# Shree H.N.Shukla College of Science

S.Y. B.Sc. [Sem-II]   [BC 301 – BIOMOLECULES]

Prepared By :- Jinesh Kaneriya

UNIT 5 Porphyrins and Vitamins

- Vitamins are compounds that are required in the diet, either because the organism cannot synthesize them, or because the rate of usage by the organism typically exceeds the rate of synthesis of the compound. In nearly all cases, only very small amounts of these compounds are required.
- Vitamins are generally classed as either water-soluble or fat-soluble. The water soluble vitamins generally act as precursors to coenzymes; the functions of the fat soluble vitamins are more diverse and less easily categorized.
- The water-soluble vitamins are readily excreted in the urine; toxicity as a result of overdose is therefore rare. However, with few exceptions, the water-soluble vitamins are not stored in large amounts, and therefore must be continually supplied in the diet. In contrast, the fat-soluble vitamins are less readily excreted, and are deleterious (and possibly lethal) in high doses. Many of the fat-soluble vitamins are stored; for example, most well nourished individuals have a three month supply of vitamin D.
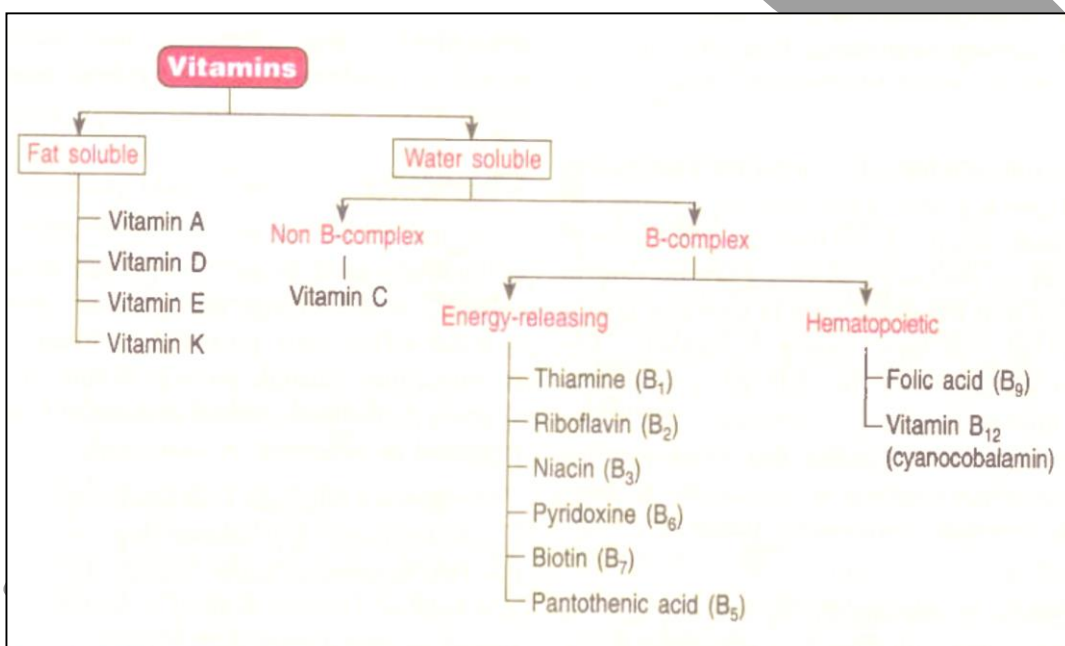

Water soluble vitamins

The water-soluble vitamins include the B complex vitamins (the actual B vitamins, biotin, and folic acid) and vitamin C.

Vitamin B1 (thiamin)

The vitamin thiamin is converted to the coenzyme thiamin pyrophosphate in an ATP-dependent reaction. Thiamin pyrophosphate is a coenzyme required for certain types of oxidative decarboxylation reactions, including the reactions catalyzed by the pyruvate dehydrogenase complex (see below) and related enzymes. Deficiency in thiamin causes **beriberi**.
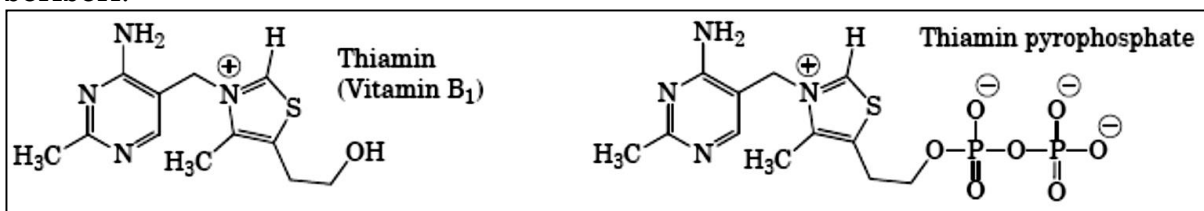
Biochemical importance

The coenzyme, thiamine pyrophosphate or carboxylase is intimately connected with the

Energy releasing reactions in the carbohydrate metabolism

1. The enzyme pyruvate dehydrogenase catalyses (oxidative decarboxylation) the irreversible conversion of pyruvate to acetyl CoA. This reaction is dependent on TPP.

2. α-Ketoglutarate dehydrogenase carbon is an enzyme of the citric acid cycle. this enzvyme its comparable wi th pyruvated ehydrogenase and requires TPP.

3. Transketolase is dependent on TPP. This is an enzyme of the hexose Pyrimidine monophosphates hunt (HMp shunt).

4. The branched chain α-keto acid dehydrogenase (decarboxylase : catalyses the oxidation of carboxylation pyrophosphate(TP P) of branched chain amino acids (valine, leucine and isoleucine) to the respective keto acids. This enzyme also requires TPP.

5. TPP plays an important role in the transmission of nerve impulse. lt is believed that TPP is required for acetylcholine synthesis and the ion t ranslocat iono f neuralt issue

Dietary Sources

Cereals, pulses, oil seeds, nuts and yeast are good sources. Thiamine is mostly concentrated in the outer layer of cereals. Polishing of rice removes about 80% of thiamine. Vitamin B, is also present in animal foods like pork, liver, heart, kidney, milk etc.

Deficiency Symptoms

The deficiency of vitamin B1 results in a condition called beri-beri. Beri-beri is mostly seen in populations consuming exclusively polished rice as staple food. The early symptoms of thiamine deficiency are loss of appetite (anorexia), weakness, constipation, n, mental depression, peripheral neuropathy, irritability etc.

Vitamin B2 (riboflavin)

- Riboflavin is the precursor to the flavin coenzymes FMN and FAD. Flavins are yellow in color and are light sensitive (flavins in food left out in the sun degrade fairly rapidly).

- Riboflavin deficiency is so rare that it has no name. Note that FMN is not really a nucleotide, and FAD is not a dinucleotide. These names are historical, and were assigned before the structures of the molecules were determined.

Riboflavin (Vitamin B₂) — Flavin mononucleotide [FMN] — Flavin adenine dinucleotide [FAD]
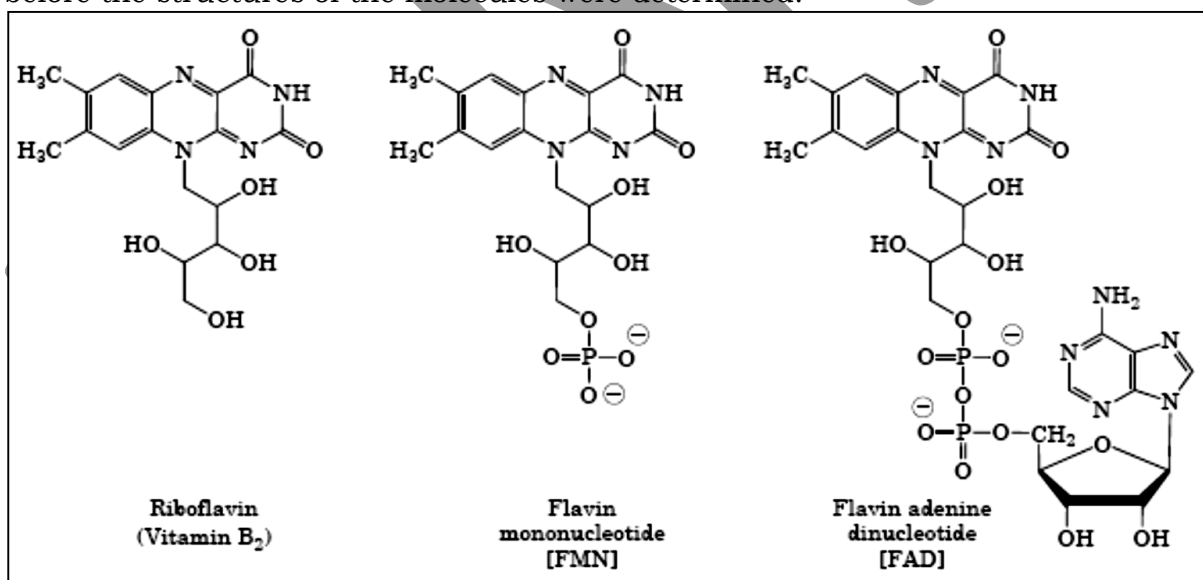
Biochemical importance

- FMN and FAD are non-covalently attached to their enzymes, but generally do not dissociate. These compounds therefore nearly always function as **prosthetic** groups, and act as storage locations for electrons within proteins.
- The isoalloxazine ring can accept or transfer electrons one at a time, although they can carry up to two electrons. This ability to accept either one or two electrons is often of critical importance for biological reactions.
- The structures below show the different electronic states observed for both flavin coenzymes.
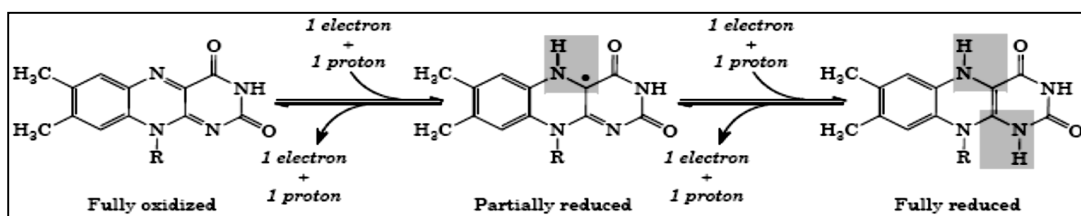
- The "partially reduced" form contains a

radical (note that the carbon with the "•" has only three actual bonds).

- This form of the compound (technically known as the **semiquinone** form of the isoalloxazine ring) is actually fairly stable.
- It is the relative stability of this state which allows flavin-containing enzymes the flexibility of transferring electrons either one or two at a time.

- The **flavin and nicotinamide coenzymes are critically important electron carriers** for a wide variety of biological processes. Both types of coenzymes are used by a number of enzymes.
- The nicotinamide coenzymes are used for carrying pairs of electrons **between** proteins, while the flavins primarily function as temporary storage for electrons **within** proteins.

Dietary Sources

Milk and milk products, meat, eggs, liver, kidney are rich sources. Cereals, fruits, vegetables and fish are moderate sources.

Deficiency Symptoms

Riboflavin deficiency symptoms include cheilosis (fissures at the corners of the mouth), glossitis (tongue smooth and purplish) and dermatitis. Riboflavin deficiency as such is uncommon

Vitamin B3 (niacin)

Niacin is the name for both nicotinamide and nicotinic acid, either of which can act as a precursor of nicotinamide coenzymes.

Biochemical importance

- Niacin is required for the synthesis of two coenzyme molecules: NAD and NADP. Note the phosphate attached to the 2′-position of the lower ribose ring in NADP, which is the only difference between the molecules.
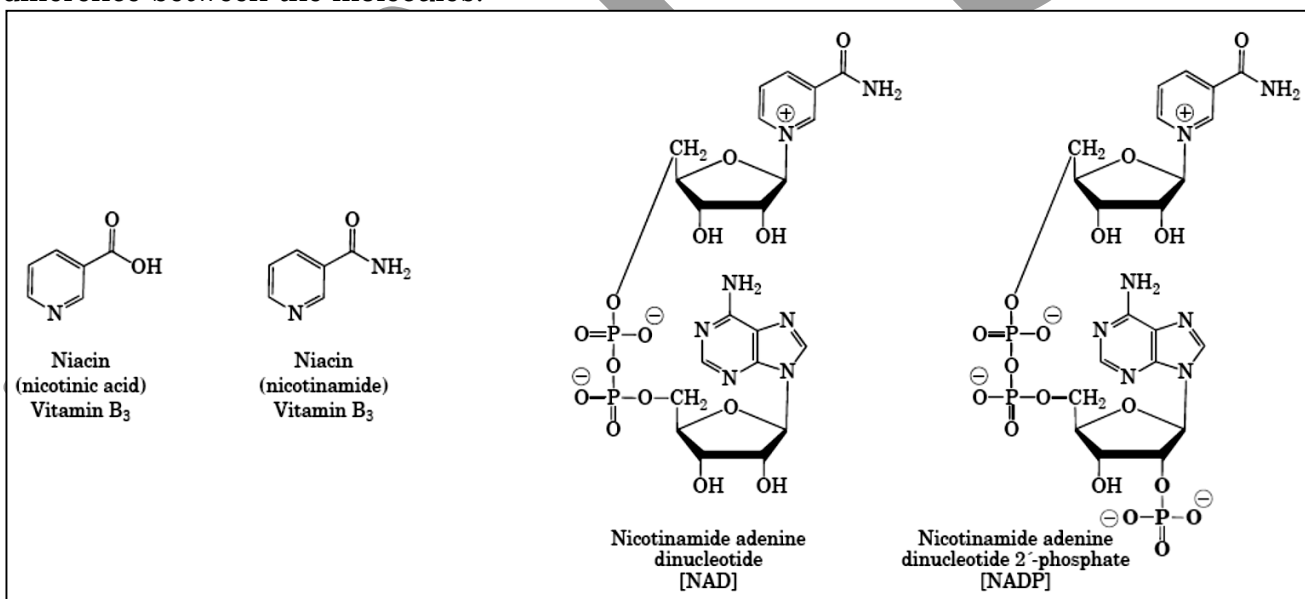
- Humans can synthesize nicotinamide cofactors from tryptophan. However, the process is somewhat inefficient; synthesis of 1 mg of niacin requires 60 mg of tryptophan.

- Niacin deficiency therefore is usually the result of a diet deficient in both niacin and tryptophan. However, some diets contain tryptophan or niacin in a biologically unavailable form.
- In corn, the niacin is poorly absorbed unless the corn is treated with alkali prior to ingestion. In the rural south of the early 20th century, this preparation step was largely ignored; the symptoms of the resulting **pellegra** (niacin-deficiency), such as sun-sensitivity and dementia, led to the pejorative term "red-neck" for individuals from this region of the US.
- Pellegra is also observed in high sorghum diets (sorghum contains niacin-synthesis inhibitors) or in some individuals taking isoniazid (isoniazid is an antibiotic used to treat tuberculosis, but also inhibits niacin uptake and synthesis).
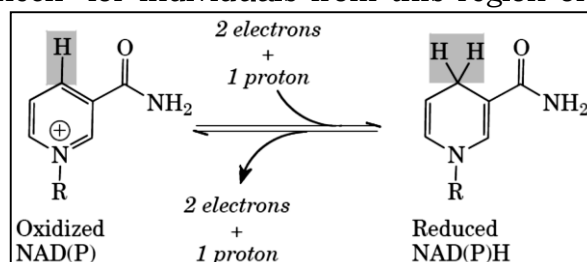
- Nicotinic acid reduces release of free fatty acids from adipose tissue, and has been used to reduce plasma cholesterol (nicotinamide is inactive for this purpose). However, some individuals cannot tolerate the level of nicotinic acid required.

- Niacin is required for the synthesis of two coenzyme molecules: NAD and NADP. Note the 2´-phosphate attached to the lower ribose ring in NADP, which is the only structural difference between the molecules.
- NAD and NADP act as **soluble electron carriers** between proteins. In effect, these compounds are substrates for enzymes involved in oxidation and reduction reactions.
- NAD is primarily involved in **catabolic reactions**. NAD accepts electrons during the breakdown of molecules for energy.
- In contrast, NADPH (the reduced form of NADP) is primarily involved in **biosynthetic reactions**; it donates electrons required for synthesizing new molecules. In most cells, NAD levels are much higher than NADH levels, while NADPH levels are much higher than those of NADP.
- The two possible electronic states for the nicotinamide cofactors are shown besides:
- The oxidized forms of both nicotinamide coenzymes can **only accept electrons in pairs**. The reduced forms of the coenzymes can **only donate pairs of electrons**.
- Note the two changes in the ring during the reduction. The addition of the electron pair is accomplished by the addition of a hydride ion to the carbon *para* to the pyridine nitrogen, and results in the loss of the positive charge on the ring.

Deficiency Symptoms

- Niacin deficiency results in a condition called pellagra (rough skin). This disease involves skin, gastrointestinal tract and central nervous system. The symptoms of pellagra are commonly referred to as three Ds. The disease also progresses in that order **dermatitis, diarrhea, dementia.**

Vitamin B5 (pantothenic acid)

- Pantothenic acid is the precursor of Coenzyme A and of the prosthetic group of the Acyl Carrier Protein domain in fatty acid syntheses.

Biochemical importance

- The active form of the cofactor is produced by formation of a peptide bond to cysteine followed by decarboxylation of the cysteine residue, and then by conjugation to the remainder of the coenzyme.
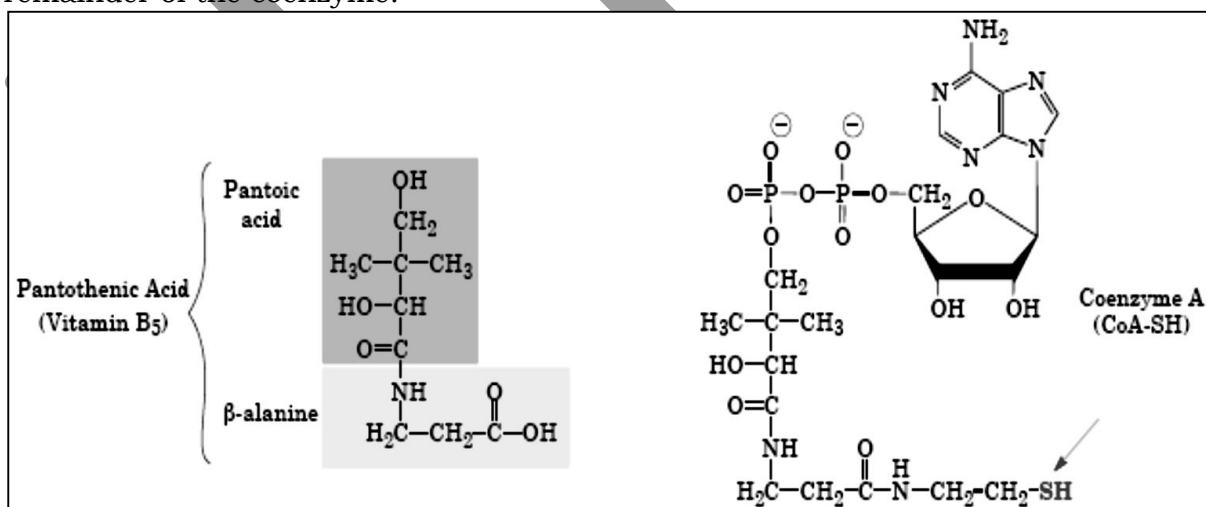
- The coenzymes produced from pantothenic acid act as carriers of acyl chains in a variety of metabolic reactions, including those in portions of the TCA cycle, in fatty acid oxidation, and in fatty acid synthesis among many others.
- Coenzyme A (usually abbreviated CoA) has a free sulfhydryl group (note the arrow in the drawing above).
- The sulfhydryl group is used to carry the carbon compounds; the remainder of the molecule acts as a "handle". In other words, the remainder of coenzyme A provides a structure that the enzyme can bind and orient when catalyzing reactions involving the attached carbon unit.
- Free CoA is often termed CoA-SH to indicate free sulfhydryl group, and to remind the reader of the attachment site for the carbon compounds.

- Pantothenic acid is readily available in most foods; deficiency in this vitamin is rare except in individuals with extremely poor diets (such as concentration and prisoner of-war camp inmates).
- An **example** of the function of several coenzymes is provided by the pyruvate dehydrogenase reaction.
- Pyruvate dehydrogenase is very large multienzyme complex (the complex contains 60 polypeptides in bacteria, and over 130 polypeptides in humans). Pyruvate dehydrogenase requires five coenzymes: thiamine pyrophosphate, lipoamide, FAD, NAD, and CoA.
- Addition of the ketoacid (pyruvate) to thiamin pyrophosphate at the position next to the N+ (carbanion addition) to form a hydroxyethyl product, with release of the carboxylate as CO_2.
- Reaction of lipoamide with the hydroxy ethyl product to re-form free thiamin pyrophosphate and acetyl lipoamide.
- Reaction of acetyl lipoamide with CoA-SH to form acetyl-CoA and dihydrolipoamide.
- Reduction of FAD by the dihydrolipoamide.
- Reduction of NAD+ by the $FADH_2$ to NADH.

Deficiency Symptoms

- It is a surpriset o biochemiststh at despite the involvement of pantothenic acid (asc oenzyme A) in a great number of metabolic reactions, its deficiency manifestations are not been reported in humans. This may be due to the widespread distribution of this vitamin or the symptoms of pantothenic acid may be similar to other vitamin deficiencies.

Vitamin B6 (pyridoxine, pyridoxal, pyridoxamine)

- Three forms of vitamin B6 can be absorbed from the diet:

Pyridoxal   Pyridoxine   Pyridoxamine
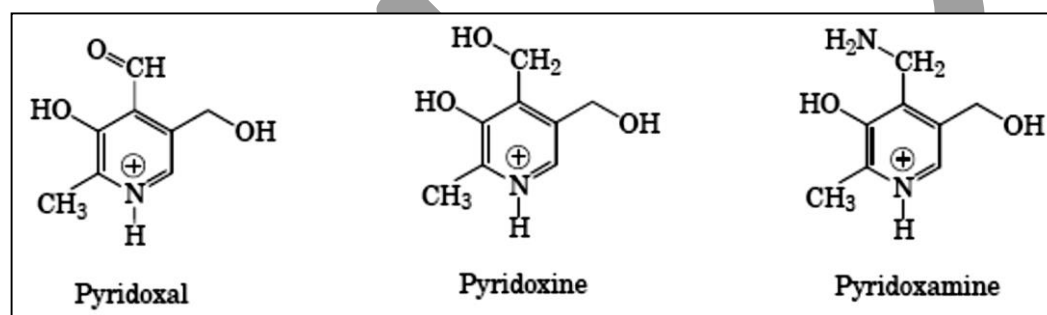
The primary cofactor form is pyridoxal phosphate:

Pyridoxal phosphate
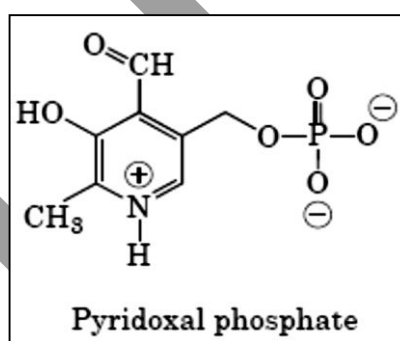
- Pyridoxal phosphate is another prosthetic group. It forms a reversible covalent association with enzymes; it is typically present as a Schiff base with a lysine ε-amino group in the resting state.
- Pyridoxal phosphate is used in a wide variety of reactions; it is especially important in reactions involving amino acids, because the aldehyde forms a Schiff base with the α-amino group, allowing stabilization of intermediates for many types of reactions.

Aldehyde   Amine   Schiff
   or   base
ketone
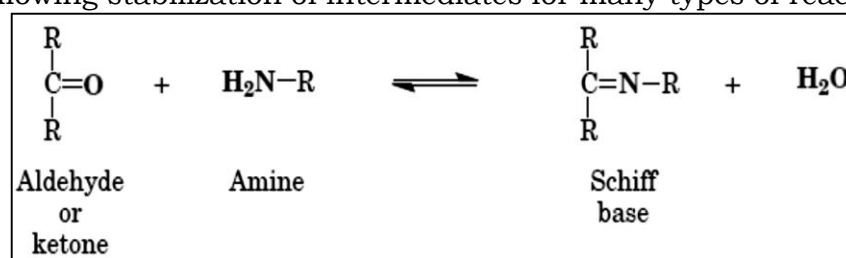
- Schiff base formation (shown above) is a readily reversible process in aqueous solution: carbon-oxygen double bonds can exchange with carbon-nitrogen double bonds. (Note that carbon-nitrogen *single* bonds do not allow this process, and therefore are in some

respects more stable than carbon-nitrogen double bonds, at least in aqueous environments.)

- Pyridoxal phosphate is also a cofactor for **glycogen phosphorylase** (it forms a Schiff base with a lysine from the enzyme); about 75% of the pyridoxal phosphate in the body is part of phosphorylase. Glycogen phosphorylase is responsible for degradation of glycogen; we will discuss this reaction later in this course.
- Deficiency in pyridoxal is fairly rare; it is either associated with other B vitamin deficiencies, with isoniazid treatment, with alcoholism, or with oral contraceptive use combined with inadequate diet (although in these cases, it is usually the breast-fed infant that suffers).
- **Examples** of pyridoxal phosphate-dependent enzymes are provided by **aspartate aminotransferase** and **serine hydroxymethyltransferase**. Aspartate aminotransferase is a member of a class of enzymes that allow exchange of amino groups from one compound to another. Serine hydroxymethyltransferase is involved in amino acid metabolism, and is the major carbon source for tetrahydrofolatedependent carbon-donation reactions.

Deficiency Symptoms

Pyridoxine deficiency is associated with neurological symptoms such as depression, Irritability, nervousness and mental confusion. Convulsions and peripheral neuropathy are observed in severe deficiency. These symptoms are related to the decreased synthesis of biogenic amine.

Biotin

Some animal carboxylase enzymes (enzymes that add CO_2 to substrates) require the water-soluble vitamin biotin.

Biochemical function

Biotin is covalently attached to the enzyme by an amide link to a lysine side chain.
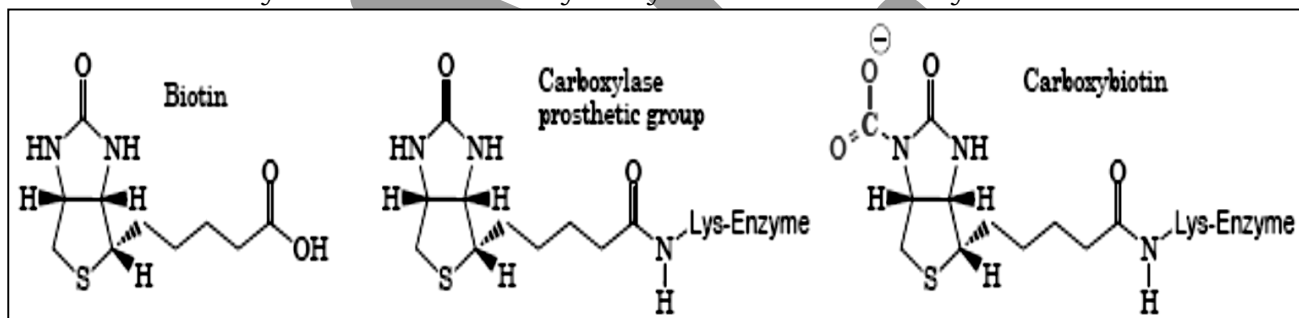
- An ATP-dependent process covalently links CO2 (using HCO3 – as the actual substrate) to one of the biotin nitrogens; the carboxybiotin then acts as a carboxylate donor for the substrate.
- Animals have four biotin dependent enzyme complexes:
- **Pyruvate carboxylase**, the first step in of the gluconeogenic pathway from pyruvate, and an important source of oxaloacetate for the TCA cycle.
- **Acetyl-CoA carboxylase**, the control step for fatty acid synthesis (this enzyme converts acetyl-CoA to malonyl-CoA).
- **Propionyl-CoA carboxylase**, which produces methylmalonyl-CoA, the first step in the conversion of propionyl CoA (generated from odd-chain fatty acid and some amino acid oxidation) to succinyl-CoA, which can enter the TCA cycle.
- β-**Methylcrotonyl-CoA carboxylase**, an enzyme required for oxidation of leucine and some isoprene derivatives.
- Biotin deficiency is sometimes found in consumers of raw chicken eggs, because eggs contain a protein called avidin that binds biotin with very high affinity and prevents its absorption.

Deficiency Symptoms

- The symptoms of biotin deficiency include anemia, loss of appetite, nausea, dermatitis glossitis etc. Biotin deficiency may also result in depression, hallucinations, muscle pain and dermatitis.

- Biotin deficiency is uncommon, since it is well distributed in foods and also supplied by the intestinal bacteria.

Folic acid

Folic acid is comprised of a pterin ring linked to **p-aminobenzoic acid (PABA)** that is in turn linked to glutamic acid.

Biochemical function

- Humans require folate in the diet because they cannot synthesize PABA (the sunscreen compound) and cannot create the link to the glutamate.
- The structure of folic acid is shown below (the shaded regions indicate the different components within the structure):
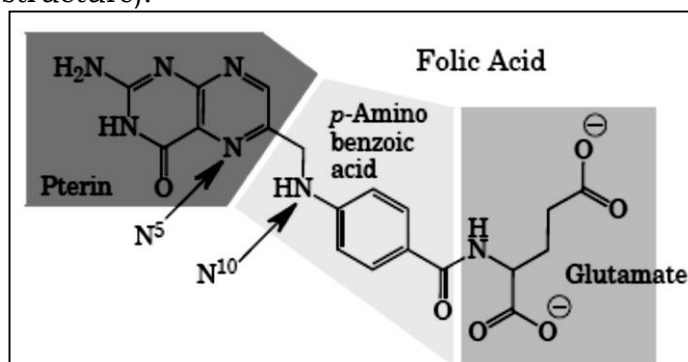
- The physiologically active form of folate has several glutamate residues (usually 5 in humans, and 7 in plants; although the absorbed form contains a single glutamate due to removal of the others in the intestines.
- Folate must be converted to the active form, **tetrahydrofolate**, by **dihydrofolate reductase**.
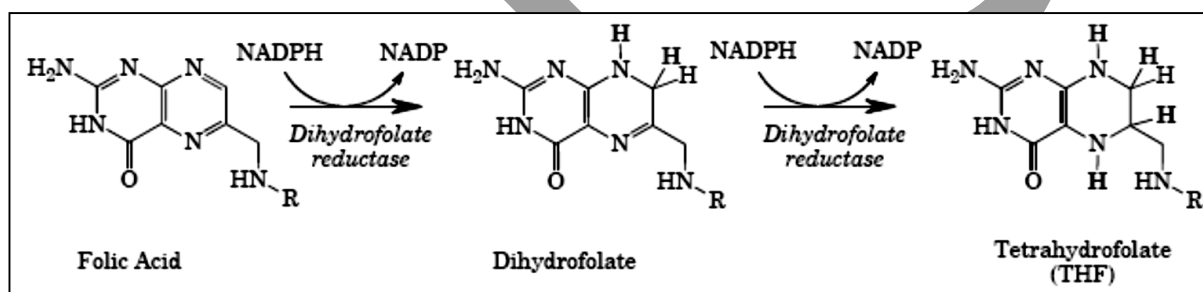
- Tetrahydrofolate acts as a single carbon carrier. The carbon can be present in most of the possible oxidation states for carbon with the exception of carbonate.

- The carbon unit is attached to the tetrahydrofolate molecule at the N5-position, N10-position, or using an arrangement that bridges both positions.

- Tetrahydrofolate is required for a number of biosynthetic enzymes. During thymidine synthesis (in the conversion of dUMP to dTMP catalyzed by thymidylate synthase), tetrahydrofolate is converted to dihydrofolate; the dihydrofolate must be reduced to tetrahydrofolate to restore the active cofactor.

- Because thymidine is required to synthesize DNA, and because dividing cells must synthesize DNA, inhibition of dihydrofolate reductase (*e.g.*, by methotrexate) prevents cell division.

- Because of its importance to growing cells, folate is required to prevent some types of birth defects. In adults, folate deficiency causes **megaloblastic anemia**.

- Folic acid is the source of the methyl group donated by methylcobalamin in the *methionine synthase* reaction, and therefore folic acid deficiency shares some symptoms with vitamin B12 deficiency.
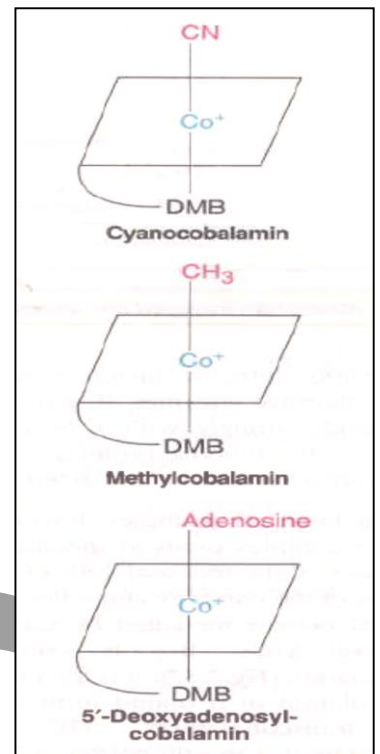
Deficiency Symptoms

- Folic acid deficiency is probably the most common vitamin deficiency, observed primarily in the pregnant women, in both developed (including USA) and developing countries (including India).
- The pregnant women, lactating women/ women on oral contraceptives, and alcoholics are also susceptible to folate deficiency. The folic acid deficiency may be due to (one or more causes) inadequate dietary intake, defective absorption, use of **anticonvulsant drugs** (phenobarbit one, d i Ia ntin , phenyltoin), and increased demand.

Vitamin B12 (cobalamin)

Vitamin B12 is a complex compound that is converted into several coenzymes.

Biochemical function

- It is used for shifting of hydrogen between carbon atoms, usually in conjunction with a shift of some other group (*e.g.*, NH2, or CH3);

- Vitamin B12 can also act as a methyl group carrier, accepting the carbon from tetrahydrofolate derivatives. In humans, vitamin B12 has only two known functions: 1) synthesis of methionine from homocysteine and 2) the rearrangement of methylmalonyl-CoA (from odd chain fatty acid metabolism and some amino acids) to succinyl-CoA.

- The structures below include the structure of the actual vitamin and of the two major coenzyme forms found in humans. (The cyanide group in cyanocobalamin is not necessarily present, and is typically an artifact of purification.) 5′-Deoxyadenosyl cobalamin is the coenzyme required by **methylmalonyl-CoA mutase**, while methylcobalamin acts as the methyl-group acceptor and donor during the **methionine synthase** reaction.

- Vitamin B12 is not made in plants; it is only synthesized by microorganisms. Strict vegetarians occasionally have difficulty obtaining enough vitamin B12, although the dietary requirements for vitamin B12 are very low (the RDA is 6 µg/day).

Deficiency Symptoms

Deficiency in vitamin B12 is called **pernicious anemia**, and may be associated with a lack of intrinsic factor, a glycoprotein required for absorption of the vitamin. Vitamin B12 is deficiently is also observed in patients who have undergone bariatric surgery.

Vitamin C (ascorbic acid)

Most animals can synthesize vitamin C from glucose, but primates are an exception.

Biochemical function

- Vitamin C acts as a reducing agent (as shown below), and is important in maintaining some metal cofactors in reduced state. It is required for proline and lysine hydroxylation (in collagen synthesis), for *dopamine β-hydroxylase* (an enzyme essential for norepinephrine and epinephrine synthesis), for bile acid synthesis, and for tyrosine degradation. It also assists in iron absorption and is a general antioxidant.
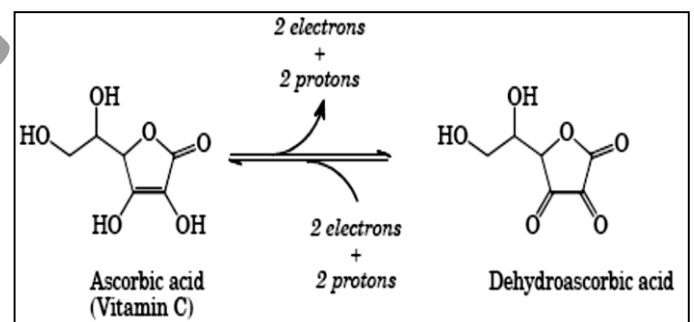
Some vitamin C is stored, especially in the adrenal. These stores can last for 3 to 4 months before symptoms of scurvy begin to appear.

Most of the functions of vitamin C are related to its property to undergo reversible oxidation-reduction i.e., inter conversion of ascorbic acid and dehydroascorbic acid.

- **Collagen formation**: Vitamin C plays the role of a coenzyme in hydroxylation of proline and lysine while protocollagen is converted to collagen.

- **Bone formation**: Bone tissue posses an organic matrix, collagen and the inorganic calcium, phosphate etc. Vitamin C is required for bone formation.

- **Iron and hemoglobin metabolism** : Ascorbic acid enhances iron absorption by keeping it in the ferrous form. This is due to the reducing property of vitamin C.

- **Tryptophan metabolism**: Vitamin C is essential for the hydroxylation of tryptophan (enzyme-hydroxylase)o hydroxyl tryptophan in the synthesis of serotonin.

- Tyrosine metabolism : Ascorbic acid is required for the oxidation of p-hydroxy phenylpyruvate(enzyme hydroxylaset)o homogentisic acid in tyrosine metabolism.

- **Folic acid metabolism** : The active form of the vitamin folic acid is tetrahydrofolate (F Hr). Vitamin C is needed for the formation of FHa (enzyme-folic acid reductase). Further, in associationw ith FHz, ascorbica cid is involved in the maturation of erythrocytes.

- **Peptide hormone synthesis** : Many peptide hormones contain carboxyl terminal amide which is derived from terminal glycine. Hydroxylation of glycine is carried out by peptidylglycine hydroxylase which requires vitamin C.

- **Synthesis of corticosteroid hormones** : Adrenal gland posses high levels of ascorbic acid, particularly in periods of stress. lt is believed that vitamin C is necessary for the hydroxylation reactions in the synthesis of corticosteroid hormones.

- Sparing action of other vitamins : Ascorbic acid is a strong antioxidant. lt spares vitamin A, vitamin E, and some B-complex vitamins from oxidation.

- **lmmunological function** : Vitamin C enhances the synthesis of immunoglobulins (antibodiesa) nd increases the phagocytic action of leucocytes.

- **Preventivea ction on cataract**: Vitamin C reduces the risk of cataract formation.

- **Preventive action on chronic diseases**: As an antioxidant, vitamin C reduces the risk of cancer, cataract, and coronary heart.

Deficiency Symptoms

- The deficiency of ascorbic acid results in scurvy. This disease is characterized by spongy and sore gums, Ioose teeth, anemia, swollen joints, fragile blood vessels, decreased immune competence, delayed wound healing.

Fat soluble vitamins

The fat-soluble vitamins have a variety of roles. Vitamin K is the only one that acts as a classical coenzyme, although retinal, a derivative of vitamin A, is a prosthetic group in the visual pigment protein rhodopsin.
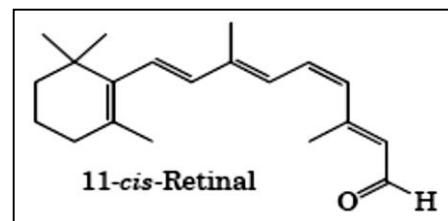
Vitamin A

Retinol is vitamin A, although most animals can convert the plant terpene β-carotene into retinol.
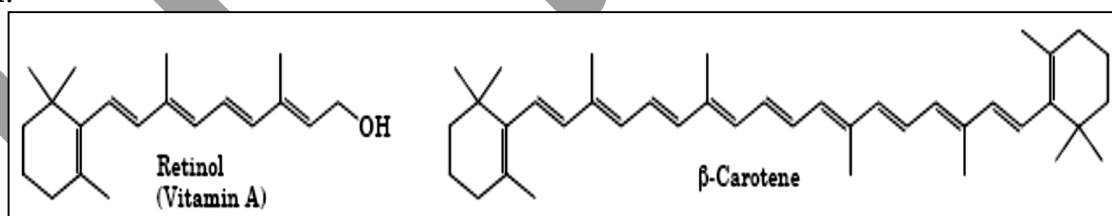
Biochemical function

- Vitamin A has a few known major functions: 1) Retinol acts as the precursor of the visual pigment 11-*cis*-retinal. Light absorption converts the 11-*cis*-retinal present as a prosthetic group in the protein rhodopsin to all *trans*-retinal; this is the first step in detecting the presence of light.

- Retinol can be converted (irreversibly) to retinoic acid. All *trans*-retinoic acid and 9-*cis*-retinoic acid are ligands for nuclear receptors, and are important in regulation of growth and development.

- Retinoic acid may have a role in glycoprotein biosynthesis. Retinol has a specific serum carrier protein, synthesized in the liver, and both retinol and retinoic acid has specific cytosolic carrier proteins.

- The main function of retinol may be to act as a precursor of retinal and retinoic acid, but retinol function is incompletely understood.

- **β-Carotene (provitamin A)** : This is found in plant foods. lt is cleaved in the intestine produce two moles of retinal. ln humans, this conversion is inefficient, hence β-carotene possesses about one-sixth vitamin A activity compared to that of retinol.

Vitamin A and vision: The biochemical function of vitamin A in the process of vision was first elucidated by Ceorge Wald (Nobel Prize 1968). The events occur in a cyclic process known as Rhodopsin cycle or Wald's visual cycle.

- Cones are specialized in bright and colour vision. Visual cycle comparable to that present in rods is also seen in bones. The colour vision is governed by colour sensitive pigments **porphyropsin** (red), **iodopsin** (green) and **cyanopsin** (blue). All these pigments are retinalopsin complexes.

- VitaminA is essent iatlo maintain healthy epithelial tissue. This is due to the fact that retinol and retinoic acid are required to prevent keratins synthesis(responsible for horny surface).

- Retinyl phosphate synthesized from retinol is necessary for the synthesis of certain glycoprotein which are required for growth and mucus secretion.

- Retinol and retinoic acid are involved in the synthesis of transferrin, the iron transport protein.

- Vitamin A is considered to be essential for the maintenance of proper immune system to fight against various infections.

- Cholesterol synthesis requires vitamin A. Mevalonate, an intermediate in the cholesterol biosynthesis, is diverted for the synthesis of

- Coenzyme Q in vitamin A deficiency. lt is pertinent to note that the discovery of coenzyme Q was originally made in vitamin A deficient animals.

Deficiency Symptoms

- Deficiency manifestations of the eyes : Night blindness is one of the earliest symptoms of vitamin A deficiency. The individuals have difficulty to see in dim light since the dark adaptation time is increased. Prolonged deficiency irreversibly damages a number of visual cells.

- Severe deficiency of vitamin A leads to **Xerophthalmia**. This is characterized by dryness in conjunctiva and cornea, and keratinisation of epithelial cells

Vitamin D

Humans can synthesize vitamin D; it is a vitamin only in humans not exposed to sunshine.
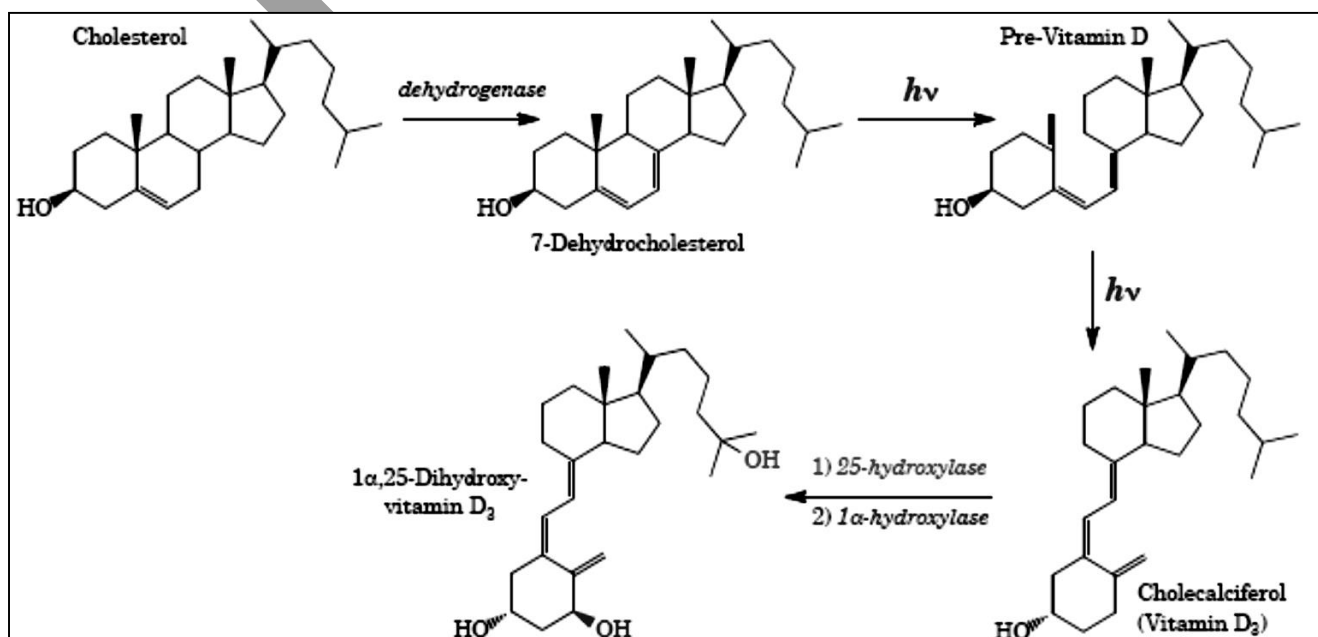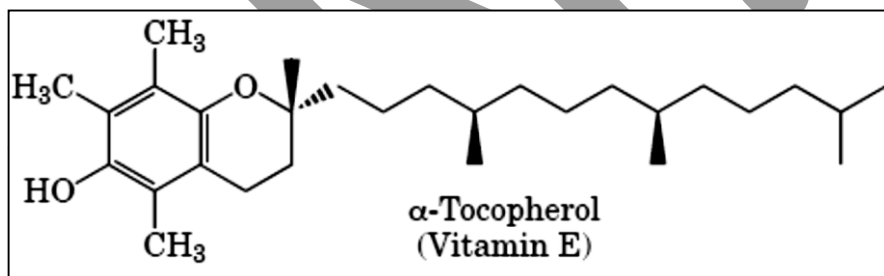

Biochemical function

- Vitamin D must be converted to the active form, 1α,25-dihydroxyvitamin D, by two sequential hydroxylations. The 1α,25-dihydroxyvitamin D acts as a hormone, and has a specific nuclear receptor.
- Deficiency in vitamin D causes Rickets (in children) and osteomalacia (in adults) due to inability to absorb calcium. Overdoses of vitamin D are associated with hypercalcemia.
- Calcitriol(1,25-DHCC)is the biologically active (form of vitamin D .lt regulates the plasma levels of calcium and phosphate Cacitriol acts at 3 different levels(intestine kidney and bone. to maintain plasma calcium(norma9 -11 mg/dl).

- **Action of calcitriol on the intestine**: Calcitriol increases the intestinal absorption of calcium and phosphate.

- **Action of calcitriol on the bone** : In the osteoblasts of bone, calcitriol stimulate calcium uptake for depositions calcium phosphate thus calcitrioils essential for bone formation

- **Action of calcitriol on the kidney**: Calcitriol is also involved in minimizing the excretion of calcium and phosphate through the kidney, by decreasing their excretion and enhancing absorption.

Deficiency Symptoms

- Vitamin D deficiency is relatively less common since this vitamin can be synthesized in the body. However, insufficient exposure to sunlight and consumption of diet lacking vitamin D results in its deficiency.

- Deficiency of vitamin D causes rickets in children and osteomalacia in adults

Vitamin E

α-Tocopherol is the most biologically active form of vitamin E. Vitamin E is an important antioxidant; it acts as a radical scavenger. It has no other known physiological function.

Biochemical Function

The biochemical functions of vitamin E, related either directly or indirectly to its antioxidant property, are given here.

1. Vitamin E is essential for the membrane structure and integrity of the cell, hence it is regarded as a membrane antioxidant.

2, lt prevents the peroxidation of polyunsaturated fatty acids in various tissues and membranes. lt protects RBC from hemolysis by oxidizing agents (e.g. H_2O_2).

3. lt is closely associated with reproductive functions and prevents sterility. Vitamin E preserves and maintains germinal epithelium of gonads for proper reproductive function.

4, lt increases the synthesis of heme by enhancing the activity of enzymes δ- aminolevulinic acid (ALA) synthase and ALA dehydratase.

5. lt is required for cellular respiration through electron transport chain (believed to stabilize coenzyme Q).

6. Vitamin E prevents the oxidation of vitamin A and carotenes.

7. lt is required for proper storage of creatine in skeletal muscle.

8. Vitamin E is needed for optimal absorption of amino acids from the intestine.

9. lt is involvedi n propers ynthesiso f nucleic acids.

10. Vitamin E protects liver from being damaged by toxic compounds such as carbon tetrachloride.

11, lt works in association with vitamins A, C and β-carotene, to delay the onset of cataract.

12. Vitamin E has been recommended for the prevention of chronic diseases such as cancer and heart disease

Deficiency symptoms

The symptoms of vitamin E deficiency vary from one animal species to another. In many animals, the deficiency is associated with sterility, degenerative changes in muscle, megaloblastic anaemia and changes in central nervous system.

Vitamin K

Vitamin K was awarded the letter K due to its role in coagulation processes (the German word for coagulation starts with a K). Menadione is a synthetic compound that can be converted into the active forms of Vitamin K.

Biochemical function

- It can be absorbed readily, but has been found to be toxic and is rarely used in supplements; the other forms require fat absorption mechanisms.

- Menaquinone is a bacterial product, and can be produced in humans from menadione (the "7" refers to the number of isoprene units; humans use 6, 7, or 9 isoprene chains).

- Phylloquinone is a plant version of the vitamin, which is used by plants in photosynthesis; the role of phylloquinone in plants is totally unrelated to the function of vitamin K in humans.
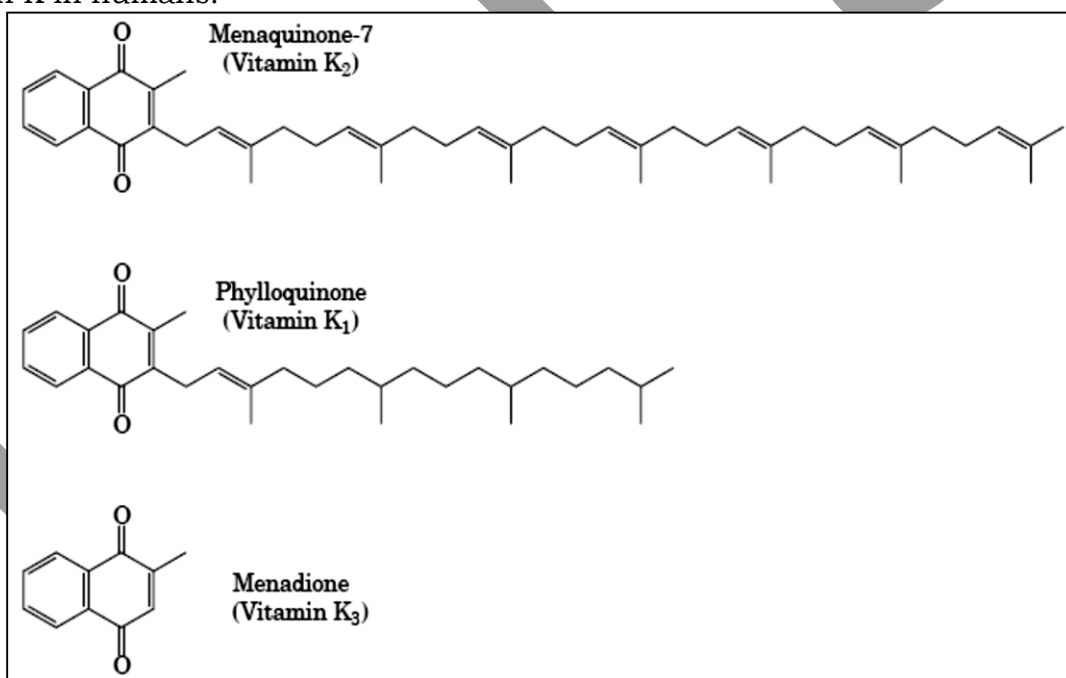
Vitamin K is required as a enzyme cofactor in the synthesis of **γ-carboxyglutamic acid**. γ-Carboxyglutamic acid is formed as a post-translational modification required for some proteins.

This unusual amino acid residue is important for the function of a number of proteins, the most notable being some of the clotting factors.

The synthesis of **γ-carboxyglutamate** from glutamate residues is a cycle. The reduced form of vitamin K acts as coenzyme for the carboxylase that produces the modified glutamate side-chain (note that this carboxylase does not require biotin).

Summary

- Vitamins are compounds that are required in relatively small amounts but that cannot be synthesized in quantities large enough to meet the normal needs of the organism.

Many vitamins, and especially the water-soluble vitamins, act as precursors for the production of coenzymes. Coenzymes allow a much larger number of reaction mechanisms than would be possible for enzymes composed only of the standard amino acids. Many of

the coenzymes act as temporary storage locations for electrons or small molecules, and as "handles" that allow proper positioning of the covalently bound substrate during the reaction.

- Vitamin-derived coenzymes are involved in a number of oxidation and reduction reactions. This is especially notable for the flavin-derived prosthetic groups FMN and FAD, and the nicotinamide-derived coenzymes NAD and NADP. Many of these enzymes catalyze physiologically reversible reactions. Due to the metabolic importance of these compounds, all biochemists need to understand the chemistry of the flavin and nicotinamide coenzymes.

- Folic acid, cobalamin, and biotin are all used for holding single carbon units. Biotin is a prosthetic group that is covalently attached to the enzyme. Tetrahydrofolate derivatives and cobalamin derivatives are used as freely diffusing carbon carriers.

- Thiamin pyrophosphate and lipoic acid are used to covalently bind small molecules (of two or more carbons). Coenzyme A is a soluble carbon carrier; it carries molecules units ranging in size from two to about 24 carbons.

- Pyridoxal phosphate is used as a prosthetic group by glycogen phosphorylase and by most of the enzymes involved in altering the α-carbon of amino acids.

- Vitamin K (and perhaps retinal) are the only fat-soluble vitamins that act as coenzymes. The other fat-soluble vitamins have non-enzymatic roles

Porphyrin and Bile Pigments

- **Defintion**: Porphyrins are tetrapyrroles (with a metal at their center) They consist of 4 pyrrole rings (which are weakly aromatic) joined by **methine (-CH=)** bridges. They have a biological significance as cofactors.

- Heme is the prosthetic group of some conjugated proteins in the body, like Hemoglobin & the cytochromes which are of paramount importance in respiration. It is also present in the enzyme catalase. The heme is a porphyrin ring containing iron in ferrous (**fe++**) condition.

- The porphyrin ring is derived from porphin, which incorporates 4 pyrrole rings connected by methene bridges (-CH=).
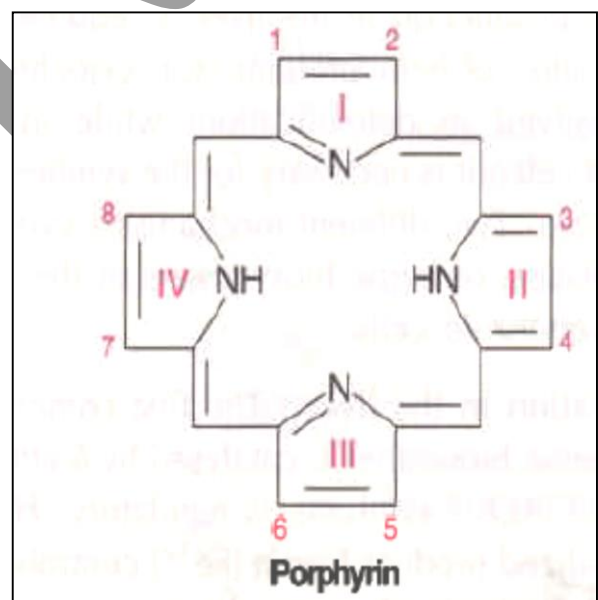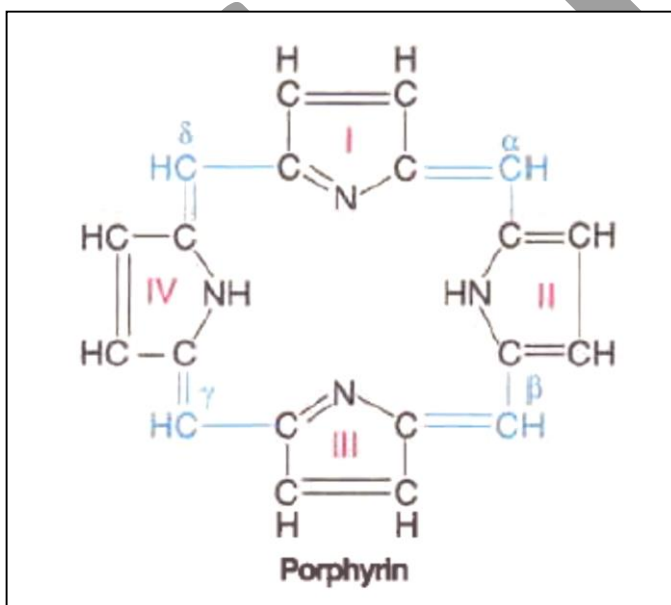
- When the porphyrin (which) has 8 (available positions accommodate) substitutents it becomes porphyrin.

- Structures of pyrole and porphyrin [l-lV] are pyrolerings; 1-8 are substituent positions;
- α,β, γ, δ are methylene (-CH=) bridge.

- (History; In 1879 German physiological chemist Hoppe Seyler showed that two of the most striking pigments of nature are related. The red iron containing heme from blood & the green magnesium complex chlorophyll of leaves have similar ring structures. Later, H. Fischer proved their structures & provided them with the names & numbering systems that are used today.)

STRUCTURES OF PORPHYRINS OR TYPES PF PORPHYRINS

- The porphyrins found in nature are compounds in which various side chains are substituted for the 8 hydrogen atoms are numbered in the porphyrin nucleus shown in figure.
-
- As a simple means of showing these substitutions, Fischer proposed a shorthand formula in which the methenyl bridges are omitted & which pyrrole ring is shown as a Bracket with the 8 substituent positions numbered as shown.
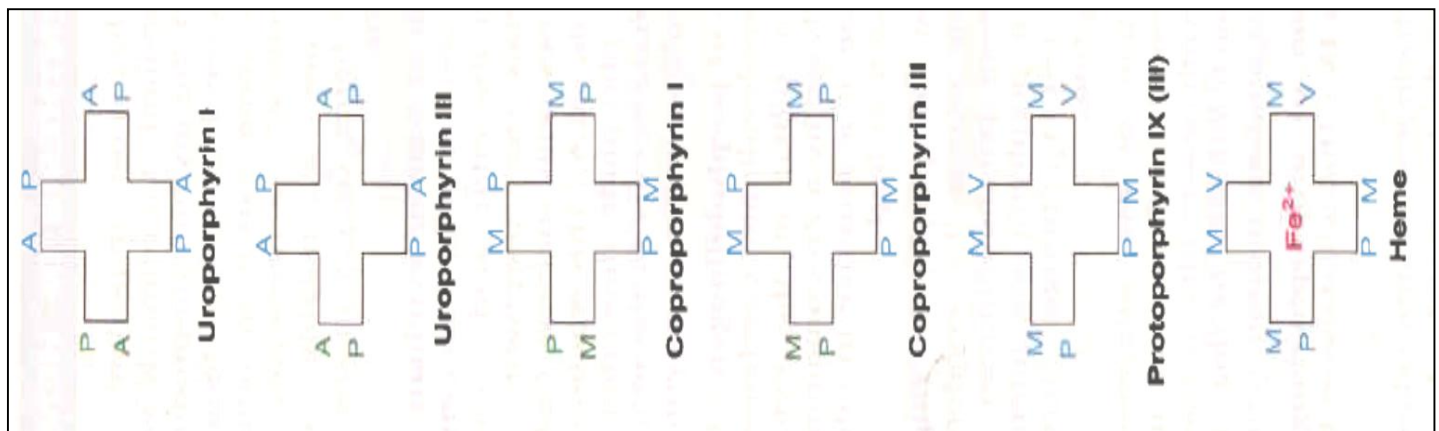
: Fischer's shorthand models of physiologically important porphyrins [A–Acetate (— CH_2COO^-); P–Propionyl (— $CH_2CH_2COO^-$); M–Methyl (— CH_3); V–Vinyl (— $CH=CH_2$)].

- Protoporphyrin has 4 methyl, 2 vinyl, & 2 propionic acid side chains. This with ferrous iron, forms heme catalysed by heme synthase or ferrochelatase. In this process, 2 hydrogen of the nitrogen atoms of 2 pyrrole are replaced by ferrous (divalent) iron,
- which is also linked to the 2 other N atoms of the porphyrin by chelation.

- **Hemoglobin**: The combination of heme with globin through different linkages (polar) & vander Waal's forces gives the red pigment hemoglobin.
- It does vital function of transport of oxygen & to a minor degree CO2 during respiration. The oxygen that is inhaled should be taken to each cell to be used for cellular respiration by mitochondria in the utilization of nutrients like glucose. Heme alone without alobin cannot combine with oxygen reversibly.
- The loose complex of Hb with O2 is called oxyhemoglobin.

- X-ray analysis has shown that the Fe of heme is linked to imidazole nitrogen of the histidine in positiobs 58 & 87 in the case of α- alpha, while they are 92 & 63 in the case of () Beta & () Gamma. With these 2 extra linkages, Fe gets a coordination valency of 6.
- The globin is made up of 4 polypeptide chain of which 2 are Alpha & are Beta in the normal adult hemoglobin (Hb A) Alpha chain 141a.a. beta chain 146 a.a.

In the formation of oxy Hb, the O-N bond being weaker, it breaks in such a way that Fe is bound only to 87th Histidine.
Various derivatives of Hemoglobin:

Heme: Ferrous plus protoporphyrin

Hematin: ferric plus protoporphyrin

Hemin: Chloride of Hematin

Hemoglobin: Heme plus native globin

Porphyrins in the body:
Porphyrins are found in urine, feces, bile, blood & bone marrow.
Role on the body: Intermediates in the synthesis of Hb, myoglobin & cytochrome.
- Uroporphyrins were first found in the urine, but they are not restricted to urine.
- Coproporphyrins were first isolated from feces, but are also found in urine.
- Uroporphyrin & coproporphyrin are different only in the nature of side chains light.

Blood	Protoporphyrins	~20mg
Blood	Coproporphrin	~1µg
Urine	Coproporphyrin	~200µg
Urine	Uroporphyrin	~20µg
Feces	Coproporphyrin	~300-1100µg
Feces	Uroporphyrin	~20µg

- In normal individual, Type III isomers are relatively abundant. Porphyrins, which are excreted, are the oxidation products of these porphyrinogens.
- Porphyrins are colored compounds while porphyrinogens are not. Porphyrinogens (highly unstable, colorless & non fluorescent) are easily oxidized to porphyrin in the presence of light, O2 or oxidizing agent. The porphyrin in strong mineral acids or organic solvents emits red fluorescence under ultraviolet. This characteristic property is used for the detection of porphyrin in urine & etc.

Clinical features of porphyrias:

- Skin lesions on sun exposed areas,
- Neurological Disturbance, Abdominal Pain,
- Mental disturbances are The most common clinical features.

Important Metalloporphyrins occuring in nature :

- In Nature, the metalloporphyrins are conjugated to proteinsto form many compounds important in biological processes. These include the following :
- Hemoglobins : Iron porphyrins attached to the protein, globin. These conjugated proteins possess the ability to combine reversibly with Oxygen. They serve as the transport mechanism for Oxygen within the blood,
- Myoglobins : Respiratory pigments that occur in the muscle cells of vertebrates and invertebrates. A Myoglobin molecule is similar to a subunit of hemoglobin.
- Cytochromes : Compounds that act as electron transfer agents in oxidation-reduction reactions. An important example is cytochrome C.
- Catalases : Iron porphyrin enzymes, are found EG. Catalase and peroxidase.
- Chlorophyll : Chlorophyll os the magnesium containing porphyrin. It is the photosynthetic pigment of plants. It is synthesized
- In the chloroplasts in the plant cell.

Bile Pigments:

- The biochemistry of the porphyrins & the bile pigments is closely related, because heme is synthesized from porphyrins & iron & the products of its degradation are the bile pigments of the iron.
- Under physiological conditions in the human adult, **$1\text{-}2 \times 10^8$** erythrocytes are destroyed per hour. Thus, in 1 day, a 70 kg human turns over approximately 6 g of hemoglobin.
- When hemoglobin is destroyed in the body, the protein portion, globin may be reutilized either such or in the form of its constituent amino acids & the iron of heme enters the iron pool, also for reuse.
- However the iron free porphyrin portion of heme is degraded in the liver, spleen & bone marrow to yield (give) bilirubin.
- The Degradation of Hb gives various bile pigments like bilirubin, biliverdib etc. The bile pigments consist of an open chain of 4 pyrrole rings joined by methylene or methane groups.

- They are derived from the proto-porphyrin IX by the oxidative breakdown of the methane bridges to form biliverdin in the reticuloendothelial cells of liver, spleen & bone marrow. Biliverdin is the first bile pigment formed.
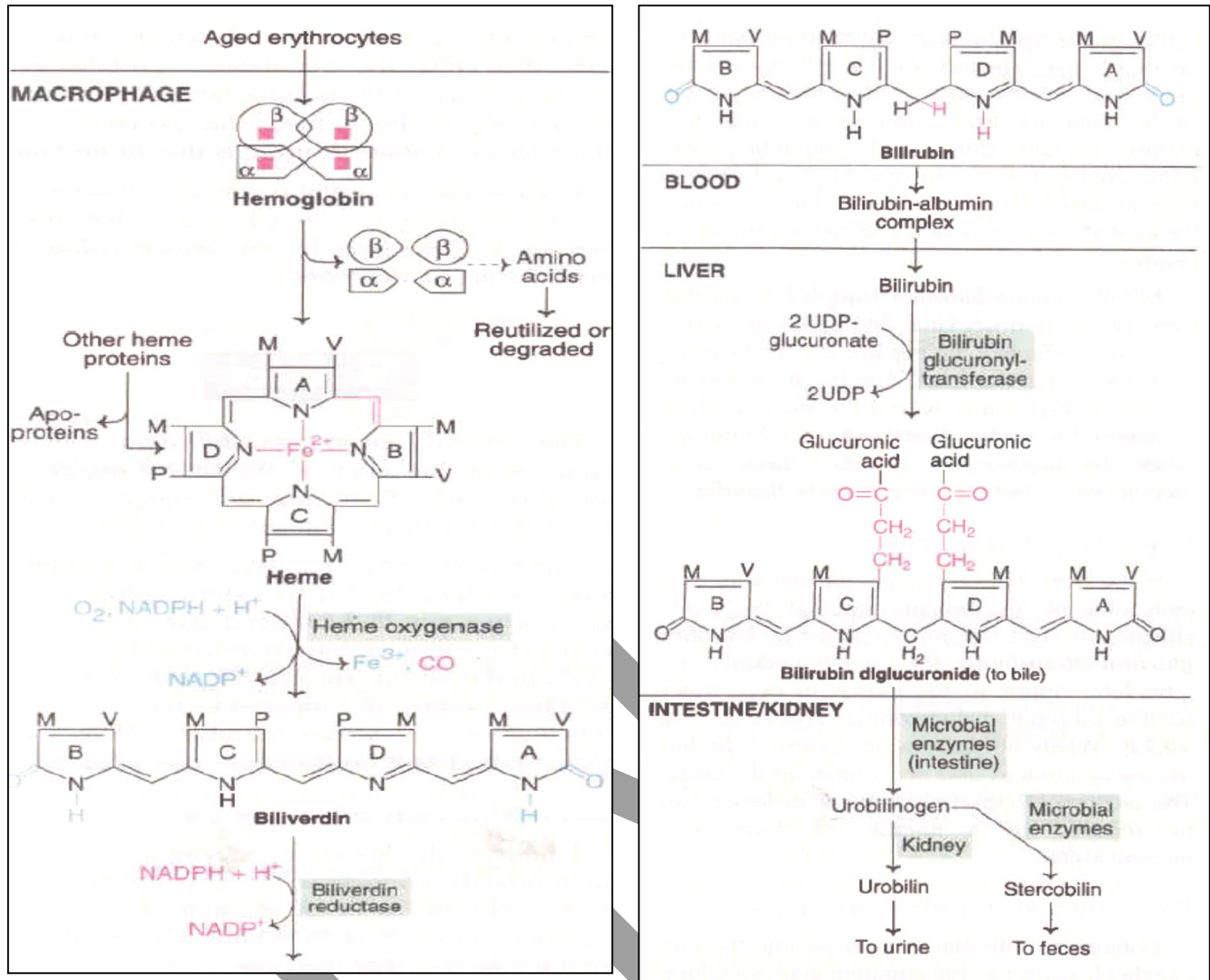
- Birds & amphibian, the green biliverdin is excreted. In Mammals, biliverdin reductase reduces the methylene bridges between pyrrole III & pyrrole IV to a methylene group to produce bilirubin.

- Bilirubin is slightly soluble in plasma and water so it binds with plasma protein albumin & is transported to the liver.

- The liver converts bilirubin to a water soluble form (it conjugates bilirubin with glucoronic acid to form bilirubin diglucoronide). This conjugated formis secreted with other components of bile into small intestine.

- In the small intestine bile pigments help in absorption of fats. After Absorption of fats, the bile pigments enter the large intestine. They are reduced the fecal flora (bacterial enzymes) to a group of colorless tetrapyrrole compounds called Urobilinogens.

- Most of the colorless urobilinogens formed in the large intestine are oxidized to colored compounds called Urobilins. Darkening of feces upon standing in the air is due to the oxidation residual urobilinogens to urobilin.

- Part of the urobilinogen is reabsorbed by entero hepatic circulation (intestine-liver) & returned to liver. Liver oxidizes it to bilirubin, which is re-excreted into bile. Normal urine therefore contains very little urobilinogens.

- The Bilirubin Accumulates In The Blood, When It Reaches A Certain Concentration (**>1 mg / 100 ml**) It Diffuses Into The Tissues, Which Then Become Yellow. The Condition Is Called Jaundic.