Harvard-MIT Division of Health Sciences and Technology
HST.151: Principles of Pharmocology
Instructor: Dr. Carl Rosow, Dr. David Standaert and Prof. Gary Strichartz

Folate

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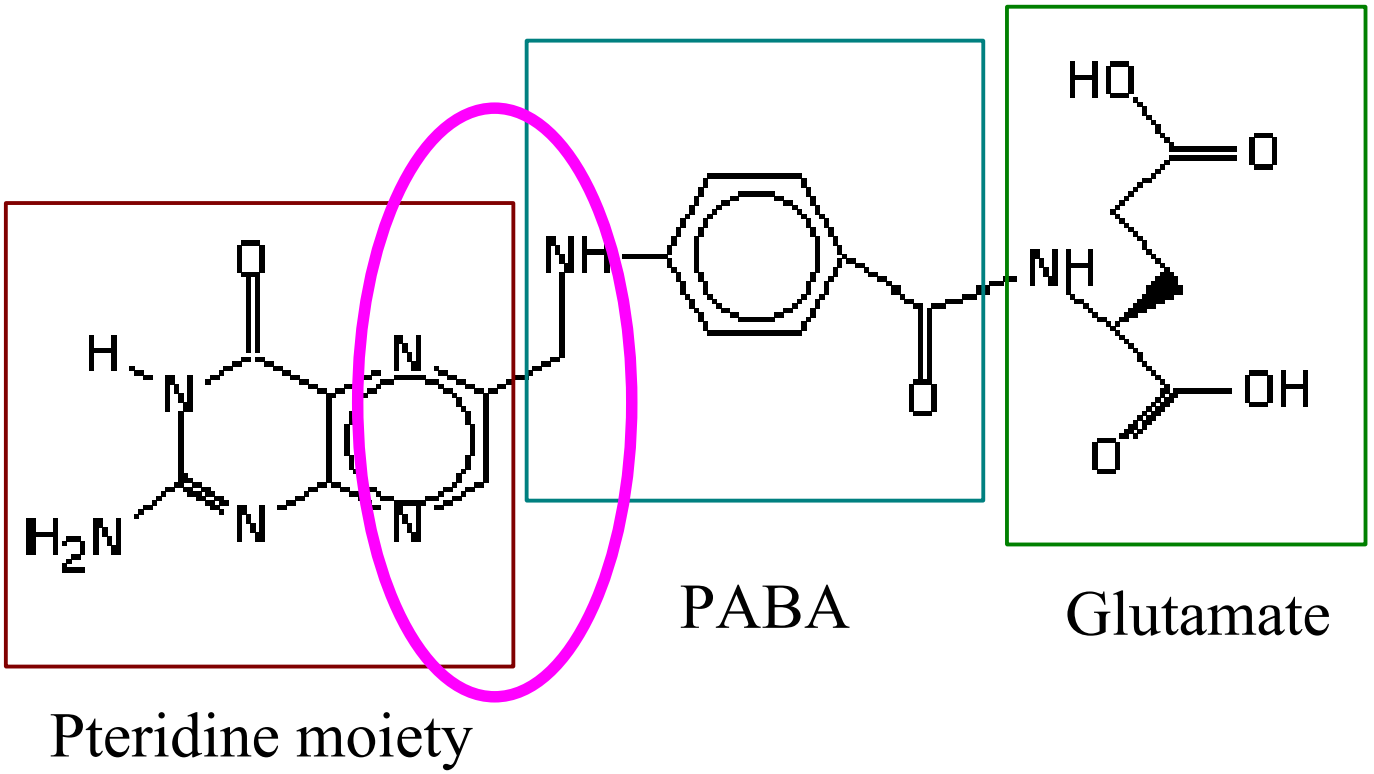
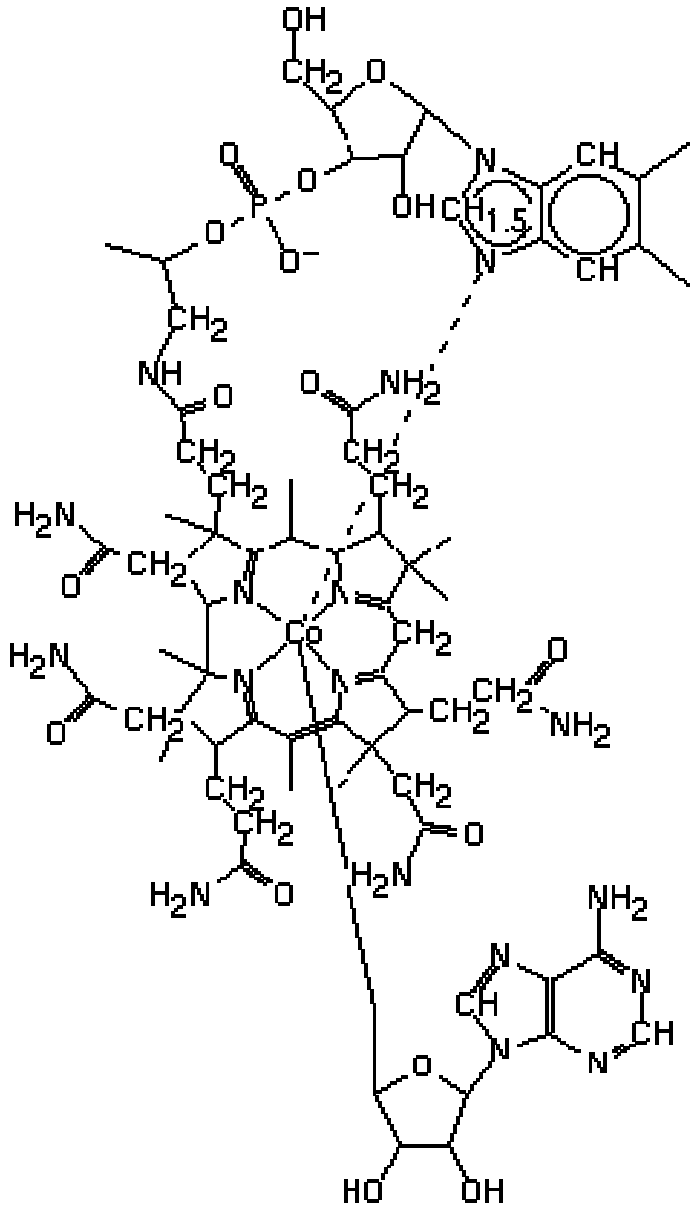
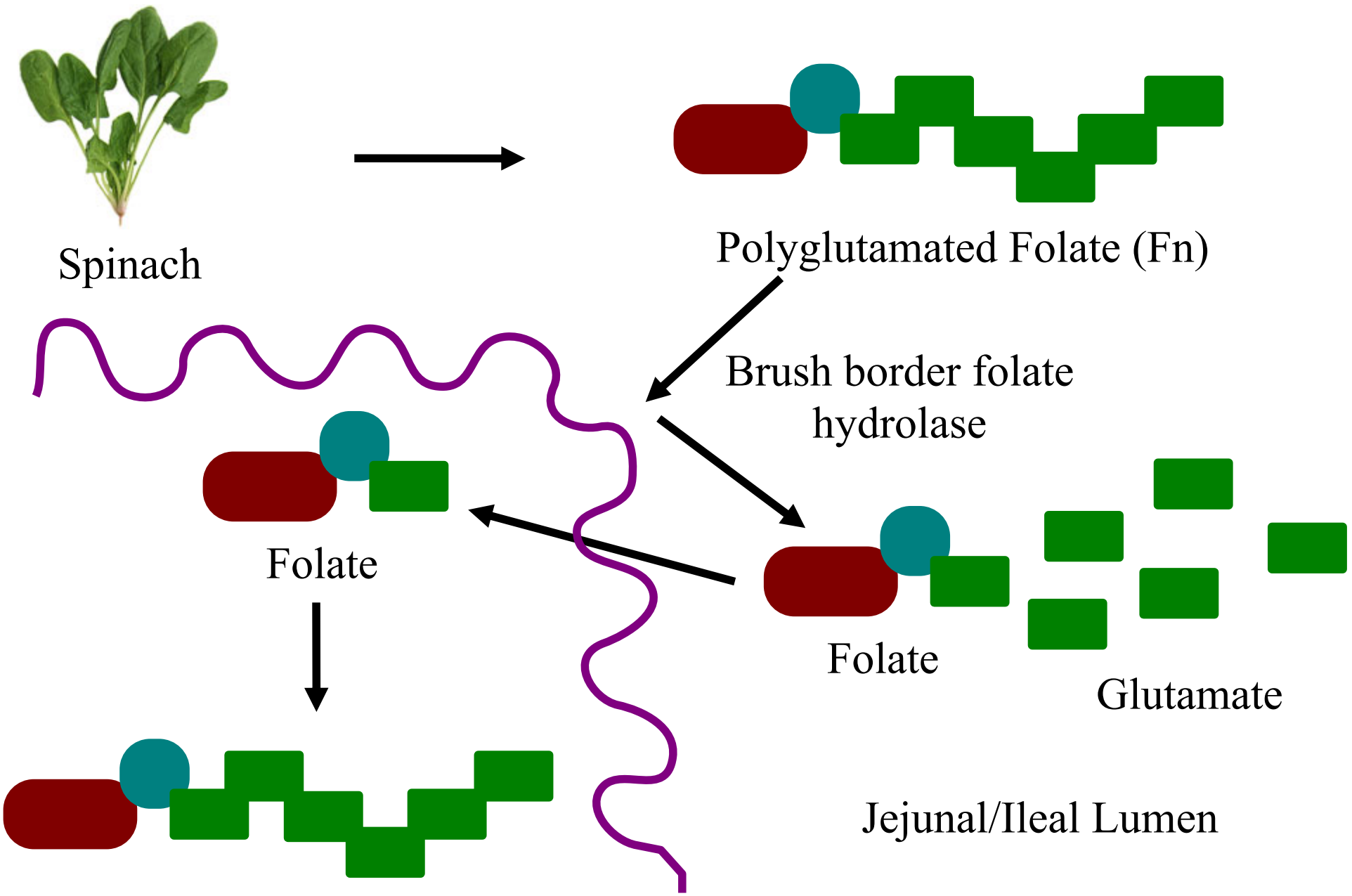
Folate (Vitamin B9)



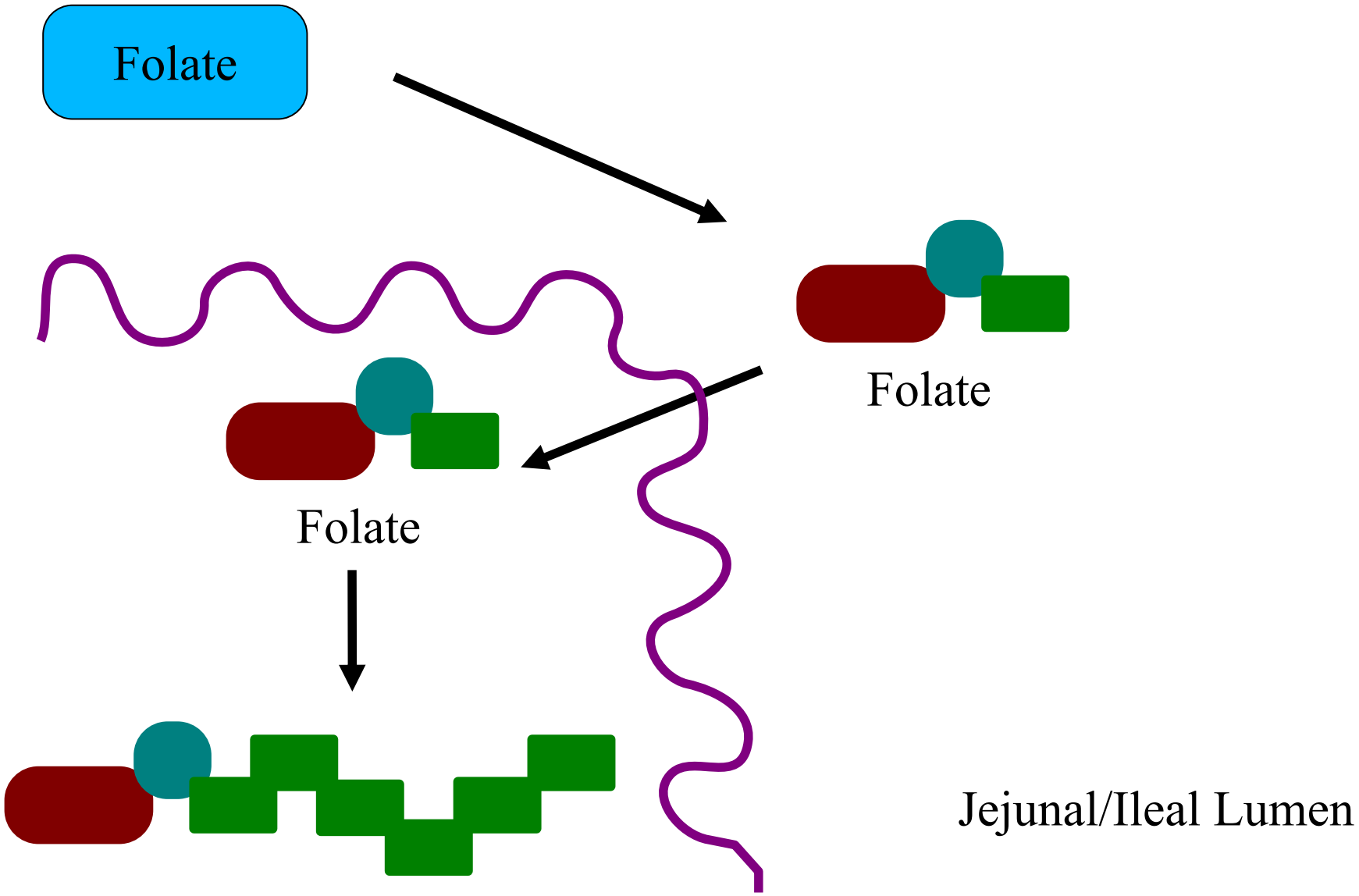
Cobalamin (Vitamin B12)



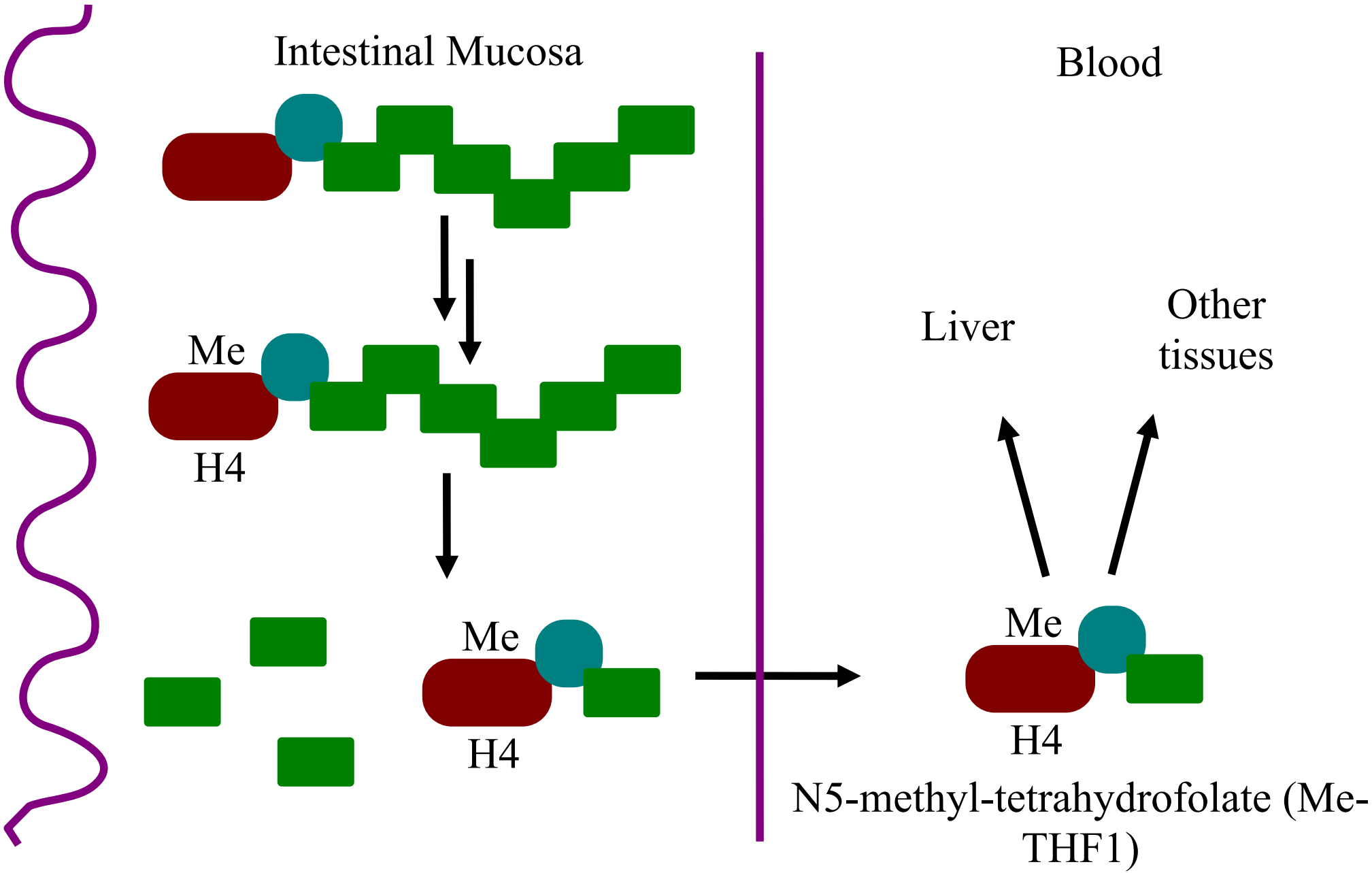
Folate Uptake



Folate Uptake



Folate Uptake (cont.)



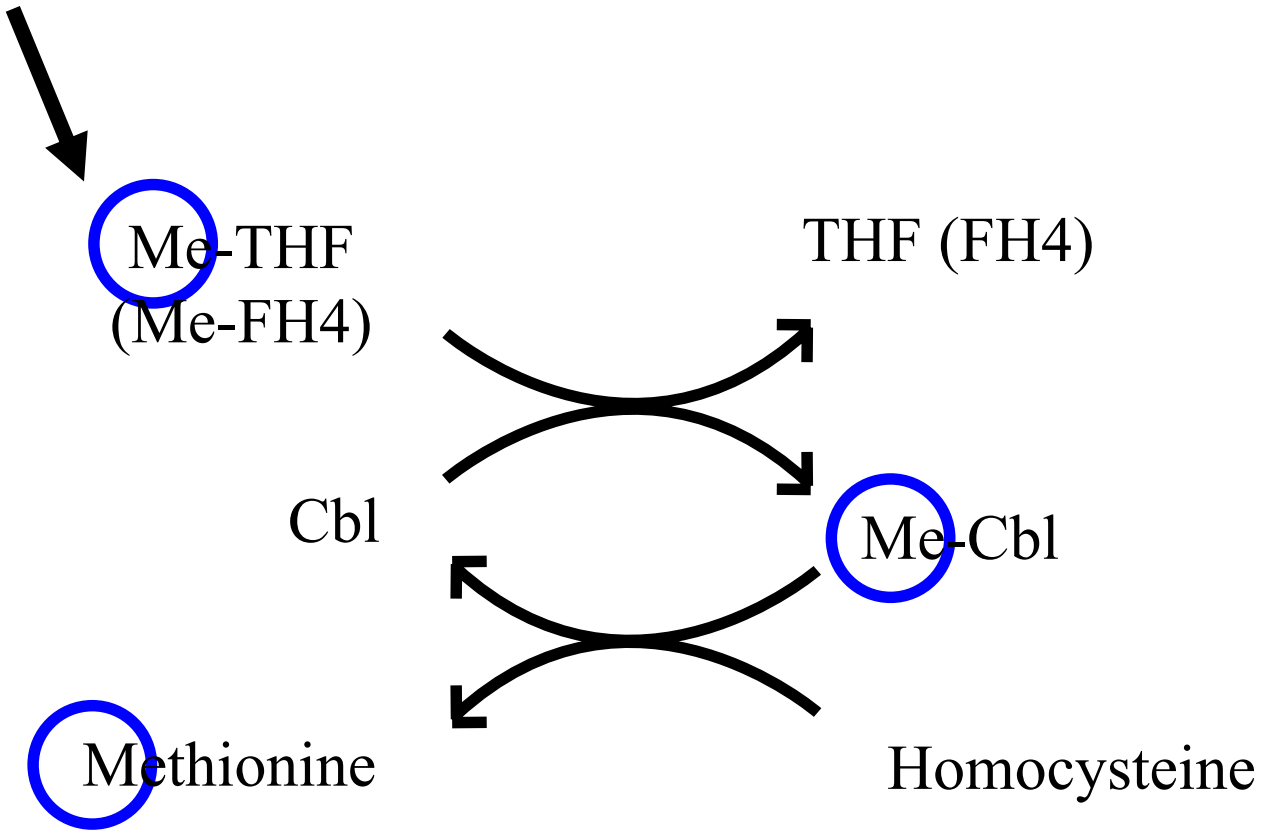
Folate Pharmacokinetics

- Many forms of folate absorbed rapidly in jejunum (and ileum)
  - Specific, saturable transporter
- At high conc., enters blood unchanged
- 2/3 protein-bound
- Rapidly cleared from plasma by cellular (mostly hepatic) uptake
- Peak serum levels 1 to 2 h after oral administration
  - Human milk folate bound to FBP
- Enterohepatic circulation, 90 μg/day

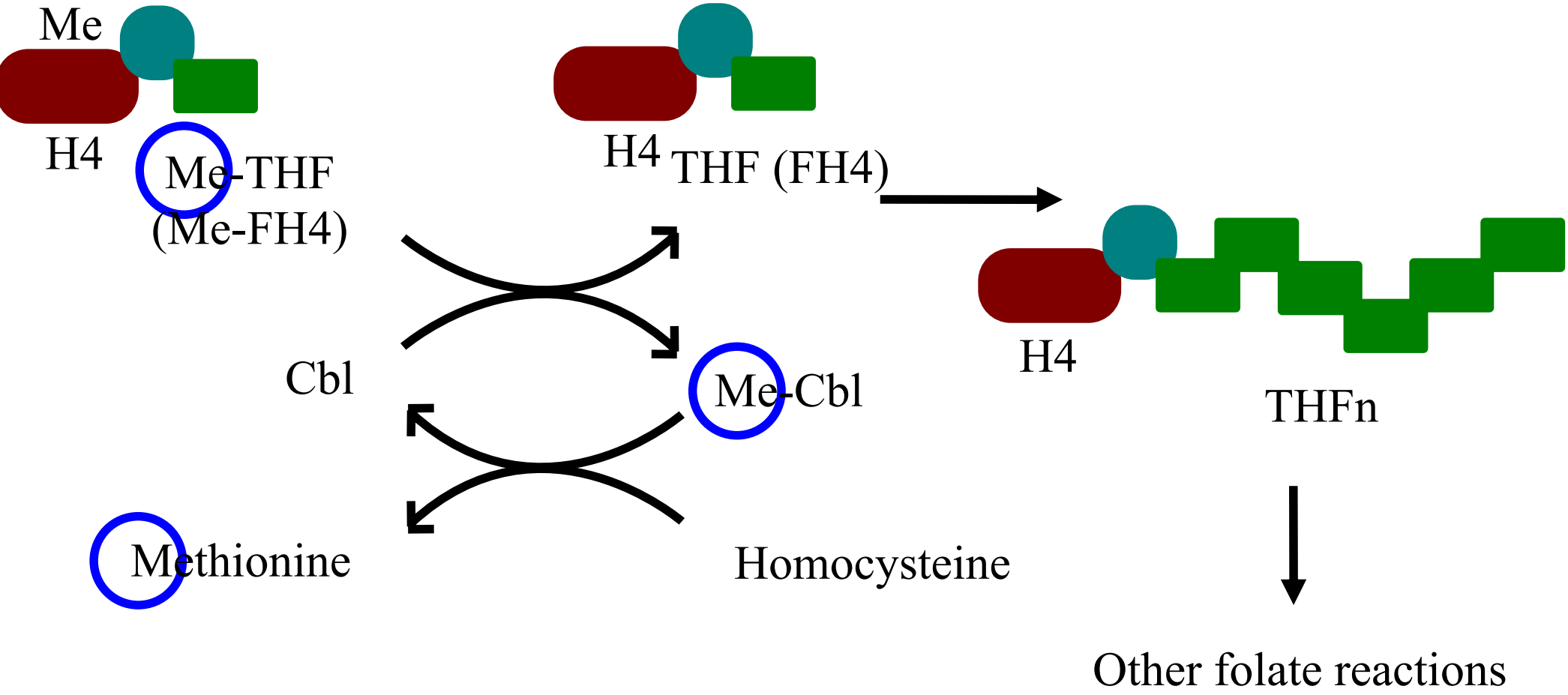
Biochemical Activities of Folate

- One-carbon donor/acceptor
- Nucleotide synthesis
  - Thymine (dUMP to TMP)
  - Purines (AMP, GMP)
- Amino acid metabolism
  - Serine and glycine metabolism
  - Homocysteine to methionone (with B12)
- Histidine, betaine, choline catabolism

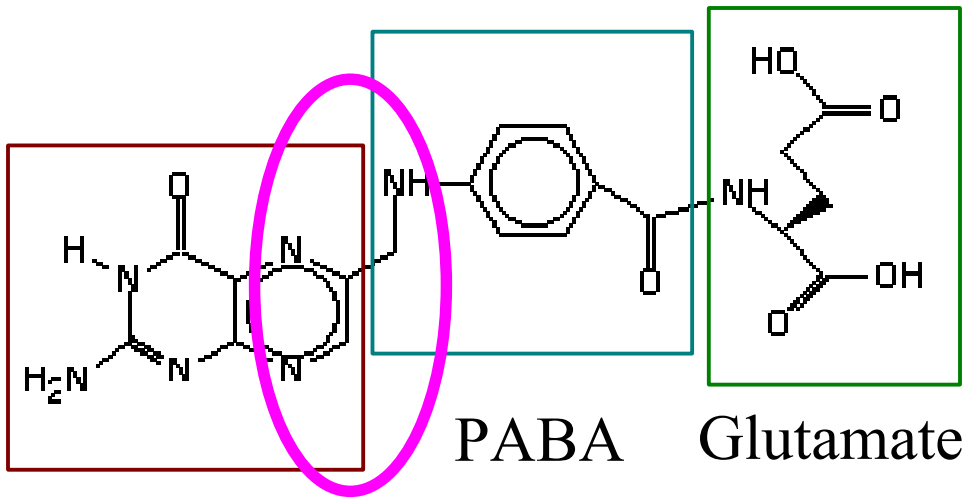
Folate, Cobalamin, and Methionine Synthesis



Folate, Cobalamin, and Methionine Synthesis



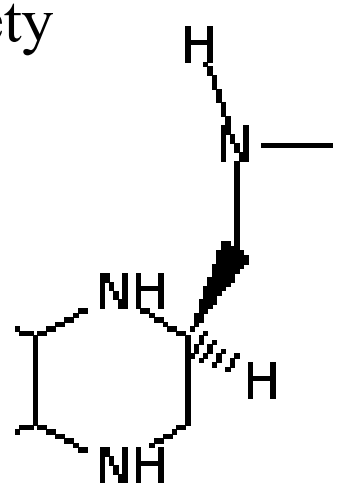
Folate Intermediates in TMP Synthesis



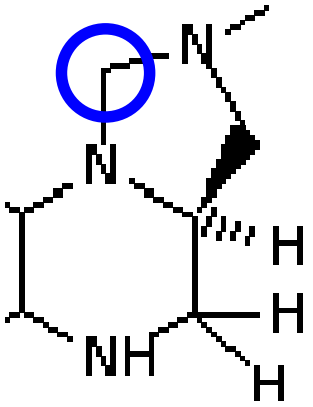
Pteridine moiety

PABA

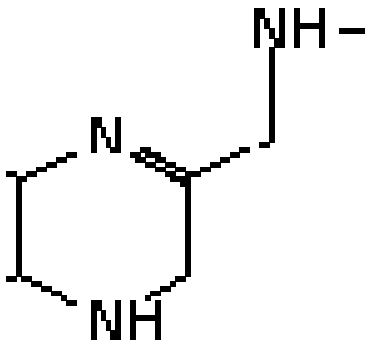
Glutamate(n)



Tetrahydrofolate (THF, FH4)

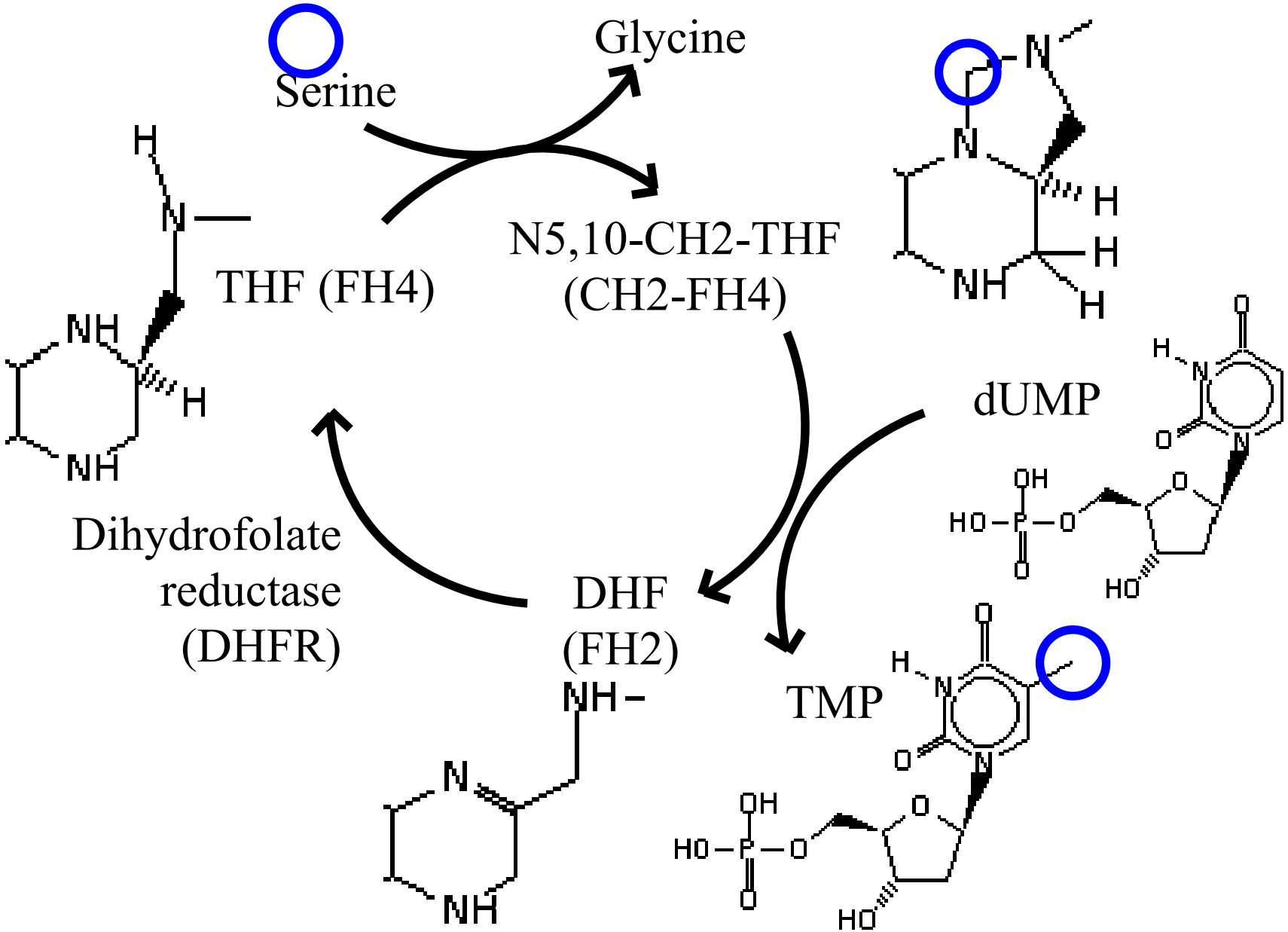


N5,10-methylene-tetrahydrofolate (CH2-FH4)



Dihydrofolate (DHF, FH2)

TMP Synthesis



Historical Understanding of Folate Deficiency

- 1817 – randomized, double-blind trial found folic acid to protect against neural tube defects in pregnant women at risk (those with a previous affected pregnancy)
- Without supplementation, only 1 in 3 women in the US ingests enough to gain the protective effect
- 1998 – FDA mandated that certain cereal products be fortified with folic acid
- Since then, rates of neural tube defects have fallen by 19% overall, and spina bifida by 23% (partly also due to better screening)

Dietary Folate

- Folate occurs naturally in leafy green vegetables, fruits, beans & legumes, and liver
- Folate is heat labile; therefore amount in raw food is reduced by cooking
- The RDA is 400 µg/day for adults unless pregnant (600) or lactating (500)
  - ½ cup cowpeas: 105 µg
  - 1 cup raw spinach: 60 µg
  - 1 oz peanuts: 40 µg

Clinical Case:

A 60 yr old male with a long history of heavy drinking sees you in clinic. Four months ago another physician told him he had an ulcer. He still has abdominal pain, but now he also complains of a painful tongue, sore mouth, and trouble walking. He attributes some of his weakness to all the diarrhea he is having. The patient's wife is impressed that her cousin had the same symptoms when he was receiving chemotherapy for his lymphoma.

→ What is the pathophysiologic process that results in the patient's symptoms?

→ Why would chemotherapy cause similar symptoms to the patient's?

Causes of Folate Deficiency

- Most common cause is nutritional, due to diet and/or alcoholism
- Ethanol reduces stored folate levels in several ways:
  - Increased renal, metabolic clearance
  - Inhibition of jejunal hydrolases
  - Inhibition of hepatic folylpolyglutamate synthase
- Alcohol abuse can rapidly cause symptoms of folate deficiency
  - Serum folate falls within 2 to 4 days
  - Megaloblastosis can occur within 5 to 10 weeks (partly also because of poor diet in alcoholics)

Nutritional deficiency
- Substance abuse
- Alcoholism
- Poor dietary intake
- Overcooked foods
- Depressed patients
- Nursing homes

Malabsorption
- Sprue
- Inflammatory bowel disease
- Infiltrative bowel disease
- Short bowel syndrome

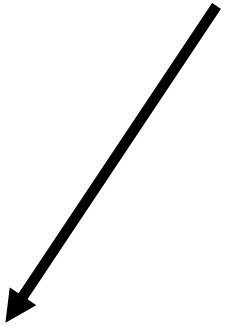
Drugs (various mechanisms)
- Methotrexate
- Trimethoprim
- Ethanol
- Phenytoin

Increased requirements
- Pregnancy, lactation
- Chronic hemolysis
- Exfoliative dermatitis

Clinical Consequences of Folate Deficiency

Look at the macroovalocyte

- Megaloblastic anemia, which is identical to that caused by vitamin B_{12} deficiency
  - Vitamin B_{12} deficiency also causes neurologic manifestations
  - Vitamin B_{12} deficiency develops much more slowly due to higher levels stored in body
- Elevated serum bilirubin and LDH from ineffective erythropoiesis
- Megaloblasts result from inadequate conversion of deoxyuridate to thymidylate, which decreases DNA synthesis
- Other symptoms are atrophic glossitis and diffuse chronic gastritis



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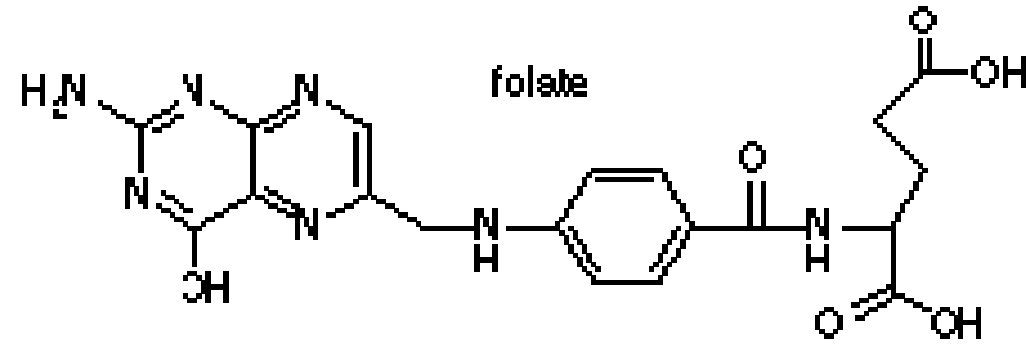
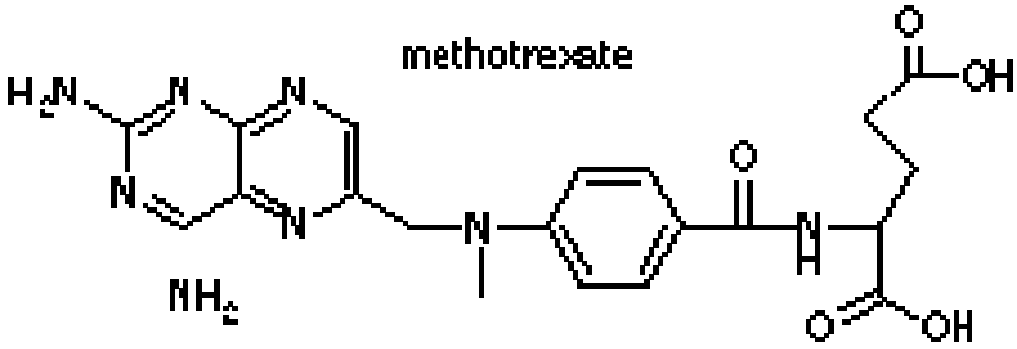
Look at the hypersegmented neutrophil

Pharmacology of Folate: Chemotherapy

- Several folate analogues exist which inhibit DHFR:
  - Methotrexate (MTX)
  - Trimethoprim
  - Pyrimethamine
- Drugs which inhibit DHFR prevent regeneration of THF from DHF; therefore they both:
  - Prevent synthesis of purine nucleotides
  - Prevent methylation of dUMP to dTMP
- What distinguishes these drugs from one another?





Pharmacology of Folate: Chemotherapy

- MTX has little selectivity for various isoforms of DHRF, while trimethoprim selectively inhibits bacterial DHFR and pyrimethamine selectively inhibits parasitic DHFR
- MTX is highly potent, producing reversible DHFR inhibition at subnanomolar concentrations
- MTX causes growth arrest in S phase of mammalian cells
- High-dose MTX chemotherapy is combined with leucovorin rescue
- Uses include carcinomas of breast, lung, head and neck; ALL; and choriocarcinoma

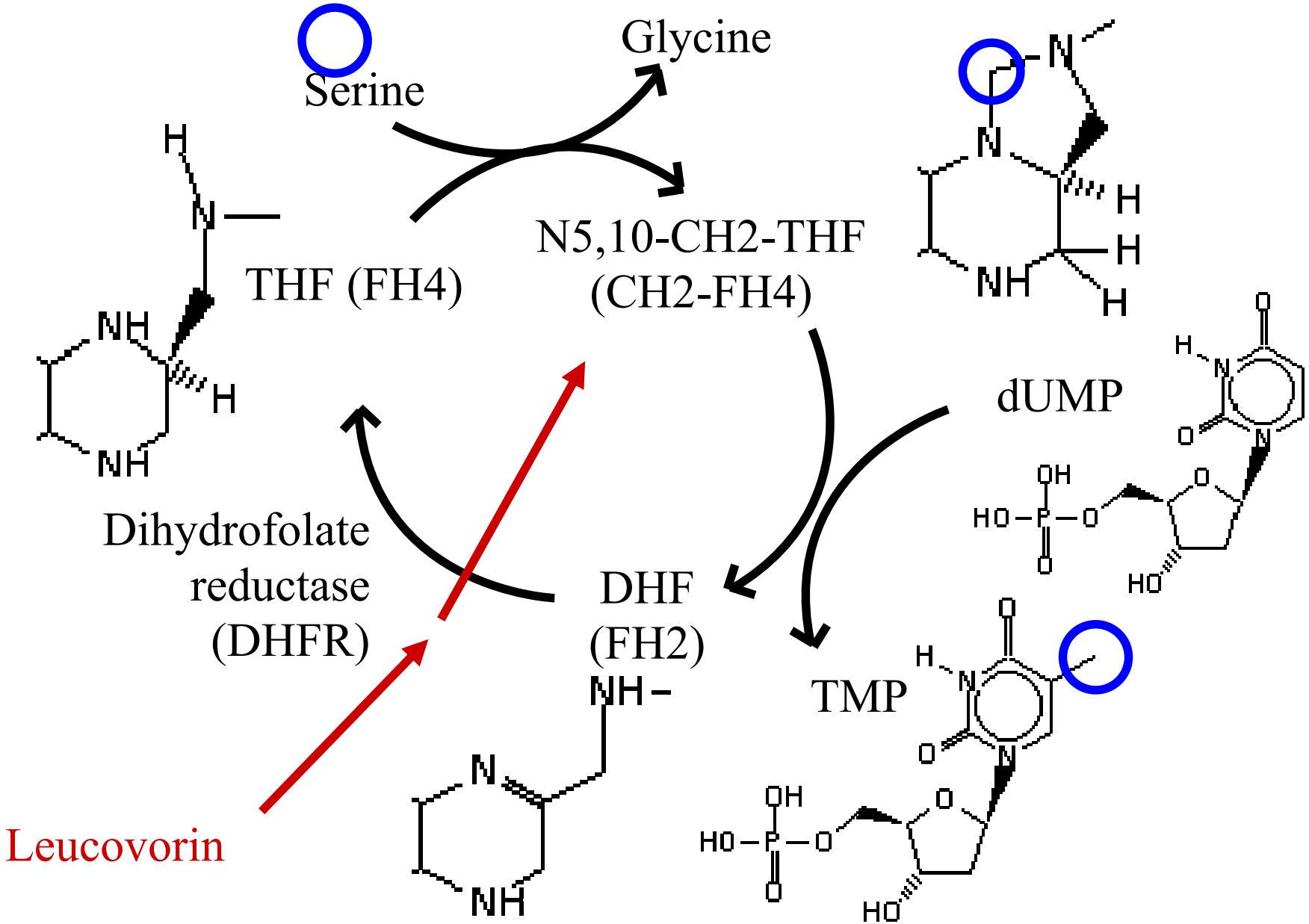
Pharmacology of Folate: Chemotherapy

- Leucovorin (folinic acid) is administered several hours after an MTX dose that would otherwise be lethal to the patient; this selectively kills malignant cells. This is known as "leucovorin rescue".

- Hypothesized mechanisms for rescue:
  - Perhaps normal cells are better able to concentrate folinic acid.
  - Alternately, perhaps malignant cells are more likely to be induced into apoptosis by MTX, thus making them unavailable for rescue.

- Toxicity of MTX includes damage to GI mucosa and to bone marrow

TMP Synthesis



Pharmacology of Folate: Antibacterials

- Bacterial cells must synthesize folic acid de novo from para-aminobenzoic acid (PABA)
- Sulfa drugs are PABA analogues which selectively inhibit dihydropteroate synthase and thereby halt bacterial synthesis of purines & pyrimidines
- These drugs are bacteriostatic, not bactericidal
- Two classes of drugs:
  - Sulfonamides (sulfanilamide, sulfadiazine, sulfamethoxazole)
  - Sulfones (dapsone)

Clinical case revisited

A 60 yr old male with a long history of heavy drinking sees you in clinic. Four months ago another physician told him he had an ulcer. He still has abdominal pain, but now he also complains of a painful tongue, sore mouth, and trouble walking. He attributes some of his weakness to all the diarrhea he is having. The patient's wife is impressed that her cousin had the same symptoms when he was receiving chemotherapy for his lymphoma.