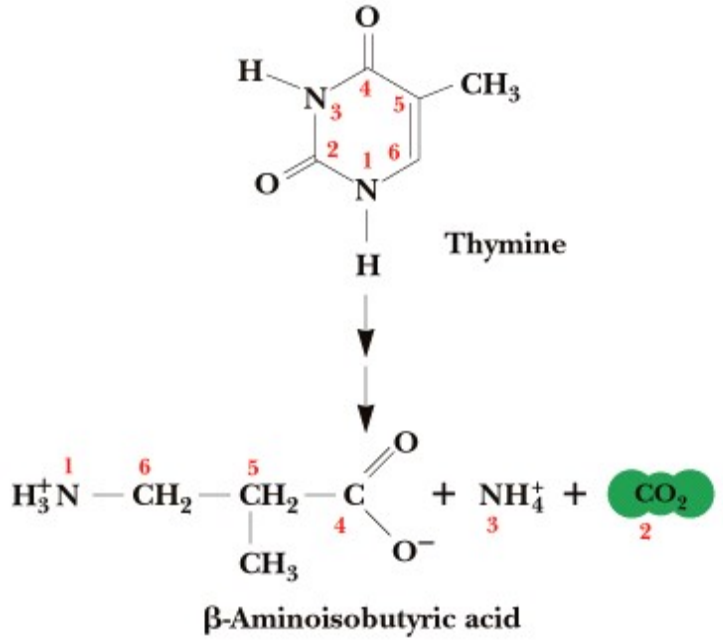
- Catabolism of the pyrimidine nucleotides leads ultimately to **β-alanine** (when CMP and UMP are degraded) or **β-aminoisobutyrate** (when dTMP is degraded) and **NH3** and **CO2**.

- The β-alanine and β-aminoisobutyrate serve as **-NH_2 donors** in transamination of α-ketoglutarate to glutamate.

- A subsequent reaction converts the products to **malonyl-CoA** (which can be diverted to **fatty acid synthesis**) or methylmalonyl-CoA (which is converted to **succinyl-CoA** and can be **shunted to the TCA cycle**).

Pyrimidine catabolism





Catabolism of Purine

- The nucleotides of a cell undergo **continual turnover**.
- Nucleotides are hydrolytically degraded to nucleosides by **nucleotidases**.
- The phosphorolytic cleavage of nucleosides to **free bases** and **ribose 1-phosphate** (*or* **deoxyribose 1-phosphate**) is catalyzed by nucleoside phosphorylases.

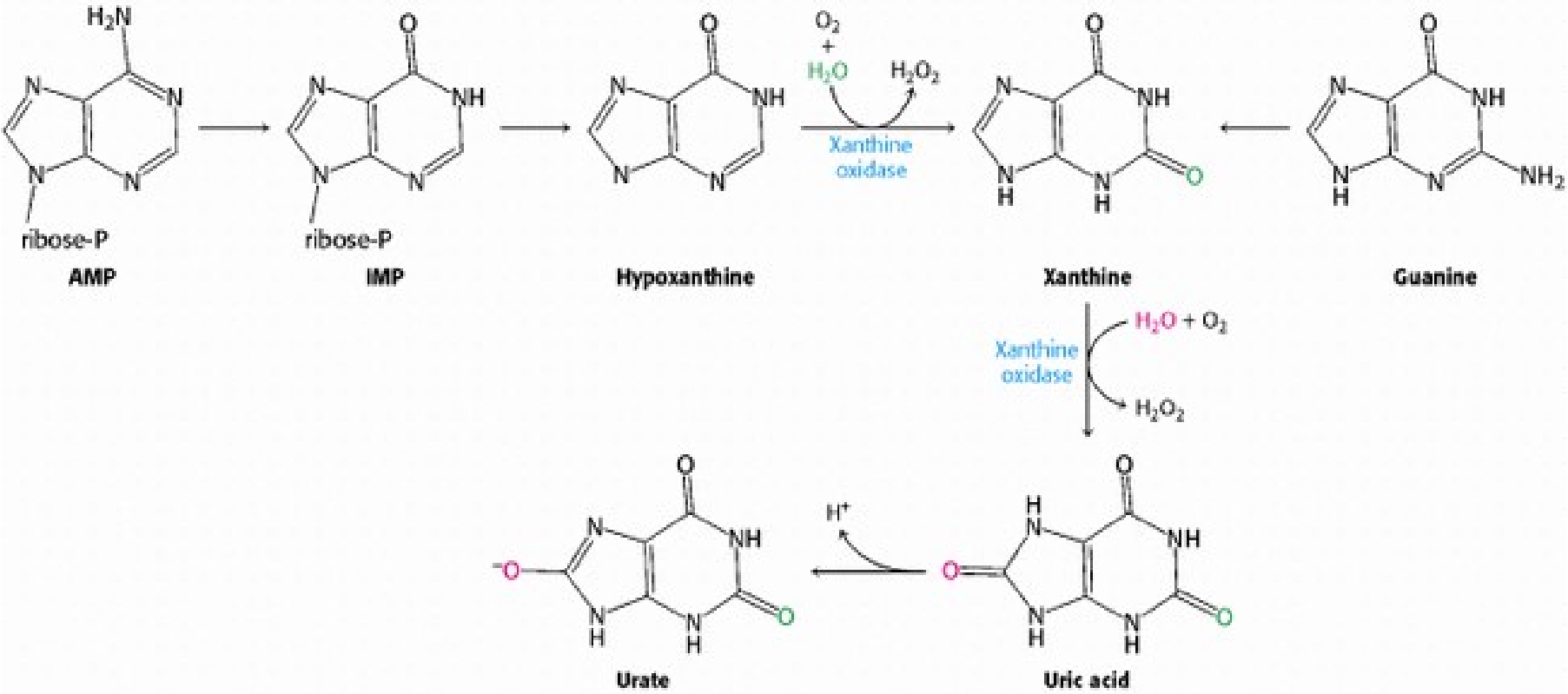
Catabolism of Purine (cont.)

- Some of the bases are reused to form nucleotides by **salvage pathways**. Others are degraded to products that are **excreted** .

- For example, **AMP** is degraded to the free base **hypoxanthine** through hydrolytic cleavage of the glycosidic bond.

- **Xanthine oxidase** transforms **hypoxanthine** to **xanthine** and the same enzyme converts xanthine to **uric acid**. Uric acid loses a proton at physiological pH to **form urate**. In human beings, urate is the final product of purine degradation and is **excreted in the urine**.

Purine catabolism



Purine Catabolism: Purine bases are converted first into xanthine and then into urate for excretion. Xanthine oxidase catalyzes two steps in this process. (Guanine deaminase converts guanine into xanthine)

References:

Title/URL	Author	Publisher	Year
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