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Vitamins

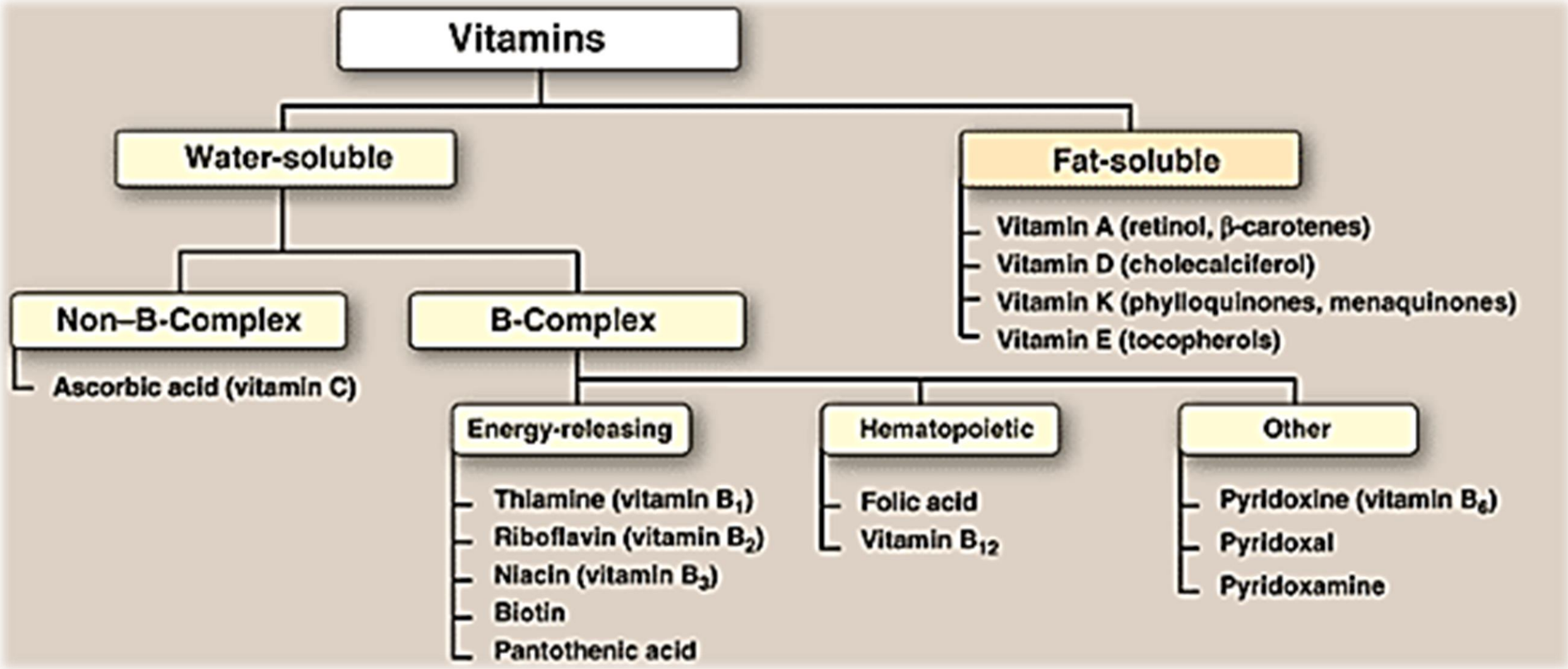
Vitamins

- Organic compounds required by an organism in tiny amounts as a vital nutrient
- Cannot be synthesized in sufficient quantities, & must be obtained from diet
- The term is conditional both on the circumstances & on the particular organism (ascorbic acid, humans, other animals) (vitamin D, human diet)
- Thirteen vitamins are universally recognized at present

Vitamins

- Vitamins have diverse biochemical functions:
  - Hormone-like functions (regulators): regulators of mineral metabolism (e.g., vitamin D), or regulators of cell & tissue growth & differentiation (e.g., vitamin A)
  - Anti-oxidants (e.g., vitamins E & C)
  - Precursors for enzyme cofactors (vitamin B subclasses)

Classification

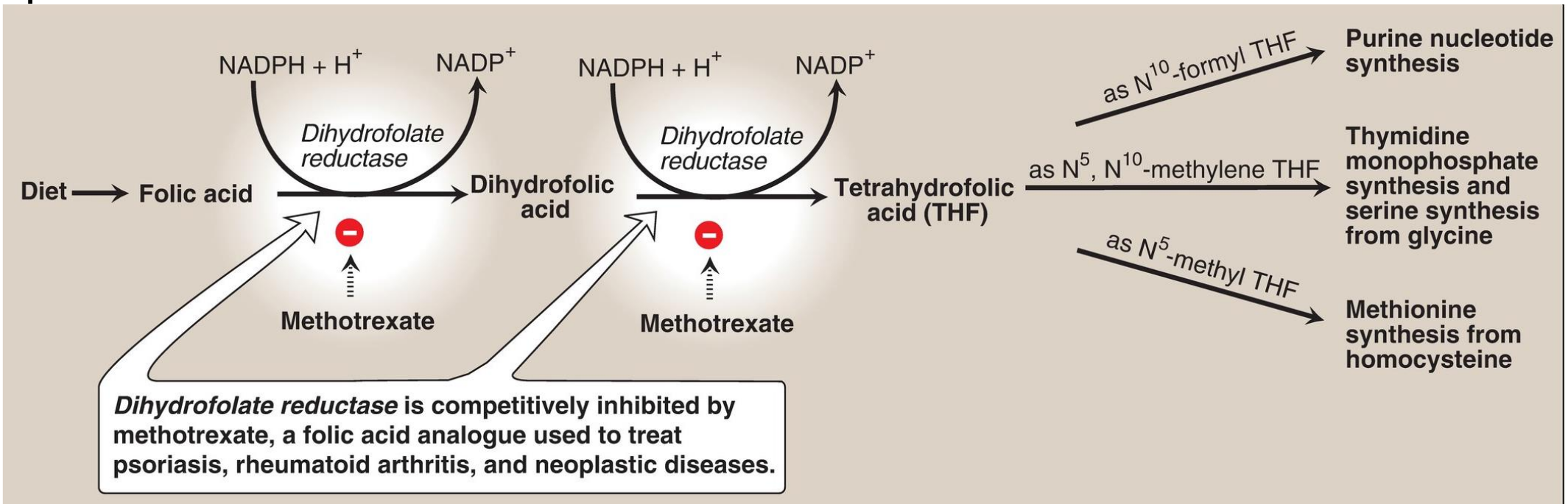


FOLIC ACID (VITAMIN B9) – Folate

- Plays a key role in one-carbon metabolism
- Folic acid deficiency is probably the most common vitamin deficiency in the United States, particularly among pregnant women and individuals with alcoholism
- Leafy, dark green vegetables are a good source of folic acid

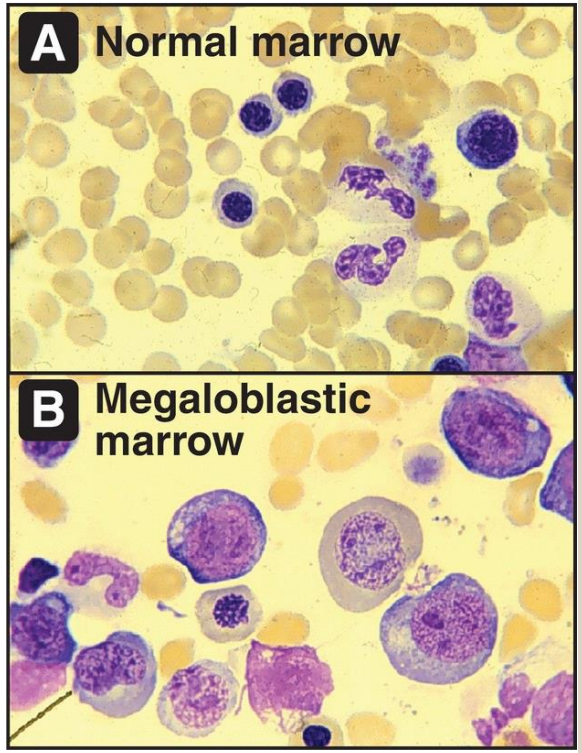
Function

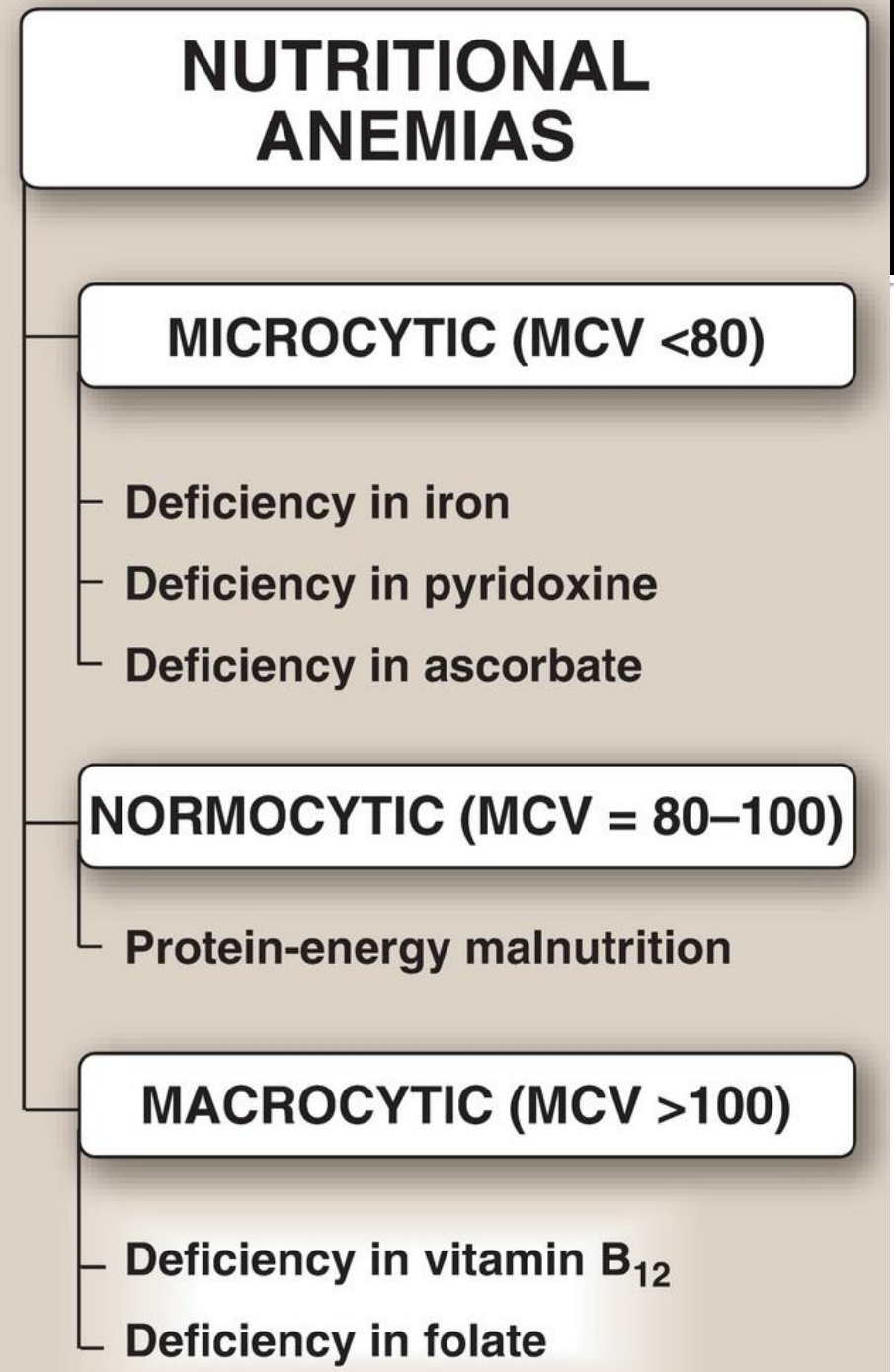
- Tetrahydrofolate (THF), the reduced, coenzyme form of folate
- Receives one-carbon fragments from donors as serine, glycine, and histidine
- Transfers them to intermediates in the synthesis of amino acids, purine nucleotides, and thymidine monophosphate (TMP), a pyrimidine nucleotide incorporated into DNA.



Nutritional anemias

- Classified according to the size of RBCs, or mean corpuscular volume (MCV)
- Microcytic: most common
- Macrocytic
- Commonly called megaloblastic (large, immature RBC precursors, known as megaloblasts, in bone marrow and blood)

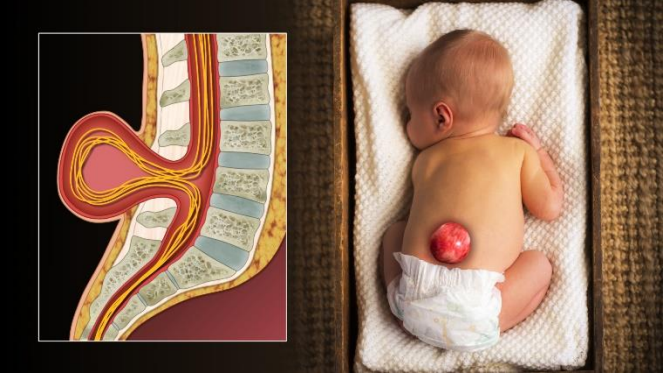




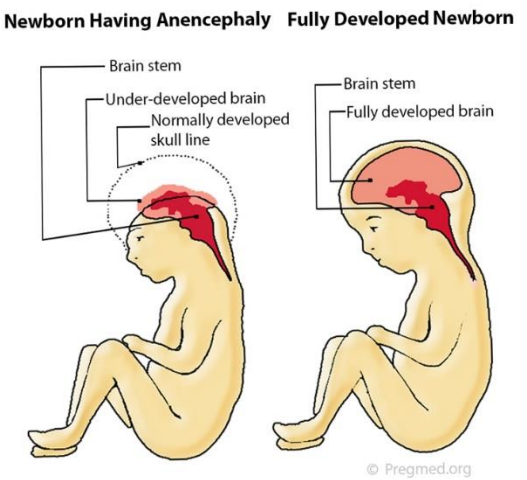
Folate and anemia

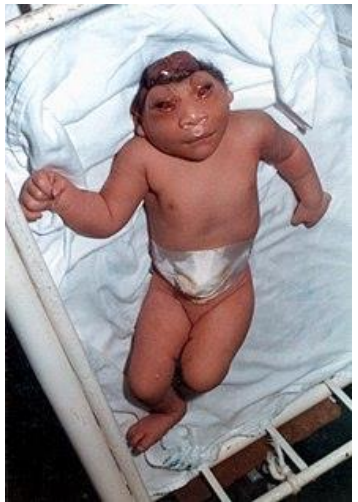
- Inadequate serum levels of folate can be caused by increased demand (pregnancy and lactation), poor absorption caused by pathology of the small intestine, alcoholism, or treatment with drugs (for example, methotrexate) that are *dihydrofolate reductase* inhibitors
- A folate-free diet can cause a deficiency within a few weeks. A primary result of folic acid deficiency is megaloblastic anemia, caused by diminished synthesis of purine nucleotides and TMP, which leads to an inability of cells (including RBC precursors) to make DNA and, therefore, an inability to divide

Folate and neural tube defects



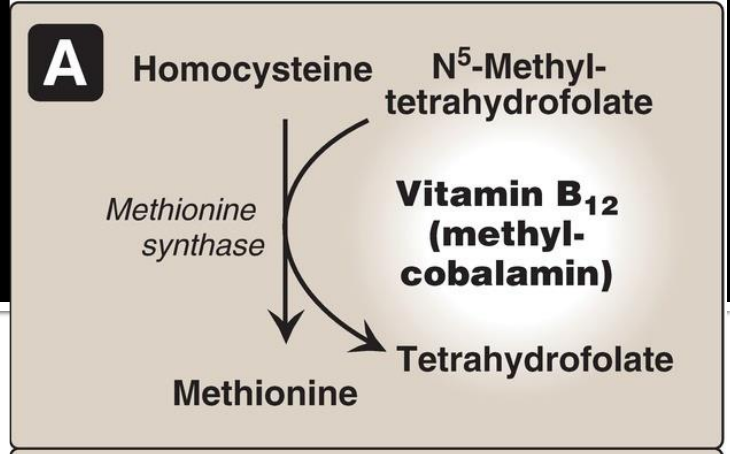
- Spina bifida and anencephaly, the most common neural tube defects (NTD), affect ~3,000 pregnancies in the US annually
- Folic acid supplementation before conception and during the first trimester
- All women of childbearing age (400 µg/day) of folic acid to reduce the risk, 10 times if a previous pregnancy was affected
- In the U.S., addition of folic acid to wheat flour and enriched grain products, resulting in a dietary supplementation of ~0.1 mg/day
- This supplementation allows ~50% of all reproductive-aged women to receive 0.4 mg of folate from all sources

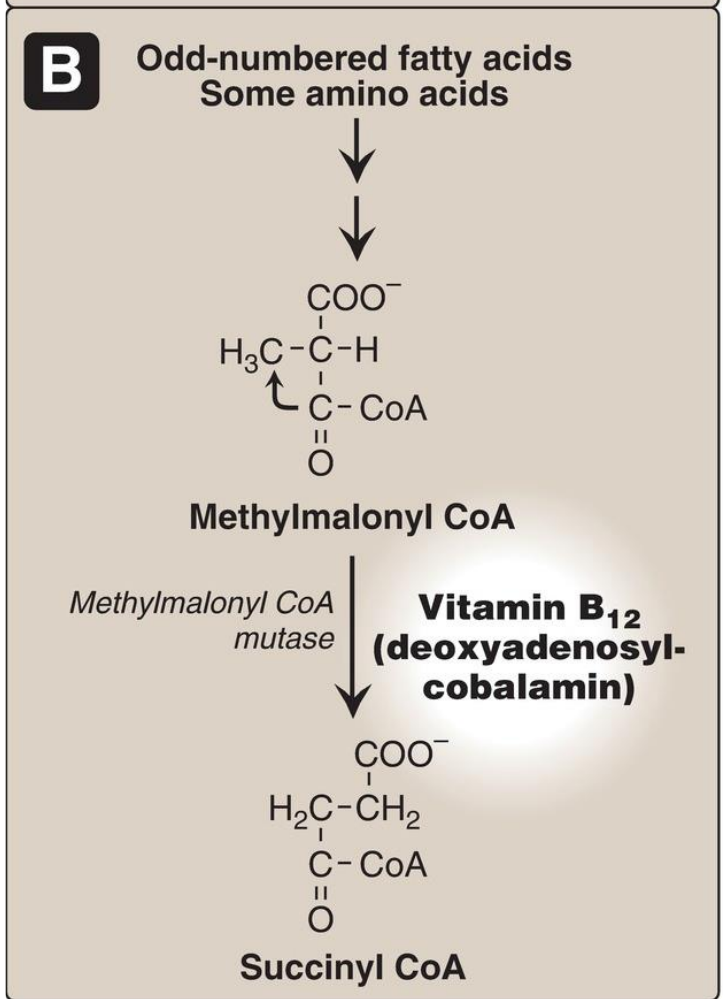




COBALAMIN (VITAMIN B12)

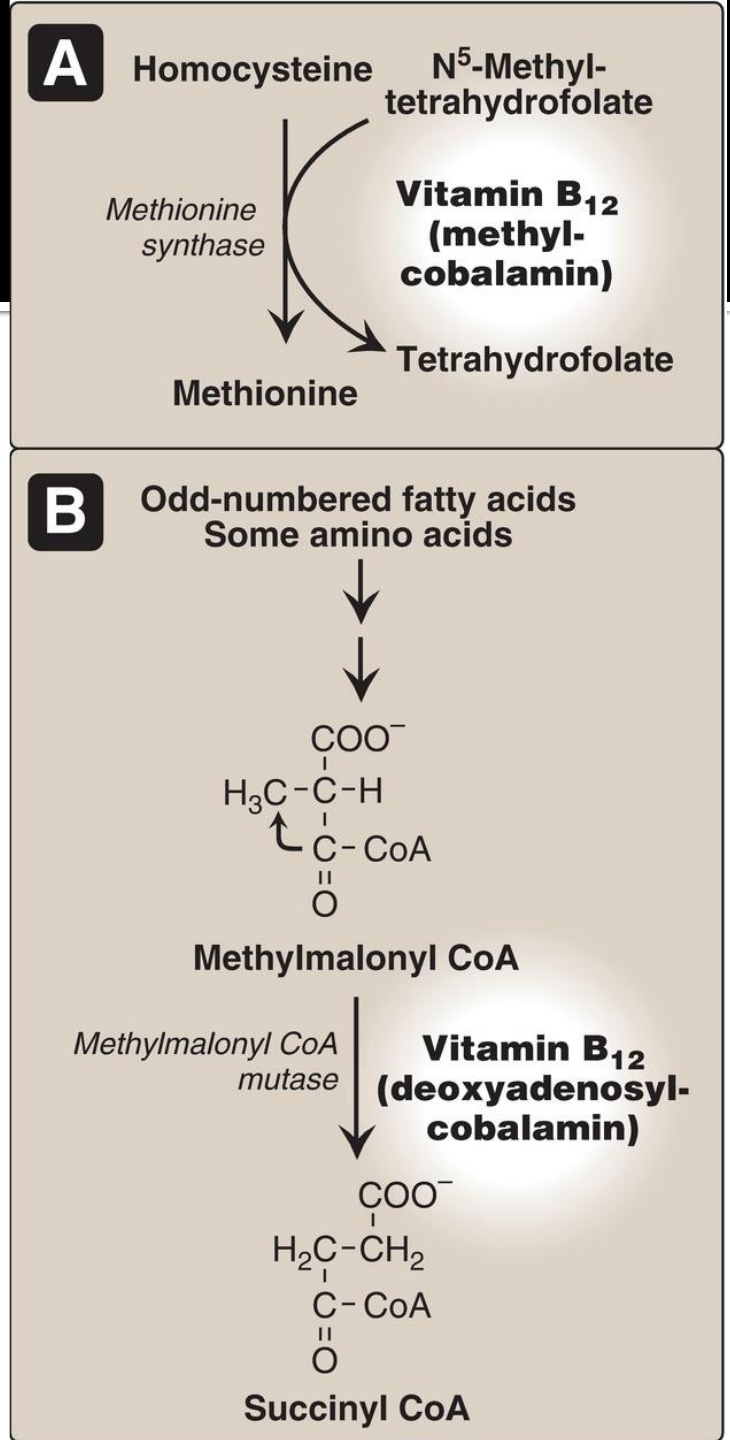
- Required for two essential enzymatic reactions
  - Remethylation of homocysteine (Hcy) to methionine
  - Isomerization of methylmalonyl coenzyme A (CoA), which is produced during the degradation of some amino acids (isoleucine, valine, threonine, and methionine) and fatty acids (FA) with odd numbers of carbon atoms





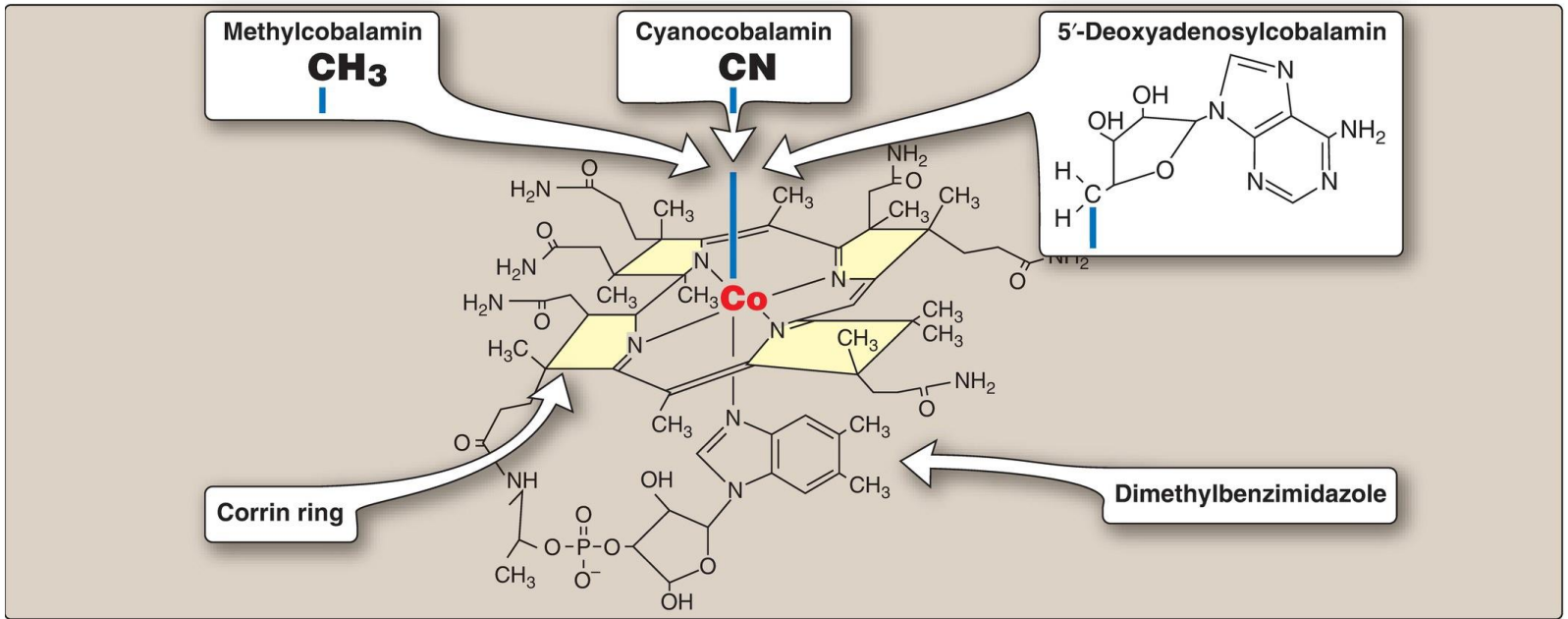
COBALAMIN (VITAMIN B12)

- When cobalamin is deficient, unusual (branched) FA accumulate and become incorporated into cell membranes, including those of the central nervous system (CNS)
- This may account for some of the neurologic manifestations of vitamin B12 deficiency
- Folic acid (as N5-methyl THF) is also required in the remethylation of Hcy. Therefore, deficiency of B12 or folate results in elevated Hcy levels



Structure and coenzyme forms

- A "corrin" ring system vs. Porphyrin (a methene bridge, Cobalt)
- The remaining coordination: nitrogen of 5,6-dimethylbenzimidazole and with cyanide in commercial preparations (cyanocobalamin)
- The physiologic coenzyme are:
  - 5′-deoxyadenosylcobalamin
  - Methylcobalamin

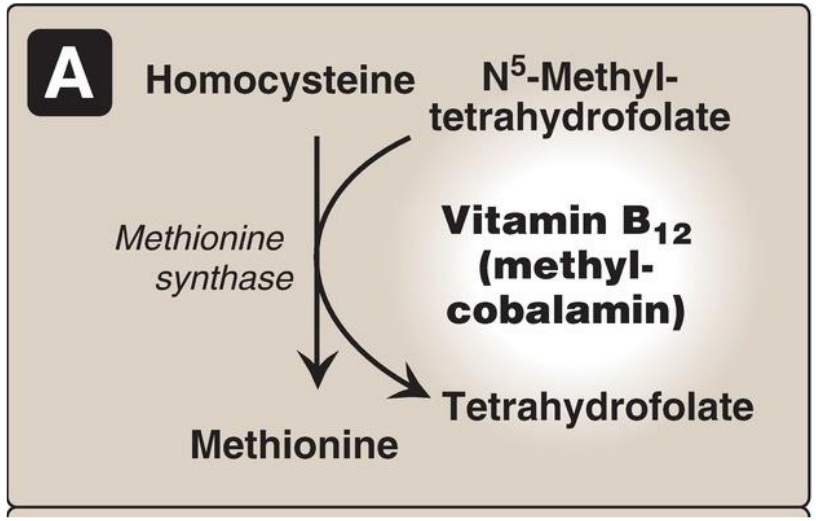


Distribution

- Vitamin B12 is synthesized only by microorganisms, and it is not present in plants
- Animals obtain the vitamin preformed from their intestinal microbiota or by eating foods derived from other animals
- Cobalamin is present in appreciable amounts in liver, red meat, fish, eggs, dairy products, and fortified cereals

Folate trap hypothesis

- Effects of cobalamin deficiency are most pronounced in rapidly dividing cells, such as the erythropoietic tissue of bone marrow and the mucosal cells of the intestine
- Such tissues need both the N5,N10-methylene and N10-formyl forms of THF for the synthesis of nucleotides required for DNA replication
- However, in vitamin B12 deficiency, the utilization of the N5-methyl form of THF is impaired - accumulates
- Deficiency of THF forms needed in purine and TMP synthesis, resulting in megaloblastic anemia

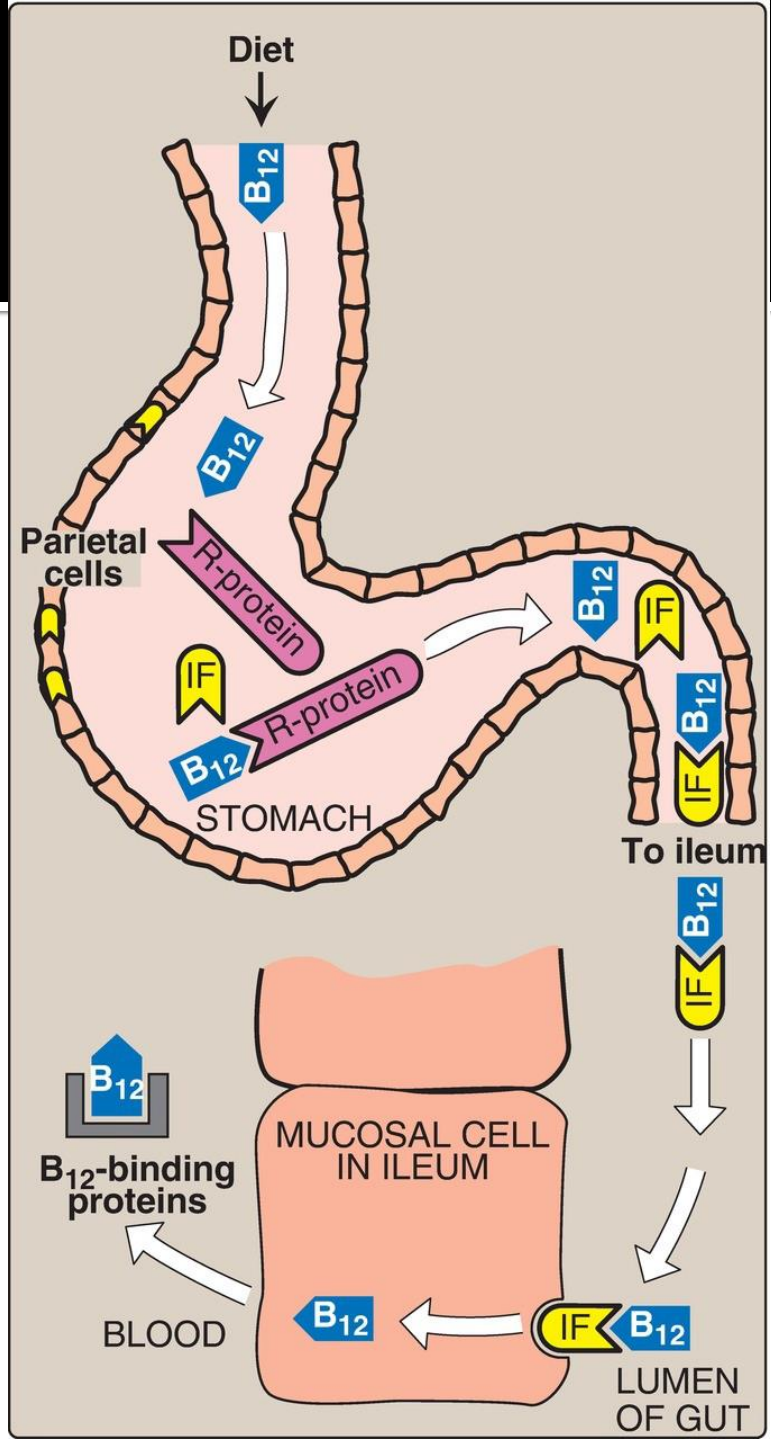


Clinical indications for cobalamin

- In contrast to WS vitamins, significant amounts (2–5 mg) of vitamin B12 are stored in the body
- May take several years for clinical symptoms to develop
- Deficiency happens much more quickly (in months) if absorption is impaired

Pernicious anemia

- Severe malabsorption of vitamin B12 leads to pernicious anemia
- Most commonly a result of an autoimmune destruction of the gastric parietal cells that are responsible for the synthesis of IF
- Impaired absorption
- Acidic environment → complexed to R-protein or haptocorrin (a glycoprotein) → intestine → complexed to intrinsic factor (IF, a glycoprotein) → cubilin (receptor) → circulation (transcobalamin)
- Malabsorption in the elderly (achlorhydria)



Pernicious anemia

- B12 is taken up and stored in the liver, primarily
- It is released into bile and efficiently reabsorbed in the ileum
- Partial or total gastrectomy become IF deficient and, therefore, B12 deficient
- Individuals with cobalamin deficiency are usually anemic (folate recycling is impaired), and they show neuropsychiatric symptoms as the disease develops
- The CNS effects are irreversible

Pernicious anemia

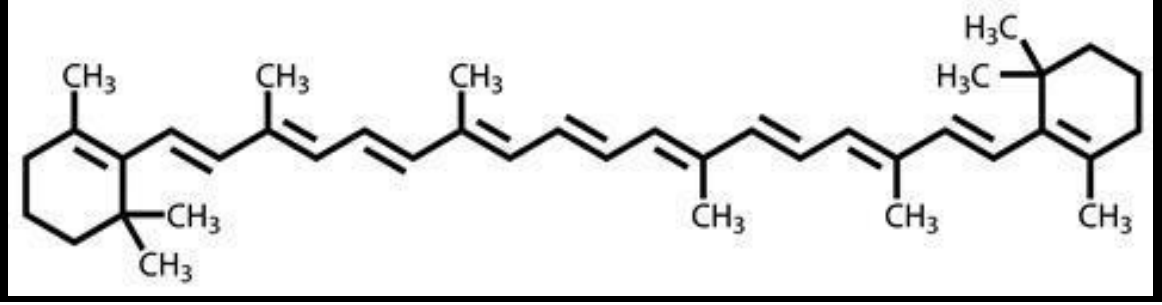
- Requires lifelong treatment with either high-dose oral B12 or intramuscular injection of cyanocobalamin
- Supplementation works even in the absence of IF because ~1% of B12 uptake is by IF-independent diffusion
- Folic acid supplementation can partially reverse the hematologic abnormalities of B12 deficiency and, therefore, can mask a cobalamin deficiency
- To prevent the later CNS effects, therapy for megaloblastic anemia is initiated with both until the cause of the anemia can be determined

Fat soluble vitamins

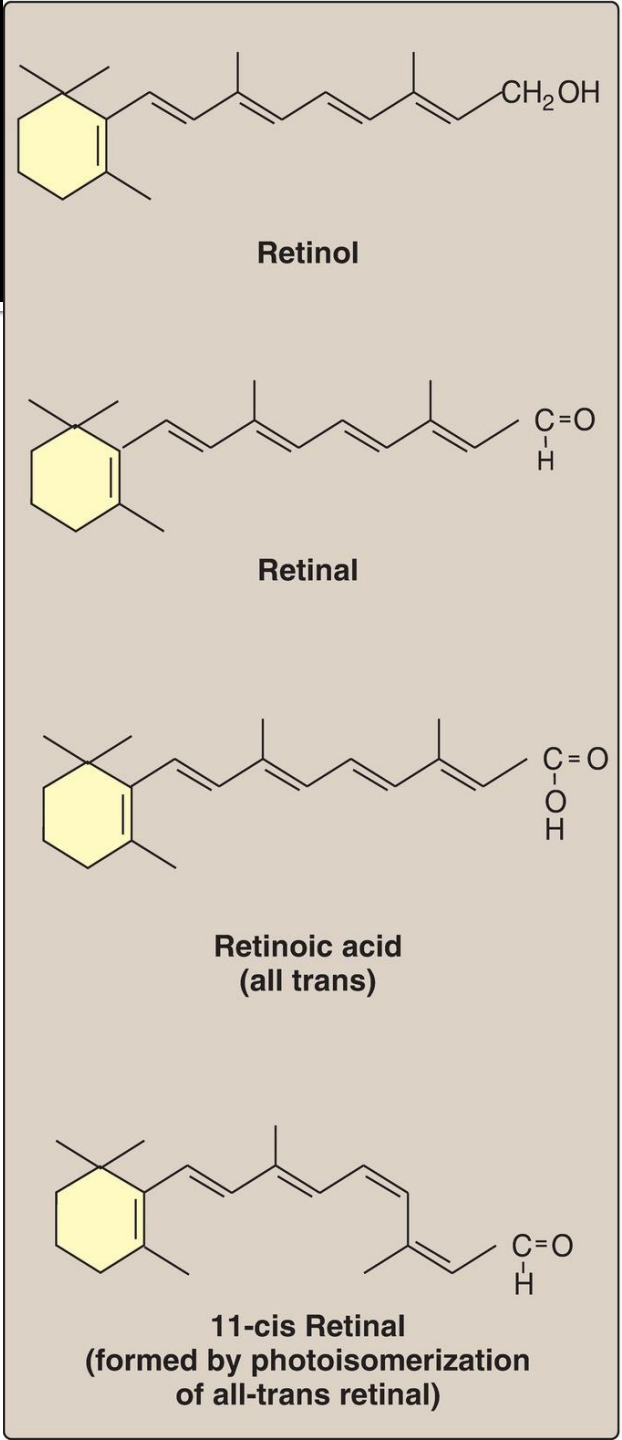
Vitamin	Main function	Deficiency
A	Roles in vision, growth, reproduction	Night blindness, cornea damage
D	Regulation of Ca^{+2} & phosphate metabolism	Rickets (children), Osteomalacia (adults)
E	Antioxidant	RBCs fragility
K	Blood coagulation	Subdermal hemorrhaging

All fat-soluble vitamins are groups and carried in chylomicrons

Vitamin A

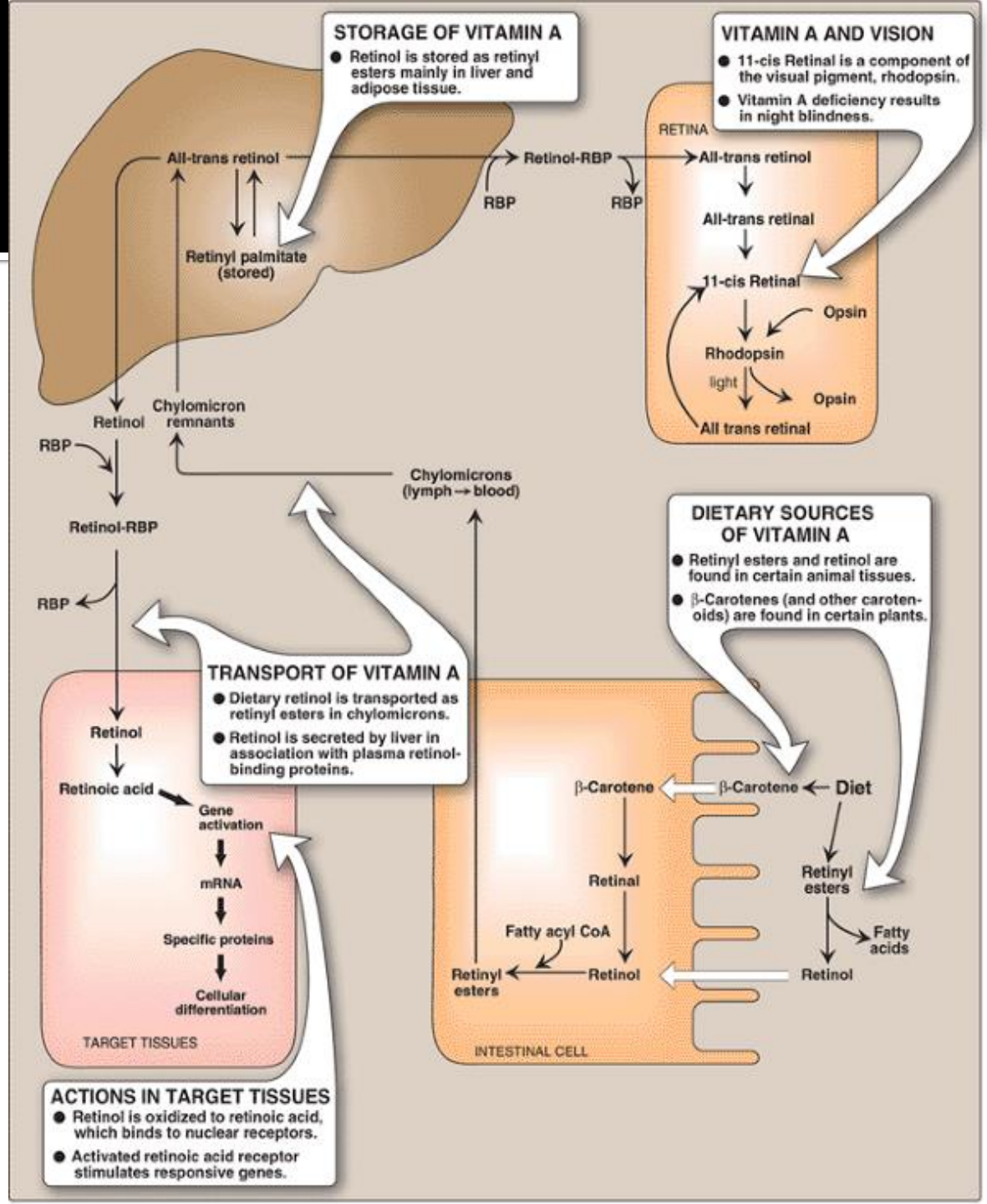


- The retinoids, a family of molecules related to retinol (vitamin A), are essential for vision, growth, reproduction, & maintenance of epithelial tissues
- Retinal and retinol are inter-convertible
- β-Carotene: oxidatively cleaved in the intestines to yield 2 molecules of retinal
- Retinoic acid, mediates most of the actions of the retinoids, except for vision and spermatogenesis



Absorption & transport

- ✓ Retinyl esters, intestinal mucosa, retinol
- ✓ Carotenes, retinal, retinol
- ✓ Chylomicrons, lymphatic system, liver (storage)
- ✓ Release from the liver: retinol binds the plasma retinol-binding protein (RBP) complexed with transthyretin
- ✓ Cellular RBP → nuclear receptors (steroids), RNA, proteins (keratin expression)

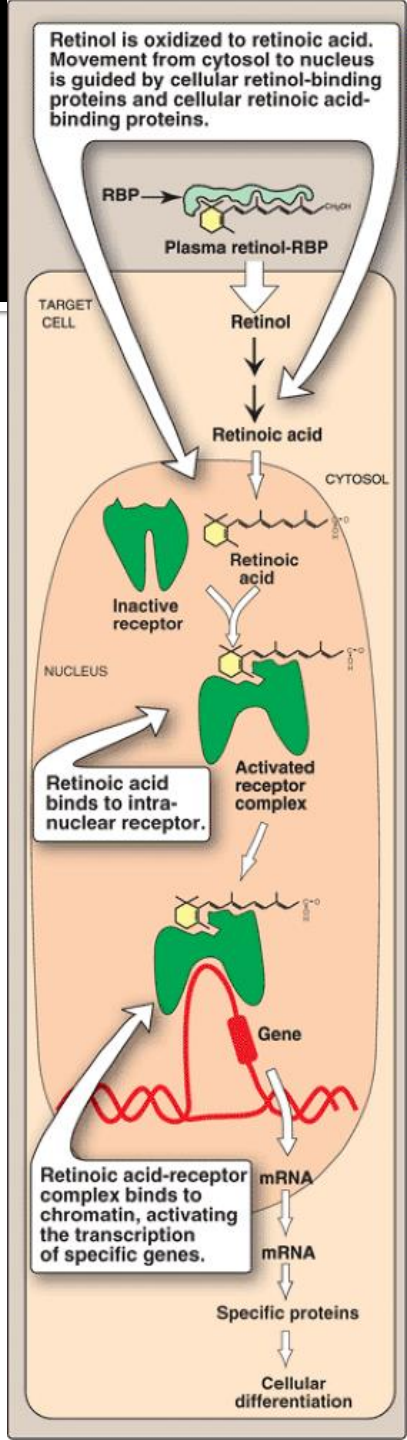


Functions of vitamin A

- ✓ Visual cycle: 11-cis retinal specifically binds the protein opsin (rhodopsin)
- ✓ Reproduction: Retinol and retinal (not retinoic acid) are essential for spermatogenesis in the male and preventing fetal resorption in the female
- ✓ Growth (retinoic acid): Vitamin A deficiency results in a decreased (growth rate & bone development) in children
- ✓ Maintenance of epithelial cells (retinoic acid): Vitamin A is essential for normal differentiation of epithelial tissues & mucus secretion
- ✓ Animals given vitamin A only as retinoic acid from birth are blind and sterile

Retinoic acid mechanism of action

- Binds with high affinity [RAR] - nucleus of target tissues such as epithelial cells
- Activated complex binds to response elements on DNA and recruits activators or repressors to regulate retinoid-specific RNA synthesis
- Retinoids control the expression of the gene for keratin in most epithelial tissues of the body
- RAR proteins are part of the superfamily of transcriptional regulators that includes the nuclear receptors for steroid and thyroid hormones and vitamin D (similar way of function)



Visual cycle

- When rhodopsin, a G protein–coupled receptor, is exposed to light, a series of photochemical isomerizations occurs, which results in the bleaching of rhodopsin and release of all-trans retinal and opsin
- This process activates the G protein transducin, triggering a nerve impulse that is transmitted by the optic nerve
- All-trans retinal is reduced to all-trans retinol, esterified, and isomerized to 11-cis retinol that is oxidized to 11-cis retinal
- 11-cis retinal combines with opsin to form rhodopsin, thus completing the cycle

Epithelial cell maintenance & Reproduction

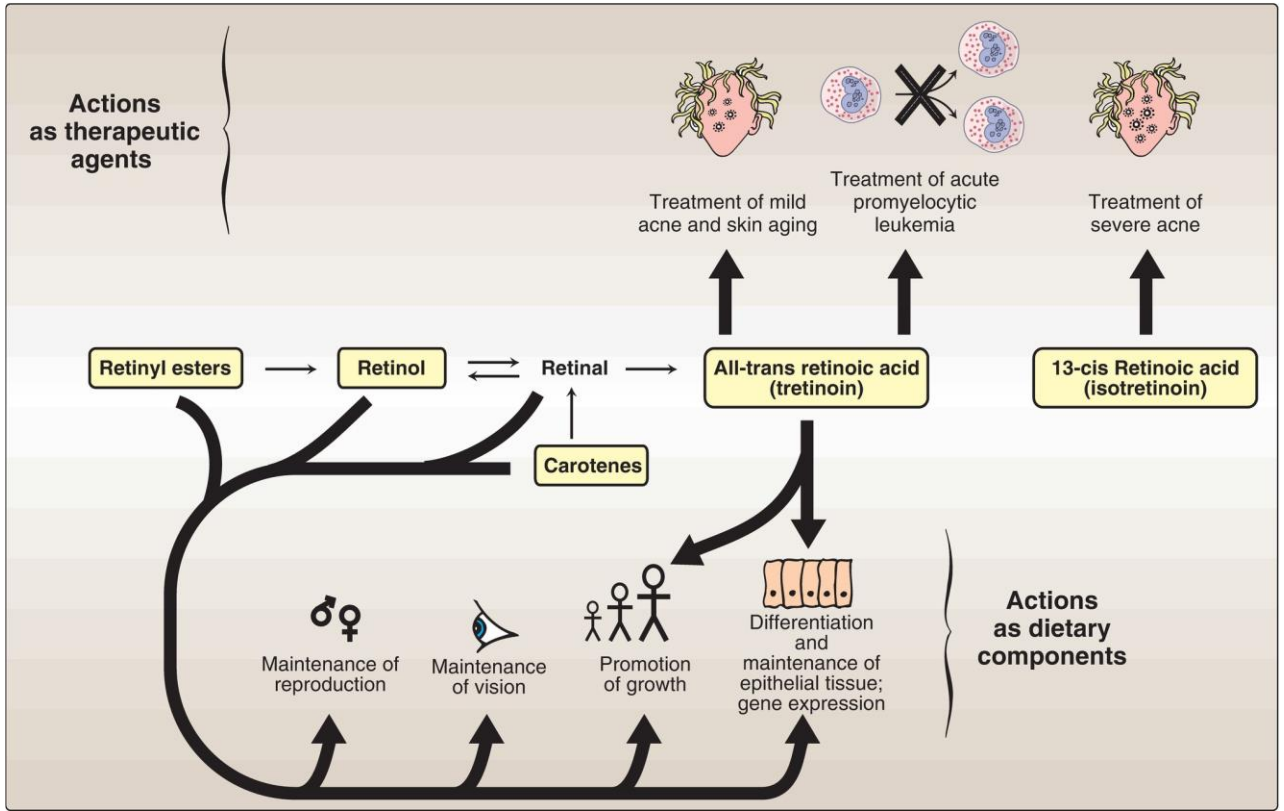
- Vitamin A is essential for normal differentiation of epithelial tissues and mucus secretion and, thus, supports the body's barrier-based defense against pathogens
- Retinol and retinal are essential for normal reproduction, supporting spermatogenesis in the male and preventing fetal resorption in the female
- Retinoic acid is inactive in maintaining reproduction and in the visual cycle but promotes growth and differentiation of epithelial cells

Distribution & Requirements

- Liver, kidney, cream, butter, and egg yolk are good sources of preformed vitamin A
- Yellow, orange, and dark-green vegetables and fruits are good sources of the carotenes (provitamin A)
- RDA for adults is 900 retinol activity equivalents (RAE) for males and 700 RAE for females. In comparison, 1 RAE = 1 µg of retinol, 12 µg of β-carotene, or 24 µg of other carotenoids.

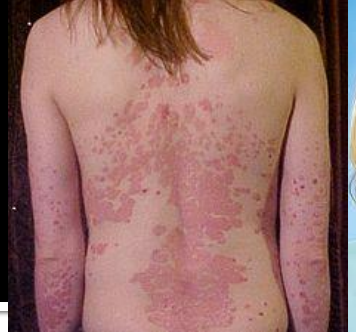
Clinical indications for vitamin A

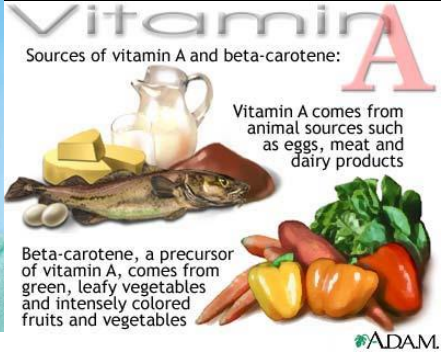
- Retinoic acid and retinol have distinctly different therapeutic applications
- Retinol and its carotenoid precursor are used as dietary supplements
- Various forms of retinoic acid are useful in dermatology



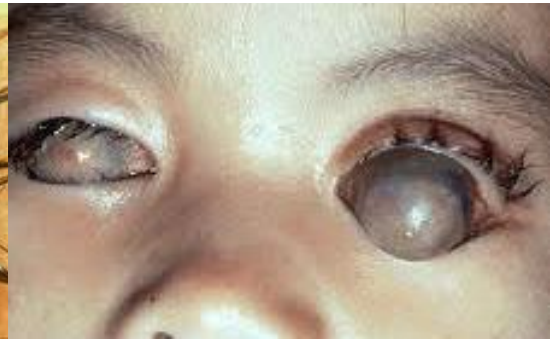
- Tretinoin is too toxic for systemic (oral) administration in treating skin conditions and is confined to topical application

Sources & indications





- ✓ Sources: excess cause hypervitaminosis A
- ✓ Clinical indications:
  - ✓ Dietary deficiency: mild (night blindness, nyctalopia), prolonged (irreversible loss for some visual cells), severe (xerophthalmia)
    - ✓ Xerophthalmia: ulceration & dryness of conjunctiva & cornea, followed by scar & blindness (affecting over 500,000 children worldwide every year)
  - ✓ Acne and psoriasis: effectively treated with retinoic acid



Toxicity - Hypervitaminosis A

- Amounts exceeding 7.5 mg/day of retinol should be avoided
- Hypervitaminosis A
  - Skin: dry and pruritic (because of decreased keratin synthesis)
  - Liver: enlarged and can become cirrhotic
  - CNS: a rise in intracranial pressure (symptoms of a brain tumor)
- Pregnant women: potential for teratogenesis
- UL is 3,000 µg of preformed vitamin A/day

Toxicity - Isotretinoin

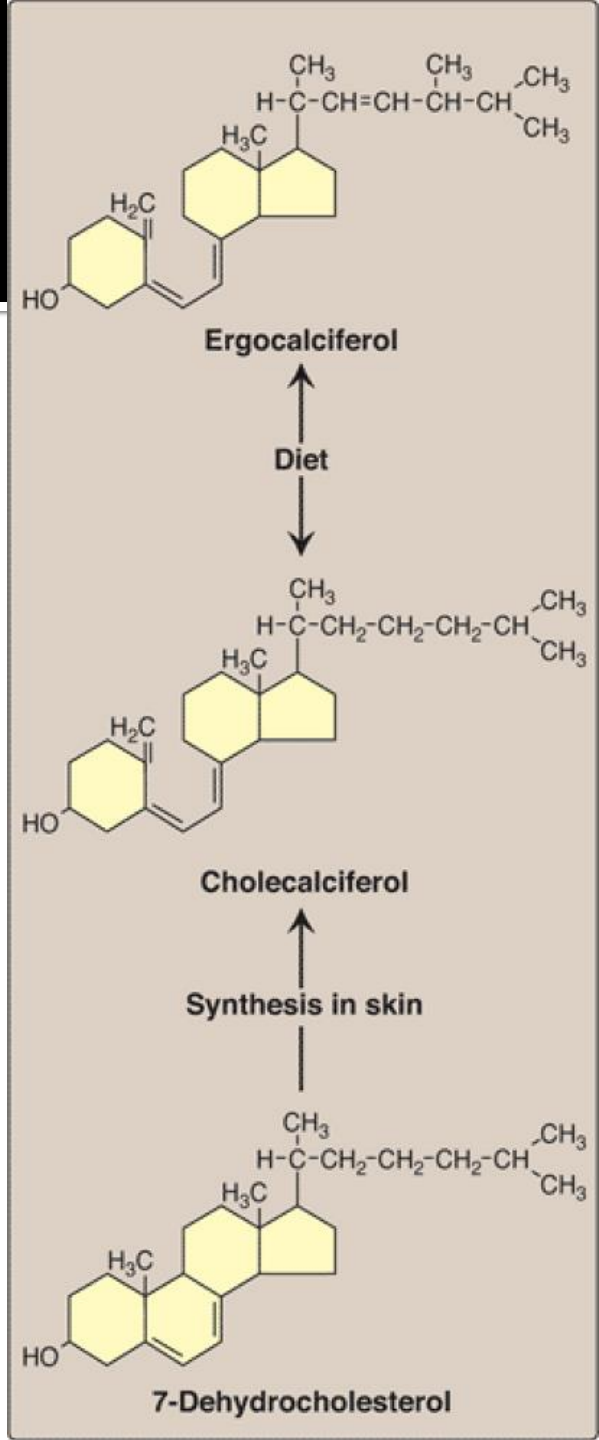
- Isomer of retinoic acid - teratogenic (pregnancy)
- Severe, disfiguring cystic acne that is unresponsive to standard therapies
- Prolonged treatment with isotretinoin can result in an increase in TAG and cholesterol, providing some concern for an increased risk of CVD

VITAMIN D

- A group of sterols that have a hormone-like function
- Active molecule, 1,25-dihydroxycholecalciferol ([1,25-diOH-D3], or calcitriol), binds to intracellular receptor proteins
- The 1,25-diOH-D3–receptor complex interacts with response elements in the nuclear DNA of target cells (vitamin A) - selectively stimulates or represses gene transcription
- The most prominent actions of calcitriol are to regulate the serum levels of calcium and phosphorus

Distribution

- 1. Endogenous vitamin precursor: 7-Dehydrocholesterol
  - Converted to cholecalciferol and transported to liver bound to vitamin D–binding protein
- 2. Diet: Ergocalciferol (vitamin D2), and cholecalciferol (vitamin D3)
- Differ chemically
- Packaged in chylomicrons
- Preformed vitamin D is a dietary requirement only in individuals with limited exposure to sunlight



Metabolism

- Vitamins D2 and D3 are not biologically active
- Converted in vivo to calcitriol, the active form of vitamin D
- The first hydroxylation (calcidiol)
  - 25 position – liver - 25-hydroxylase
  - The predominant form in serum and the major storage form
- Further hydroxylated
  - 1 position – kidney - 25-hydroxycholecalciferol 1-hydroxylase
  - Formation of 1,25-diOH-D3 (calcitriol)

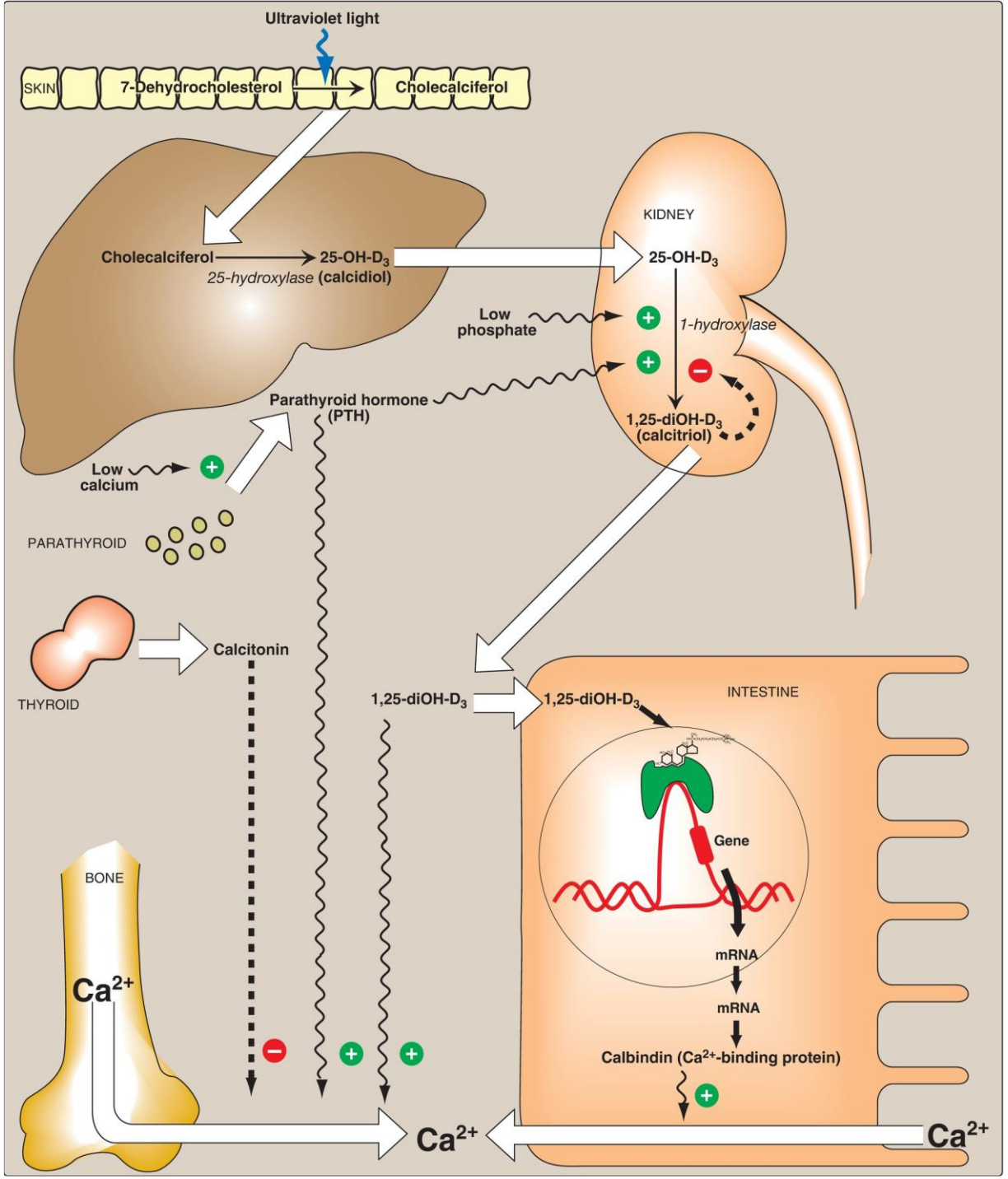
Hydroxylation regulation

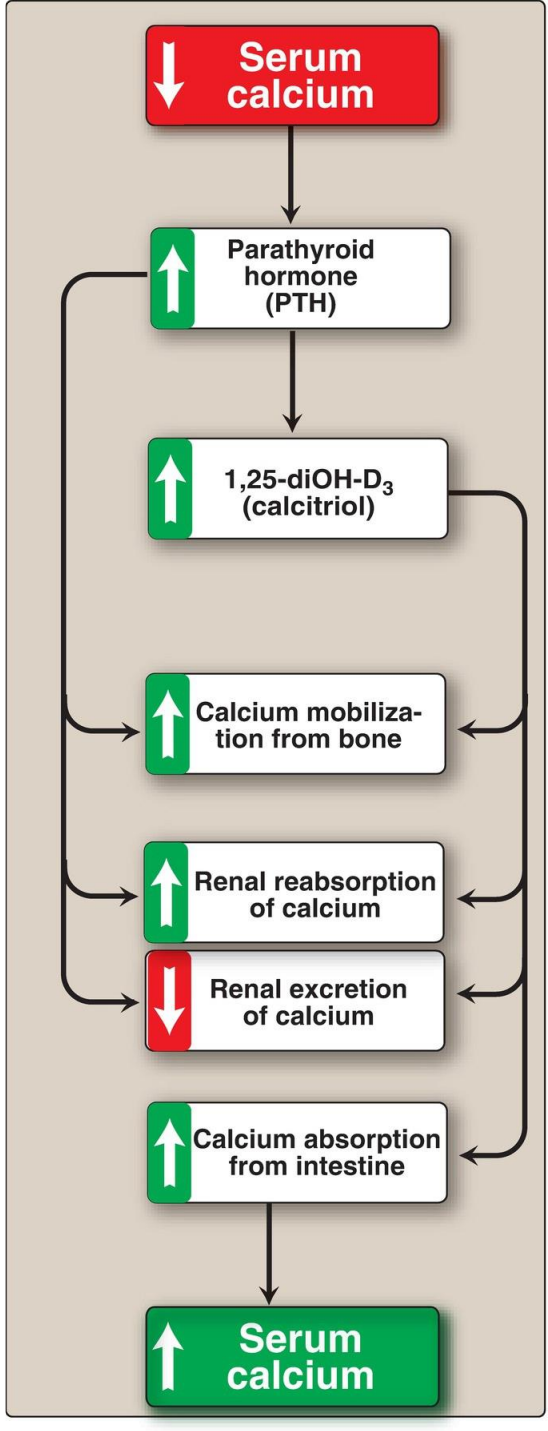
- Formation of Calcitriol is tightly regulated by the level of serum phosphate (PO_4^{3-}) and calcium ions (Ca^{2+})
- *25-Hydroxycholecalciferol 1-hydroxylase* activity is increased
  - Directly by low serum PO_4^{3-}
  - Indirectly by low serum Ca^{2+} (through PTH)
- Thus, hypocalcemia caused by insufficient dietary Ca^{2+} results in elevated levels of serum 1,25-diOH-D3
- 1,25-diOH-D3 inhibits expression of PTH, forming a negative feedback loop. It also inhibits activity of the *1-hydroxylase*

Hydroxylation regulation

Calcitriol increases intestinal absorption and renal reabsorption of phosphate

PTH decreases renal reabsorption of phosphate





Function

- To maintain adequate serum levels of Ca^{2+} (intestine, kidney, and bone)
- Increased expression of the calcium-binding protein calbindin (typical of steroid hormones)
- Bone is composed of collagen and crystals of $Ca_5(PO_4)_3OH$ (hydroxylapatite) - enhanced by PTH
- PTH and calcitriol also work together to prevent renal loss of Ca^{2+}

Distribution and requirement

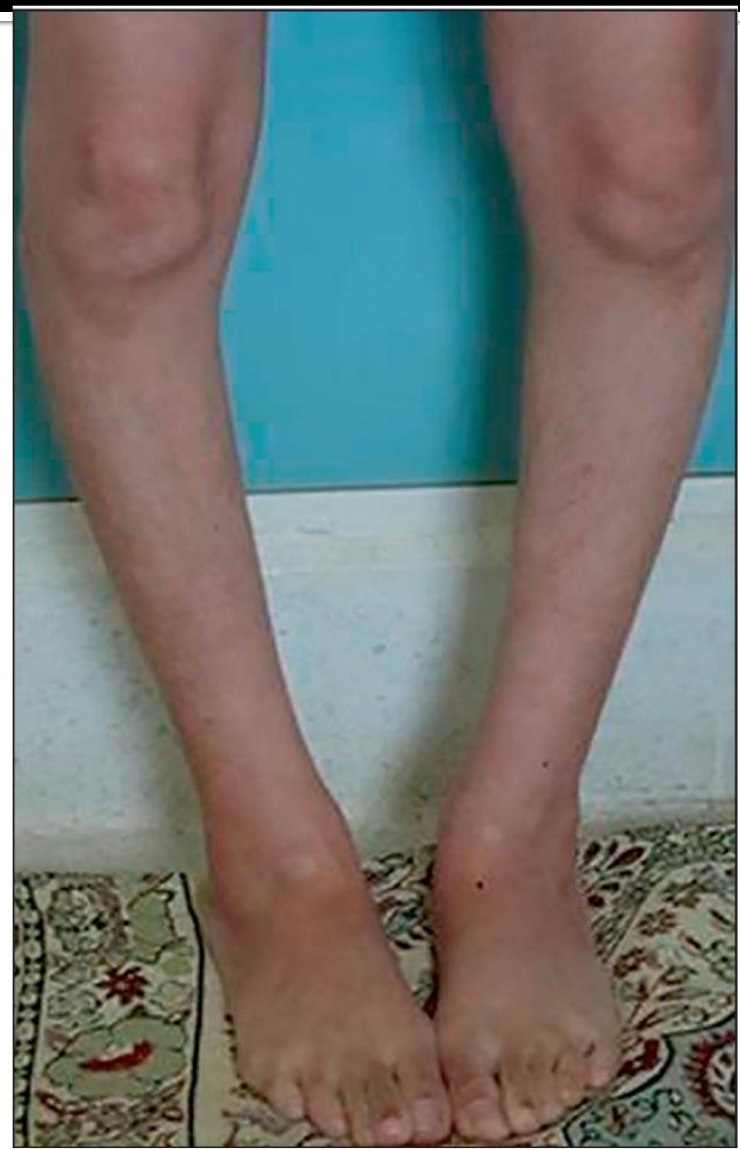
- Naturally in fatty fish, liver, and egg yolk
- Milk, unless it is artificially fortified, is not a good source
- The RDA for individuals ages 1–70 years is 15 µg/day and 20 µg/day if over age 70 years
- Experts disagree, however, on the optimal level of vitamin D needed to maintain health
- 1 µg vitamin D = 40 international units (IU)
- Because breast milk is a poor source of vitamin D, supplementation is recommended for breastfed babies

Clinical indications for vitamin D

- 1. Nutritional rickets: Vitamin D deficiency causes a net demineralization of bone, resulting in rickets in children and osteomalacia in adults
- Rickets is characterized by the continued formation of the collagen matrix of bone, but incomplete mineralization results in soft, pliable bones
- Osteomalacia, demineralization of preexisting bones increases their susceptibility to fracture. Insufficient exposure to daylight and/or deficiencies in vitamin D consumption occur predominantly in infants and the elderly

Clinical indications for vitamin D

- Vitamin D deficiency is more common in the northern latitudes, because less vitamin D synthesis occurs in the skin as a result of reduced exposure to ultraviolet light
- Loss-of-function mutations in the vitamin D receptor result in hereditary vitamin D–deficient rickets



Clinical indications for vitamin D

- 2. Renal osteodystrophy: Chronic kidney disease causes decreased ability to form active vitamin D as well as increased retention of PO_4^{3-}, resulting in hyperphosphatemia and hypocalcemia
- The low blood Ca^{2+} causes a rise in PTH and associated bone demineralization with release of Ca^{2+} and PO_4^{3-}
- Supplementation with vitamin D is an effective therapy. However, supplementation must be accompanied by PO_4^{3-} reduction therapy to prevent further bone loss and precipitation of calcium phosphate crystals

Clinical indications for vitamin D

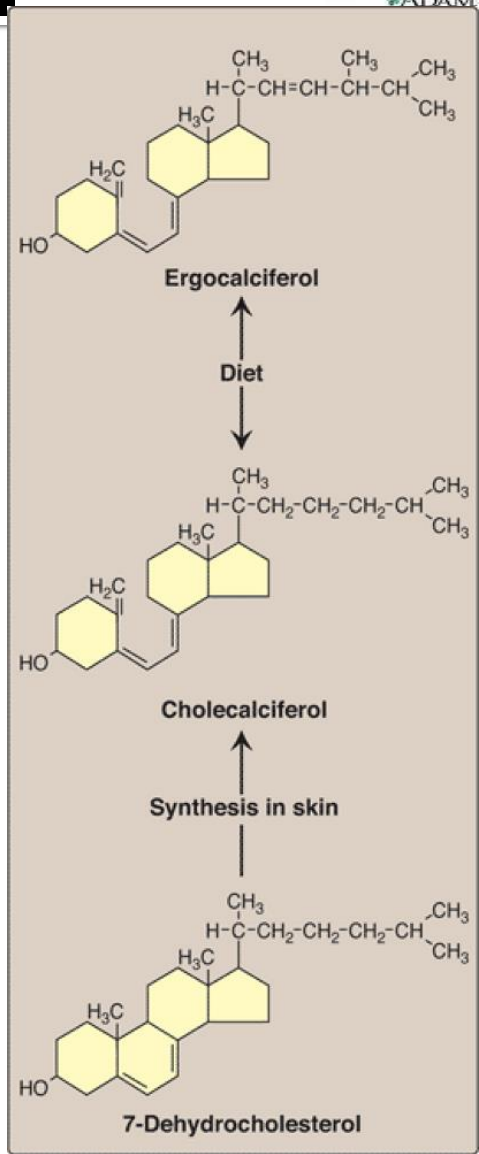
- 3. Hypoparathyroidism: Lack of PTH causes hypocalcemia and hyperphosphatemia
- PTH increases phosphate excretion. Patients may be treated with vitamin D and calcium supplementation

Toxicity

- High doses (100,000 IU for weeks or months) can cause loss of appetite, nausea, thirst, and weakness
- Enhanced Ca^{2+} absorption and bone resorption results in hypercalcemia, which can lead to deposition of calcium salts in soft tissue (metastatic calcification)
- UL is 100 µg/day (4,000 IU/day) for individuals ages 9 years or older, with a lower level for those under age 9 years
- Toxicity is only seen with use of supplements. Excess vitamin D produced in the skin is converted to inactive forms

Vitamin D

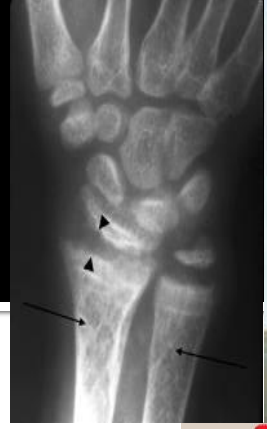
- ✓ Is a group of sterols that have a hormone-like function
- ✓ The active molecule, 1,25-dihydroxycholecalciferol (1,25-diOH-D3), binds to intracellular receptor proteins
- ✓ The most prominent actions are to regulate the plasma levels of calcium & phosphorus
- ✓ Sources:
  - ✓ Diet: Ergocalciferol (vitamin D_2, plants)
  - ✓ Cholecalciferol (vitamin D_3, animals)
  - ✓ Endogenous: 7-Dehydrocholesterol is converted to cholecalciferol in the skin

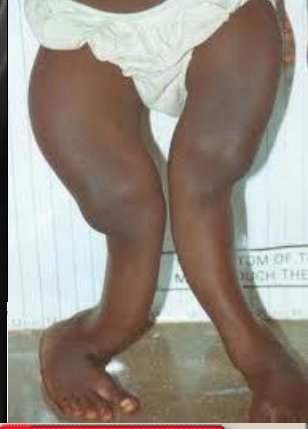


Metabolism of vitamin D

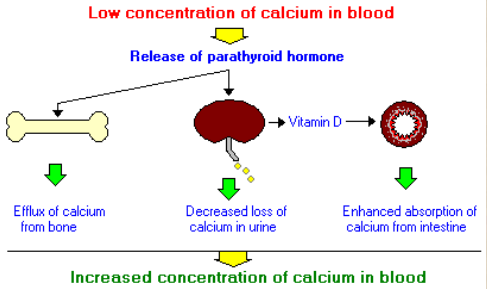
- Vitamins D_2 and D_3 are not biologically active
- Converted by two sequential hydroxylation reactions to the active 1,25-diOH-D_3
  - The first occurs in liver (25-hydroxycholecalciferol)
    - The predominant form of vitamin D in plasma
    - The major storage form
  - The second occurs in kidneys (25-hydroxycholecalciferol 1-hydroxylase)
- Regulation of 25-hydroxycholecalciferol 1-hydroxylase:
  - Increased directly by low plasma phosphate
  - Increased indirectly by low plasma calcium (PTH)
  - Decreased by excess 1,25-diOH-D3, the product of the reaction

Functions & indications

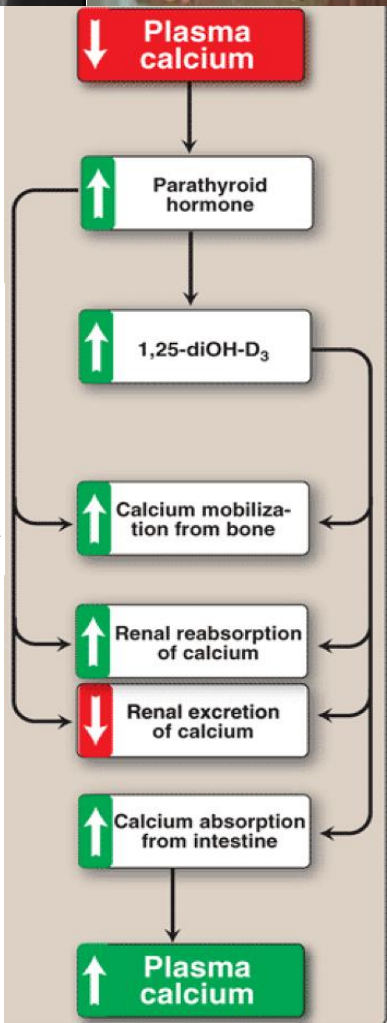




- The overall function is to <u>maintain adequate plasma levels of calcium</u>
  - 1) Increasing calcium uptake by intestines (the mechanism is typical of steroid hormones)
  - 2) Minimizing calcium loss by kidneys
  - 3) Stimulating resorption of bone when necessary



- Indications:
  - Nutritional rickets: rickets in children & osteomalacia in adults
  - Renal rickets (renal osteodystrophy): chronic renal failure
  - Hypoparathyroidism

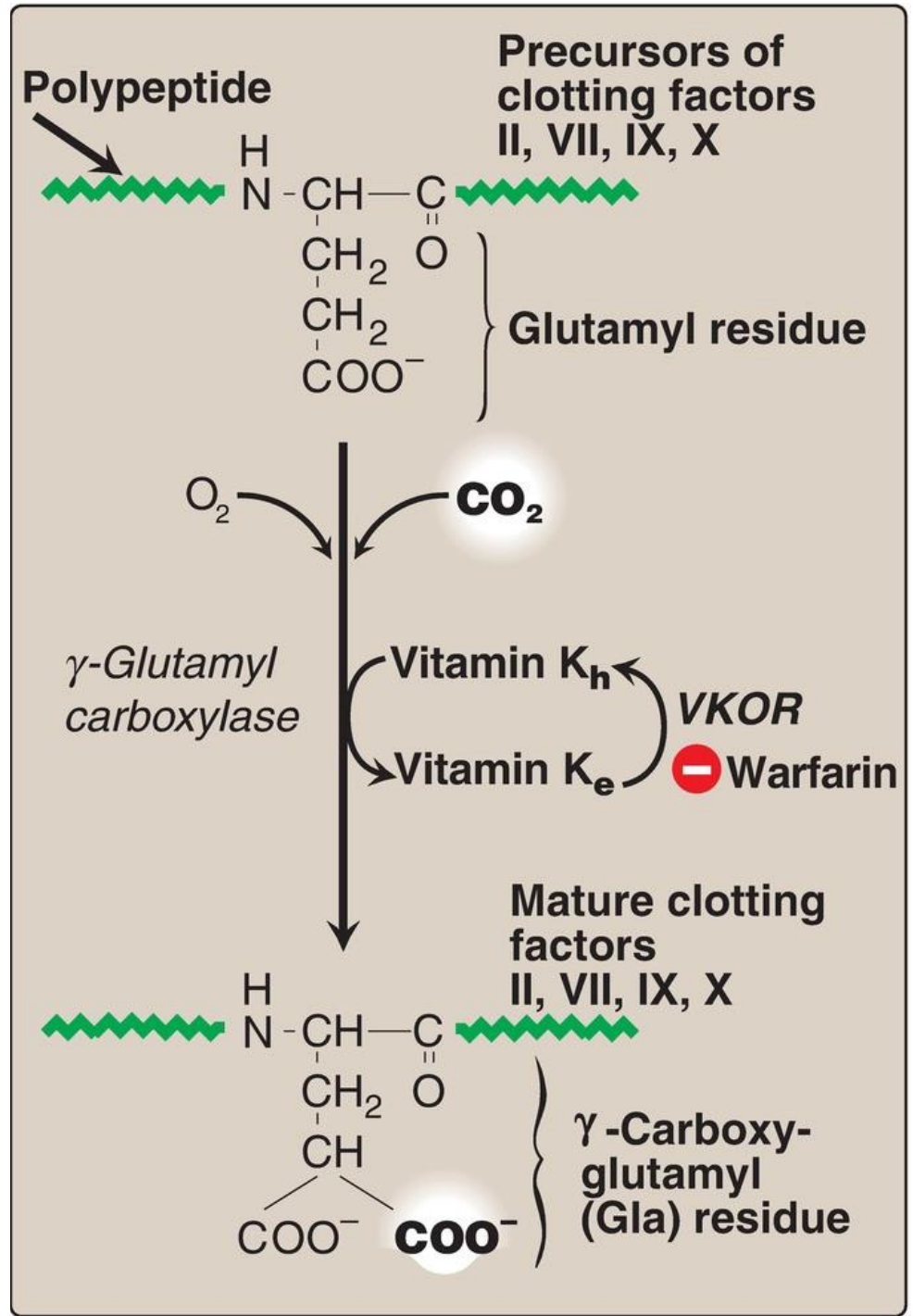


VITAMIN K

- The principal role is in the posttranslational modification of a number of proteins (most of which are involved with blood clotting), in which it serves as a coenzyme in the carboxylation of certain glutamic acid residues in these proteins
- Vitamin K exists in several active forms
- In plants as phylloquinone (or vitamin K1), and in intestinal bacteria as menaquinone (or vitamin K2).
- A synthetic form of vitamin K, menadione, is able to be converted to K2

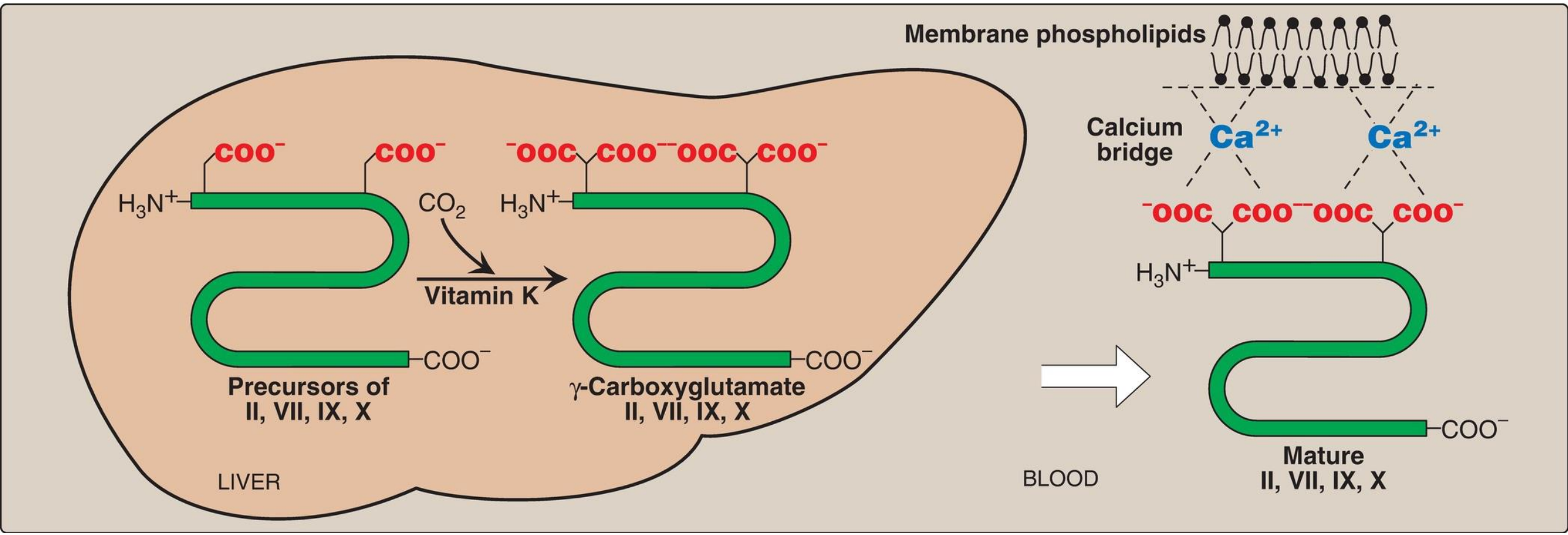
Function

- γ-Carboxyglutamate formation: Vitamin K is required in the hepatic synthesis of the blood clotting proteins, prothrombin (factor [F]II) and FVII, FIX, and FX
- The carboxylation reaction requires γ-glutamyl carboxylase, O_2, CO_2, and the hydroquinone form of vitamin K (which gets oxidized to the epoxide form)
- Formation of Gla residues is sensitive to inhibition by warfarin, a synthetic analog of vitamin K that inhibits vitamin K epoxide reductase (VKOR), the enzyme required to regenerate the functional hydroquinone form of vitamin K


Polypeptide
Precursors of clotting factors II, VII, IX, X
H
N-CH—C
CH2 O
CH2
COO⁻
Glutamyl residue
O2
CO2
γ-Glutamyl carboxylase
Vitamin Kh
Vitamin Ke
VKOR
Warfarin
Mature clotting factors II, VII, IX, X
H
N-CH—C
CH2 O
CH
COO⁻ COO⁻
γ-Carboxy-glutamyl (Gla) residue

Function

- Gla residues are good chelators of positively charged calcium ions, because of their two adjacent, negatively charged carboxylate groups
- Prothrombin–calcium complex is able to bind negatively charged membrane phospholipids on the surface of damaged endothelium and platelets
- Attachment to membrane increases the rate at which the proteolytic conversion of prothrombin to thrombin can occur


Membrane phospholipids
Calcium bridge
Ca2+
Ca2+
COO−
COO−
−OOC
COO−
−OOC
COO−
H3N+
CO2
H3N+
Vitamin K
COO−
COO−
Precursors of
II, VII, IX, X
γ-Carboxyglutamate
II, VII, IX, X
LIVER
BLOOD
−OOC
COO−
−OOC
COO−
H3N+
COO−
Mature
II, VII, IX, X

Distribution and requirement

- Found in cabbage, spinach, egg yolk, and liver
- Adequate intake for vitamin K is 120 µg/day for adult males and 90 µg for adult females
- There is also synthesis of the vitamin by the gut microbiota

Clinical indications for vitamin K

- 1. Deficiency:
- A true vitamin K deficiency is unusual – long antibiotic treatment
- In addition, certain cephalosporin antibiotics (for example, cefamandole) cause hypoprothrombinemia, apparently by a warfarin-like mechanism that inhibits *VKOR*. Consequently, their use in treatment is usually supplemented with vitamin K

Clinical indications for vitamin K

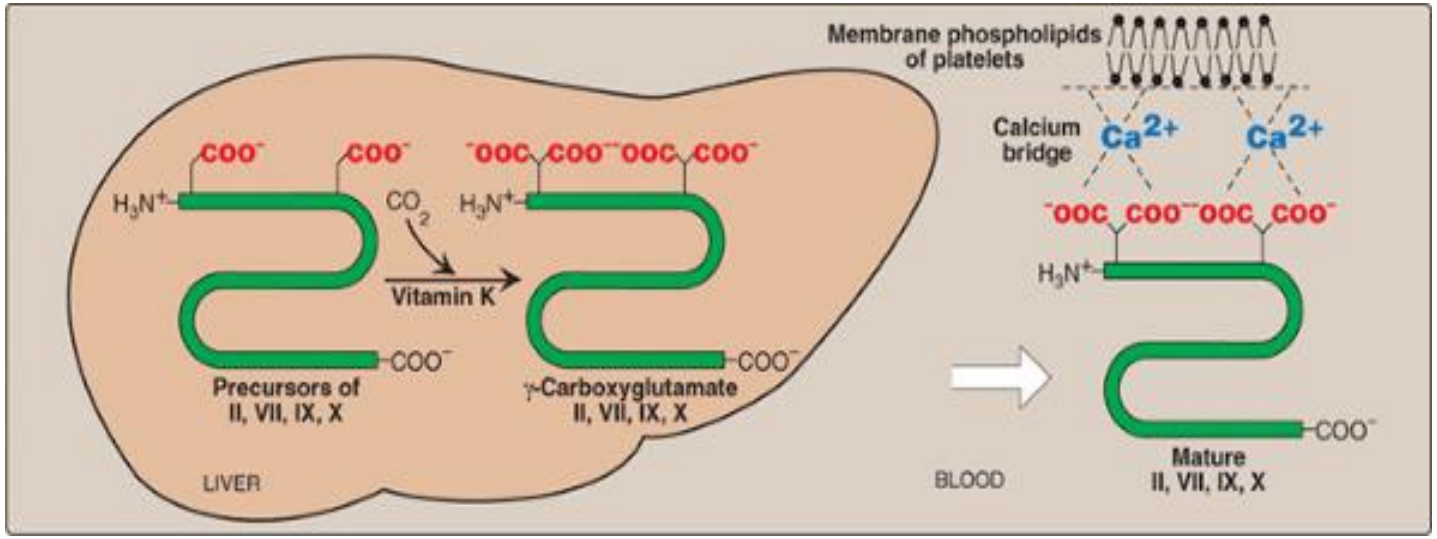
- 2. Deficiency in the newborn: sterile intestines. Human milk provides only about one fifth of the daily requirement for vitamin K, it is recommended that all newborns receive a single intramuscular dose of vitamin K as prophylaxis against hemorrhagic disease of the newborn

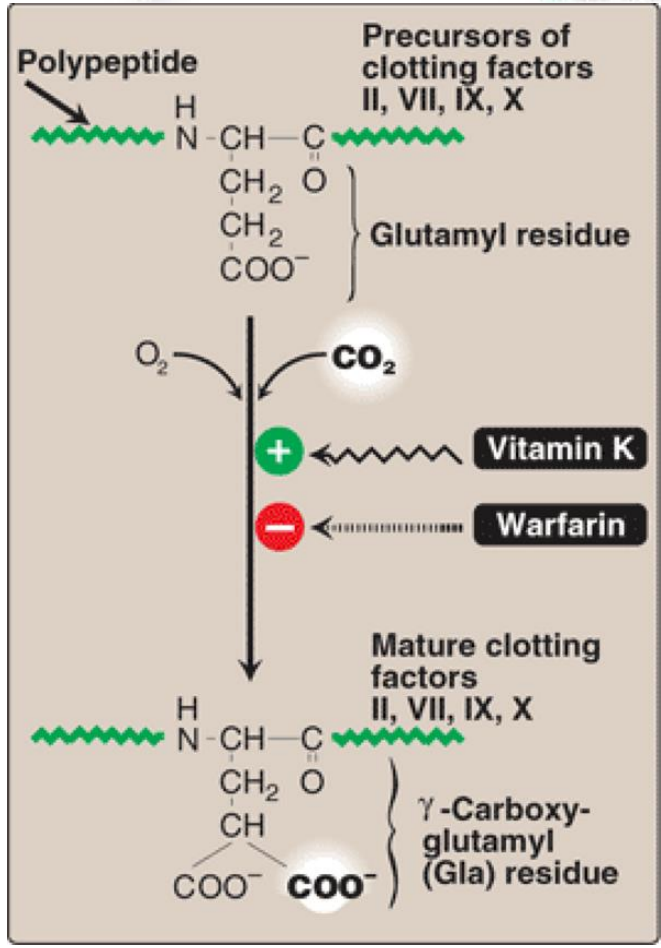
Toxicity

- Prolonged administration of large doses of menadione can produce hemolytic anemia and jaundice in the infant, because of toxic effects on the RBC membrane
- Therefore, it is no longer used to treat vitamin K deficiency
- No UL for the natural form has been set

Vitamin K

- Exists in several forms:
  - In plants: phylloquinone (vitamin K_1), Intestinal bacterial flora: menaquinone (vitamin K_2)
- Principal role: posttranslational modification of hepatic synthesis of clotting factors II (prothrombin), VII, IX, and X
- Present in low concentration in milk





Vitamin K

- Clinical indications:
  - Deficiency of vitamin K: unusual (intestinal bacteria), antibiotics
  - Deficiency of vitamin K in the newborn (sterile intestines). Human milk (20%) of the need, so it is recommended that all newborns receive a single intramuscular dose of vitamin K as prophylaxis against hemorrhagic disease

Vitamin E

- ✓ 8 naturally occurring tocopherols
- ✓ α-tocopherol is the most active form
- ✓ The primary function is as an antioxidant
- ✓ Vitamin E deficiency is almost entirely restricted to premature infants
- ✓ When observed in adults, it is usually associated with defective lipid absorption or transport