

## 17 Vitamins

Till the beginning of the twentieth century, it was believed that the human diet needed to contain only some of the proximate principles, that is, carbohydrates, fats, proteins, minerals and water. But when experiments were carried out with animals maintained on a chemically defined diet containing purified protein, carbohydrate, fat and mineral salts, it was found that the sustenance of life was rendered difficult. The presence of some 'accessory food factors' in natural food was recognised and many of these factors were characterised as 'organic compounds'. The first such substance isolated from rice husk which was a potent growth factor, was found to be an amine and was therefore called 'vitamine' to mean vital amine. This name had to be changed to 'vitamin' – dropping the last letter 'e' as many were not amines. Vitamins differ from each other chemically, but they share some common general functions as growth factors as well as agents promoting metabolic reactions. The deficiency or lack of a vitamin produces symptoms of hypovitaminosis which may lead to pathological conditions. The pathological, anatomical and biochemical lesions in many vitamin-deficient states have been clearly studied, and the functions of each of these vitamins have also now been established. Most of the vitamins needed by humans have to be obtained through the diet, with the exception of vitamin D and choline. The bacterial flora living in the intestinal tract are able to synthesise a few of the B-vitamins in small amounts.

Vitamins may be defined as organic molecules required in small amounts by mammals, including humans, in their diet, for metabolic purposes. Most of the B-vitamins function as coenzymes or co-factors in many enzymatic reactions.

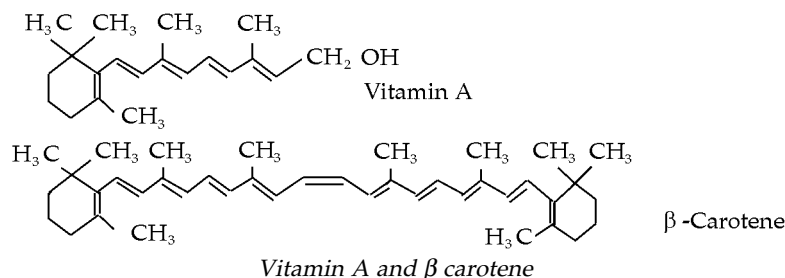
Vitamins are classified as water soluble and fat soluble, depending upon their solubility in water or fats and oils, as well as in fat solvents.

### FAT SOLUBLE VITAMINS

The fat soluble vitamins are vitamins A, D, E and K.

#### Vitamin A (Retinol) (Antixerophthalmic vitamin)

*Chemistry* Vitamin A is a complex primary alcohol, with the formula  $C_{20}H_{29}OH$ . It contains a beta ionone ring. Carotenes are the precursors (or provitamins) of vitamin A. Beta-carotene is converted into vitamin A in the tissues, that is, in the intestines and liver.



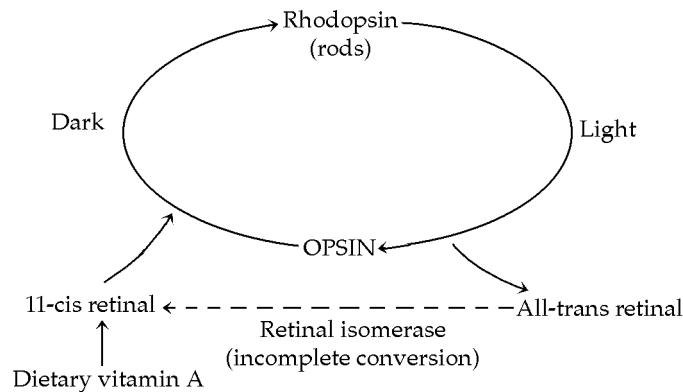
Vitamin A<sub>1</sub> and vitamin A<sub>2</sub> alcohols are known to exist. Vitamin A<sub>2</sub> has one double bond more than vitamin A<sub>1</sub>.

Vitamin A is soluble in fats and oils and in fat solvents like chloroform. Heat does not destroy it, but heating in the presence of oxygen may oxidise it slowly. Vitamin E by its anti-oxidant action spares vitamin A. Ultraviolet light destroys this vitamin. Vitamin A has a characteristic absorption spectrum at 324 mμ.

*Sources* The best sources of this vitamin are the fish liver oils, shark, halibut and cod liver oils. Animal tissues, egg yolk, cheese and butter contain moderate amounts of this vitamin. Plants do not contain any preformed vitamin A. However, carrots, yellow vegetables, spinach, etc., contain provitamins A, viz., alpha, beta and gamma carotenes and cryptoxanthine. Beta carotene is the most potent of all and is readily converted to vitamin A in the intestinal wall. Its absorption requires the presence of bile and fat in the intestine.

*Biochemical action* Vitamin A is an alcohol and is also called 'retinol.' Its oxidation products are vitamin A aldehyde, that is, retinal and vitamin A acid (retinoic acid). These three together are called 'retinoids'. Retinol probably serves as a hormone and its action is similar to that of the steroid hormone. It is bound to cellular retinol-binding protein and taken to the nucleus where it gets bound to the nuclear proteins. Retinol supports the normal function of the reproductive system in males and females.

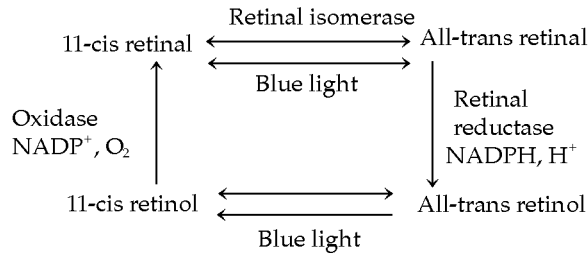
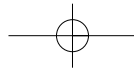
Retinal has an important role in the visual process. The photosensitive pigment 'rhodopsin' (visual purple) of the rods of the retina contains 11-cis vitamin A aldehyde and the protein opsin. On exposure to light (chiefly blue light) this gets bleached and is converted to all-trans retinal (visual yellow) and opsin. All-trans retinal has to be converted to 11-cis retinal to be used again. This conversion is brought by retinal isomerase (Fig. 17.1).



**Fig. 17.1** Wald's visual cycle

11-cis retinal then combines with opsin in the dark to reform rhodopsin. As the rods are meant for vision in dim light (night vision), rhodopsin should be available adequately for night vision. It is to be noted that all-trans retinal is incompletely converted to 11-cis retinal. Hence, a constant supply of vitamin A is needed in the diet. In vitamin A deficiency, the retinal pigment of the rods is not regenerated efficiently and hence night blindness or nyctalopia results.

Retinoic acid is required for supporting growth and differentiation of epithelia. Its distinct biochemical function is in the synthesis of glycoproteins, as carrier of oligosaccharides. There is cellular



**Fig. 17.2** Isomerisation of all-trans to 11-cis forms

retinoic acid-binding protein also but there is no corresponding nuclear protein. Retinoic acid has many other biological and biochemical *in vitro* responses from cells which are 1) increase of epidermal growth, 2) differentiation of embryonal carcinoma cells, and 3) inhibition of collagenase. Retinoic acid keeps the mucous membrane healthy and moist because it is necessary for the synthesis of glycoproteins. In vitamin A deficiency, keratinisation of the mucous membrane occurs and the skin becomes dry, scaly and rough.

Gluconeogenesis is also promoted by vitamin A. This is considered to be through adrenal corticoids.

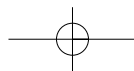
In blood vitamin A is carried by the retinol-binding protein cemented to it by transthyretin (pre-albumin).

**Colour vision** Colour vision is related to three retinol-containing pigments of the cone cells, porphyropsin, iodopsin and cyanopsin. Depending on the colour of the light falling on the retina, one or more of the above pigments undergo bleaching and get converted to all-trans retinal and the protein moiety (opsin) is released. The nerve impulses given out during this reaction are read as colour by the brain.

Porphyropsin is related to red colour while iodopsin and cyanopsin are linked up with green and blue colours, respectively. The congenital deficiency of any of these pigments may be related to colour blindness.

**Effects of deficiency** In general, vitamin A deficiency results in keratinising metaplasia, that is, replacement of the normal secretory epithelia by dry nonsecretory keratinised epithelia. The various deficiency manifestations are as follows:

1. Failure of growth in the young. The collagenous tissue is affected.
2. Night blindness (nyctalopia) due to defective resynthesis of rhodopsin.
3. Dryness of the eye (xerophthalmia) due to decreased lachrymal secretion (tears).
4. Destruction of the cornea (keratomalacia).
5. Drying of skin and atrophy of sebaceous glands, appearance of pustules around hair follicles.
6. Drying of the mucous membrane, keratinisation and loss of cilia on the epithelium of the mucous membrane of the respiratory tract resulting in infections.
7. Degeneration of germinal epithelium, thus affecting reproduction. This causes sterility in men and cornification of the vaginal epithelium in women. Though ovulation and implantation may occur, the placentas are defective and abnormalities in fetus occur, mostly resulting in fetal death.
8. Defective formation of the enamel of teeth.
9. Imbalance between osteoblasts (bone-forming cells) and osteoclasts (bone-destroying cells) causing aberrations in the shape of bones. If the foramina are involved, neurological effects due to pressure are encountered.



**Requirements** One international unit of vitamin A is equal to the activity contained in 0.3  $\mu\text{g}$  of vitamin A alcohol or 0.344  $\mu\text{g}$  of synthetic vitamin A<sub>1</sub> acetate. For adults and growing children, the recommended daily allowance is 5000 I.U. In women, during pregnancy and lactation, this allowance is increased to 6000–8000 I.U.

**Hypervitaminosis A** This is possible in children by ingestion of a large dose of vitamin A. The manifestations are painful joints, thickening of long bones and loss of hair.

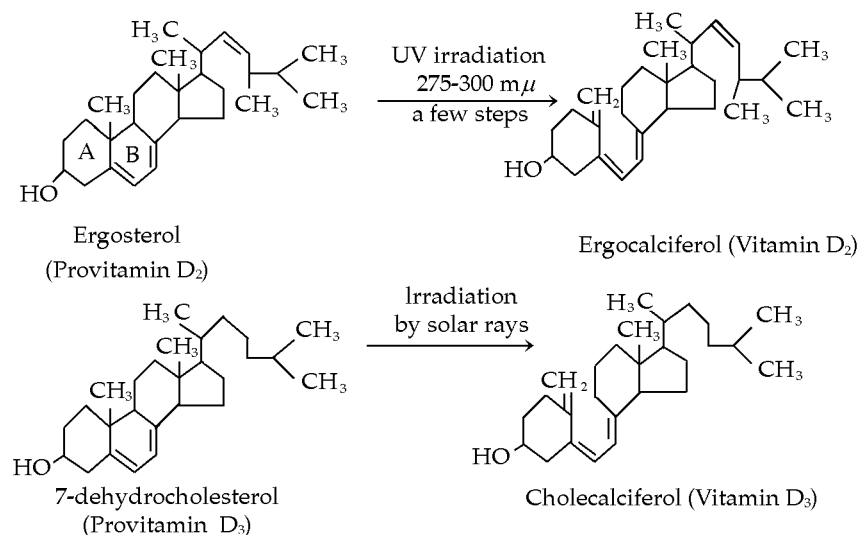
**Estimation of vitamin A** Vitamin A present in foods, such as fish liver oils, is estimated colorimetrically using the Carr–Price reaction. When a chloroformic solution of  $\text{SbCl}_3$  is added to a dilute solution of vitamin A extract from the oils, a blue colour appears which is immediately measured at 620  $\text{m}\mu$ . The colour may fade away quickly and so readings have to be taken immediately after the addition of the reagent.

Spectrophotometric methods are also available for the estimation of vitamin A at 436  $\text{m}\mu$ .

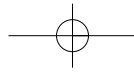
### Vitamin D (Antirachitic vitamin)

Vitamin D is the fat soluble factor present in animal fats, and is related to calcium and phosphate metabolism. Its absence leads to rickets in infants, in which there is failure of calcification of bones. This vitamin is present in the nonsaponifiable fraction of fish liver oils and is related to the sterols from which they are derived by ultraviolet irradiation. These sterols are called provitamins D.

**Chemistry** Two most important D vitamins are known. The natural vitamin D present in fish liver oil and animal fats is called cholecalciferol. Vitamin D<sub>2</sub> is not present in animal fats but may be obtained artificially by irradiation of ergosterol. The D vitamins are not, strictly speaking, sterols since the B ring is broken up between C<sub>9</sub> and C<sub>10</sub> and the methyl group on C<sub>10</sub> is converted to a methylene group, during irradiation of the provitamin sterol



*Irradiation of provitamins to yield vitamin D<sub>2</sub> and D<sub>3</sub>*



Vitamin D<sub>2</sub> and D<sub>3</sub> are quite stable compounds. They are not destroyed by heat or oxidation. They are soluble in fat solvents. They have been purified as white crystalline substances. In many of their properties they resemble sterols.

*Sources* Vitamin D<sub>3</sub> is readily synthesised in the body from cholesterol with 7-dehydrocholesterol as an intermediate. Irradiation of 7-dehydrocholesterol of skin lipids with ultraviolet exposure of the body to sunlight results in the synthesis of this vitamin. All animal fats contain vitamin D<sub>3</sub>, the best source being the fish liver oils. Egg yolk fat contains moderate amounts; milk contains only small quantities of vitamin D<sub>3</sub> but irradiated milk contains more of the antirachitic vitamins. Commercially, vitamin D<sub>2</sub> is produced by the irradiation of ergosterol obtained from ergot. Many other sterols also exhibit antirachitic properties after ultraviolet irradiation.

*Biochemical effects* Vitamin D<sub>3</sub>, as such, has no biochemical effect. It is converted to 25-hydroxy vitamin D<sub>3</sub> in the liver and further hydroxylated to 1,25-dihydroxy vitamin D<sub>3</sub> in the kidneys. This is the most potent active form of vitamin D<sub>3</sub> which regulates the metabolism of calcium and phosphorus.

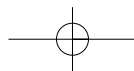
Adequate phosphate in serum along with Ca should be available for proper bone mineralisation and bone formation. When the serum phosphate level is abnormally low, the formation of 1,25-dihydroxy vitamin D<sub>3</sub> in the renal tubule is stimulated. This active form of vitamin D<sub>3</sub> is bound to the cytoplasmic receptor molecule and then translocated to the nucleus. It brings about an increase in the synthesis of the calcium-binding protein in the intestines. Thus, the intestinal absorption of both Ca and P is increased. The increased absorption of P is both indirect through increased Ca absorption and direct by an unknown mechanism. In the kidneys, 1,25-dihydroxy D<sub>3</sub> enhances the re-absorption of filtered tubular phosphate but this is usually masked by the inhibition of phosphate re-absorption by the parathyroid hormone.

1,25 dihydroxy vitamin D<sub>3</sub> affects the cross-linking of bone collagen and increases the synthesis of the vitamin K-dependent calcium-binding protein, osteocalcin of the bone, thereby influencing the mineralisation of bone tissues. This would mask the reported permissive role of 1,25-dihydroxy vitamin D<sub>3</sub> in the parathyroid hormone-mediated mobilisation of calcium and phosphate from the bone.

The active metabolite of vitamin D<sub>3</sub> also seems to prevent myopathy. 25-hydroxy D<sub>3</sub> also gives, in a minor pathway, 24,25-dihydroxy vitamin D<sub>3</sub>. This also increases intestinal Ca absorption but decreases serum Ca and P though, strangely, it promotes normal bone mineralisation. PTH causes a decrease in the formation of 24,25-dihydroxy vitamin D<sub>3</sub>. The biochemical functions of the two hydroxy compounds are reciprocally related.

*Effects of deficiency* The deficiency of vitamin D leads to rickets in children (Fig. 17.3) and osteomalacia in adults. Rickets is characterised by defective ossification leading to soft and pliable bone, bow-legs, knock-knees, bead-like swellings at the rib junctions (rachitic rosary), enlargement of the epiphyses and contracted pelvis. X-ray of the bone reveals abnormal ossification. The rachitic bone has different composition from the normal bone. The content of calcium phosphate (bone mineral) decreases while the organic matter and water content increase in the bone during vitamin D deficiency. A lowering of inorganic phosphate level in the blood is observed and a marked rise in the alkaline phosphatase, although the serum Ca level may remain more or less normal. The product of Ca x P of normal blood which is usually around 50–60 in growing children and 30–40 in adults, decreases to a lower value in the case of rickets. In this condition, dentition is also delayed.

Osteomalacia occurs in adults suffering from a deficiency of vitamin D. In osteomalacia or adult rickets, the bones become very soft and the Ca x P ratio falls to below 30. Serum calcium is lowered, unlike in rickets. In the bone, more Ca is lost than P, and sometimes there is a relative increase in



the Mg content. The ionic serum calcium is reduced to low levels resulting in neuromuscular irritability and tetany. Osteomalacia is seen in a condition known as celiac disease, which is associated with malabsorption of vitamin D from the intestines.

In both rickets and osteomalacia, a decreased serum phosphate increases the formation of 1,25-dihydroxy vitamin D<sub>3</sub>. A decreased Serum Ca<sup>2+</sup> stimulates the secretion of the parathyroid hormone which also increases the formation of 1,25-dihydroxy vitamin D<sub>3</sub>. This increases the intestinal absorption of calcium and phosphorus and enhances calcium reabsorption from the kidneys. Thus, there is replenishment of serum Ca<sup>2+</sup> and PO<sub>4</sub><sup>3-</sup>, 1,25 dihydroxy vitamin D<sub>3</sub> increases the synthesis of osteocalcin and increases **bone mineralisation in the newly forming bones**. Thus, rickets and osteomalacia are corrected by vitamin D<sub>3</sub>. However, the real effect of 1,25-dihydroxy vitamin D<sub>3</sub> **is bone demineralisation as an agent of parathyroid hormone. This is found in old bones**. If the hydroxylation of vitamin D<sub>3</sub> does not take place, rickets cannot be cured by intake of vitamin D, for example, vitamin D resistant rickets (renal rickets).

*Requirements* For infants and children, a requirement of 400 I.U. per day has been proposed. Pregnancy and lactation demand 600–800 I.U. per day.

Excess vitamin D causes toxicity, characterised by nausea, anorexia, digestive disturbances, metastatic calcification of soft tissues and calculi formation and therefore should be avoided.



**Fig. 17.3** Child with rickets (Courtesy: Department of Paediatrics, Jawaharlal Institute, Pondicherry)

*Vitamin D as hormone* Recent research on the mechanism of action of vitamin D has revealed an interesting fact – vitamin D functions more as a hormone than as a vitamin for the following reasons:

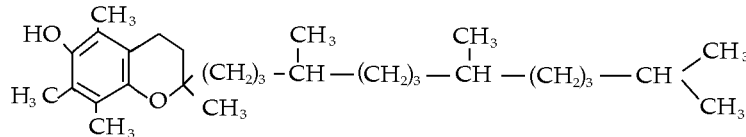
1. Vitamin D can be synthesised by the human body.
2. Vitamin D proper is inactive and is only a storage form. It has to be converted to 25-hydroxy and 1,25-dihydroxy compounds for its functions.

3. It has definite target organs – bone, kidneys and small intestines (property of hormone).
4. The formation of active forms of vitamin D<sub>3</sub> is subject to feedback control (property of hormone).
5. The active forms of vitamin D<sub>3</sub> maintain calcium homeostasis along with two other hormones, the parathyroid hormone and calcitonin. The parathyroid hormone is even considered a tropic hormone for 1,25-dihydroxy vitamin D<sub>3</sub>.
6. 1,25-dihydroxy D<sub>3</sub> resembles steroid hormones in its mode of action. In the intestines, it enters the cell and is bound to a cytoplasmic receptor molecule. This complex is translocated to the nucleus. By some mechanism it effects an increase in the synthesis of the intestinal calcium-binding protein necessary for the intestinal absorption of calcium.

### Vitamin E (Tocopherols) (Antisterility vitamin)

Vitamin E activity is exhibited by a class of fat soluble compounds – tocopherols. Alpha, beta, gamma and delta tocopherols have been obtained from natural sources, and their relationship with fertility, and prevention of muscular dystrophy has been recognised.

*Chemistry* Alpha tocopherol is the most active form. It has a chromane ring system containing four methyl groups, one phenolic group and a phytyl side chain. Beta and gamma tocopherols have one methyl group less in the ring than alpha tocopherol.



*α-Tocopherol (5.7.8. trimethyl tocol)*

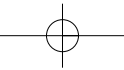
Tocopherols are very stable to heat, acids, alkalis and mild oxidising agents. However, they are destroyed on exposure to ultraviolet light for a prolonged time. Tocopherols are oily liquids and are insoluble in water.

*Sources* Among the vegetable sources of tocopherol, wheat germ oil and cotton seed oil have been found to be the richest. However, all vegetable oils and animal fat contain at least small quantities of this vitamin. Good sources of animal foods include eggs, meat, fish and liver. Milk is a poor source of vitamin E.

*Biochemical action* Vitamin E deficiency manifestations occur in certain species like rabbit, rat and guinea pig but not significantly in humans. These manifestations are related to 1) gonadal and reproductive functions, and 2) muscle metabolism and structure.

The protective effect of vitamin E on reproduction and prevention of sterility is due to its physiological function in the gonadal structures like the germinal epithelium, and the embryo. All the three layers of the embryo – ectoderm, endoderm and mesoderm – are preserved by vitamin E.

Vitamin E is required in higher animals such as poultry and cattle for fertility. There is no reliable evidence that vitamin E is necessary for fertility in humans. However, in severely impaired intestinal fat absorption, vitamin E deficiency can occur in humans. This is because vitamin E has to be absorbed with fat. The signs of vitamin E deficiency in humans are muscular weakness, creatinuria and fragile erythrocytes. Vitamin E is required for the preservation or storage of creatine in the muscles and is a most potent fat soluble antioxidant. It offers the first line of defence against the peroxidation of cellular and subcellular phospholipids. This explains the fragility of erythrocytes in vitamin E deficiency.



Selenium also offers protection against the peroxidation of membrane lipids. Selenium and vitamin E spare each other by co-operative functioning. The role of selenium as an antioxidant is through its presence in glutathione peroxidase. Cystine, selenium-containing organic compound, and vitamin E act synergistically in the prevention of hepatic necrosis. Vitamin E being the most potent antioxidant also protects vitamin A from oxidative destruction.

Vitamin E protects the unsaturated fatty acids in the erythrocyte cell membrane from oxidation and thus prevents hemolysis. In infants, some types of macrocytic anemia respond to vitamin E therapy. Vitamin E is also one of the inducers of ALA synthetase and ALA dehydratase enzymes involved in heme synthesis.

*Effects of deficiency* The deficiency of this vitamin causes resorption of the fetus in female rats and atrophy of spermatogenic structures in male rats leading to permanent sterility. The anti-sterility value is not established in humans.

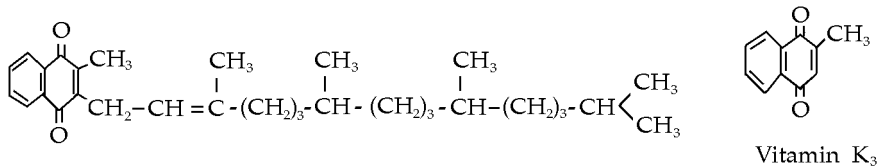
The lack of this vitamin in the diet can also cause degenerative changes in the muscles. The muscle fibres atrophy, and are replaced by connective tissue and fat. The glycogen as well as creatine content of the muscle decreases in vitamin E deficiency, while cholesterol and phospholipids are increased. In experimental animals maintained on a vitamin E deficient diet necrosis and fibrosis of heart muscles have been reported. Vitamin E is useful in the control of some eye diseases as it protects vitamin A from oxidation and some other diseases like cancer and atherosclerosis where there is oxidant assault.

*Requirements* The dietary requirement of this vitamin is related to the amount of polyunsaturated fatty acids ingested in the diet. As more unsaturated fats are ingested, more vitamin E has to be taken as well. The daily requirement of vitamin E in humans is about 15–30 mg.

There is evidence for the need of supplemental vitamin E in the diet of pregnant and lactating women and for newborn infants.

## VITAMIN K

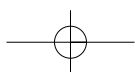
*Chemistry* A large number of compounds are found to exhibit vitamin K activity. Two important ones among them are vitamin K<sub>1</sub> or 2-methyl, 3-phytyl, 1,4-naphthoquinone, and K<sub>2</sub> or 2-methyl, 3-difarneysyl 1,4-naphthoquinone. The synthetic vitamin K is K<sub>3</sub> (menadione). It is 2-methyl 1,4-naphthoquinone. This is also fat soluble. They all have a common ring system, the difference being only in the side chain.



The K vitamins are fat soluble and are stable in heat although destroyed by strong acids and oxidising agents. They are also destroyed by sunlight.

*Sources* Green leafy vegetables, spinach and alfalfa are the best sources for vitamin K<sub>1</sub>. Vitamin K<sub>2</sub> is synthesised by the intestinal flora. Commercially, vitamin K<sub>2</sub> is produced from putrefied fish meal.

*Biochemical action* Vitamin K has been known to be required for the maintenance of normal levels of the blood clotting factors II (prothrombin), VII, IX and X, all of which are synthesised in the liver.



These are produced as inactive precursors and require gamma carboxy glutamate for their modification into the active form. The main function of vitamin K is to serve as an essential co-factor for the carboxylase enzyme that forms gamma carboxy glutamate (GLA) from glutamate residues in specific proteins. As vitamin K is involved in the formation of the active forms of clotting factors including prothrombin, it is required for normal blood clotting. Dicoumarol is antagonistic to vitamin K and prolongs clotting time. It inhibits an enzyme required for vitamin K in its function of gamma carboxylation.

An important function of vitamin K, recognised recently, is its role as co-factor in oxidative phosphorylation being associated with mitochondrial lipids. Dicoumarol which is an antagonist to vitamin K is known to uncouple oxidative phosphorylation. Ubiquinones resembling vitamin K in structure are known to take part in electron transport.

*Effects of deficiency* Deficiency of vitamin K, such as in fat malabsorption syndrome or in the sterilisation of large intestines with inadequate dietary intake results in the prolongation of clotting time and a tendency to bleed profusely. Vitamin K deficiency symptoms are observed in man in obstructive jaundice because this vitamin cannot be absorbed from the intestines in the absence of bile salts.

In the first few days of life, hypoprothrombinemia may occur in infants, as in the immediate post-natal period vitamin K is not synthesised by the intestinal flora. This can be prevented by giving vitamin K to the mother before parturition or to the infant in small amounts and this prevents melena neonatorum. (Large dose of vitamin K should be avoided as it may lead to hyperbilirubinemia.)

*Requirements* The minimum requirement of vitamin K in human adults is about 2 mg. Deficiency usually does not occur with a mixed diet. Severe deficiency does not usually occur because the intestinal flora are able to produce this vitamin in small quantities.

## WATER SOLUBLE VITAMINS

The water soluble vitamins consist of the members of the B-group (or B-complex), vitamin C and certain other compounds possessing vitamin activity. These are characterised by their high solubility in water and their insoluble nature in fats and oils.

### B-GROUP OF VITAMINS

The B-group of vitamins or the vitamin B complex group consists of 12 important members. They are:

Thiamine or aneurin (anti-beriberi vitamin), B<sub>1</sub>

Riboflavin B<sub>2</sub>

Pantothenic acid B<sub>3</sub>

Niacin and Niacinamide (anti-pellagra vitamin) B<sub>6</sub>, pellagra preventive factor

Pyridoxine B<sub>6</sub>

Biotin (anti-egg white injury factor) B<sub>7</sub>

Folic acid (pteroyl glutamic acid) and folinic acid, B<sub>9</sub>

Lipoic (thioctic acid) acid

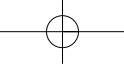
Para-aminobenzoic acid (PABA)

Inositol

Choline

Cyanocobalamin (antipernicious anemia factor) B<sub>12</sub>

Some of the members of the B vitamins are present as natural constituents in yeast, cereals and/or liver. All the B-vitamins are growth promoting factors for microorganisms and they function as coenzymes in cellular reactions.

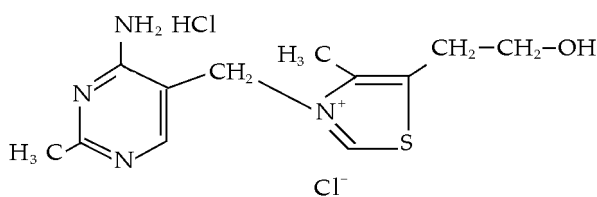


### Thiamine or vitamin B<sub>1</sub>

(anti-beriberi vitamin, antineuritic vitamin)

*Chemistry* The vitamin is present in crystalline form as thiamine hydrochloride, C<sub>12</sub>H<sub>17</sub>ClN<sub>4</sub>OS. HCl. The molecule consists of two important parts, that is, a pyrimidine part and a thiazole part, joined together by a methylene bridge. There is a quaternary nitrogen atom on the thiazole moiety. The alcoholic group on the thiazole moiety gets esterified with phosphoric acid to form the thiamine coenzyme, TPP (thiamine pyrophosphate).

Thiamine is a white crystalline compound highly soluble in water. It has the odour of yeast. It is stable on heating upto 100 °C in the dry state and in the presence of acids but heating in moist conditions and alkalis destroys it.



*Thiamine chloride hydrochloride*

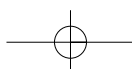
*Sources* Thiamine is widely distributed in many plant and animal foods. Whole grains, rice bran, wheat bran, nuts and yeast are some of the best sources while liver, eggs and fish are reasonably good sources. Polishing of the rice removes the vitamin B<sub>1</sub> content by 80 per cent, since most of the vitamin is present in the bran. During cooking of rice, the water in which it is cooked carries almost all the thiamine in the form of a solution.

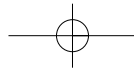
*Biochemical functions* Thiamine in the form of thiamine pyrophosphate (TPP) and in conjunction with lipoic acid is the coenzyme for the decarboxylation of alpha ketoacids like pyruvic and alpha ketoglutaric acid. Hydroxy ethyl thiamine pyrophosphate is an integral part of the pyruvate dehydrogenase complex which functions in association with lipoamide. Thus thiamine is involved in carbohydrate metabolism in all the cells of the body. It acts as co-carboxylase in the conversion of pyruvic acid to acetyl CoA and alpha ketoglutaric acid to succinyl CoA, thus helping the oxidation of glucose to CO<sub>2</sub> and also converting glucose to fatty acid. TPP also acts as a coenzyme transketolase in the HMP shunt pathway of glucose metabolism. Thus, in thiamine deficiency, there is an accumulation of pyruvic acid and other alpha keto acids in the blood. The neuritic symptoms are due to an excess of pyruvic acid.

The addition of thiamine to the diet or administration of thiamine intraperitoneally restores the condition to normal.

*Effects of deficiency* One of the first symptoms of thiamine deficiency is loss of appetite (anorexia) which leads to decreased food intake and loss of weight as well as arrested growth. The arrested growth may also be independent of decreased food intake (Fig. 17.4 and 17.5).

In humans, the symptoms of thiamine deficiency were first observed in the rice-eating population in Java and the condition was called beriberi. Beriberi also occurs in many parts of South Asia and Japan. This disease is characterised by polyneuritis with muscular atrophy, edema and cardiovascular changes. Weakness, fatigue, anorexia, headache, insomnia, gastrointestinal disorders and tachycardia are usually seen in beriberi, in its early stages. Beriberi is of four types:

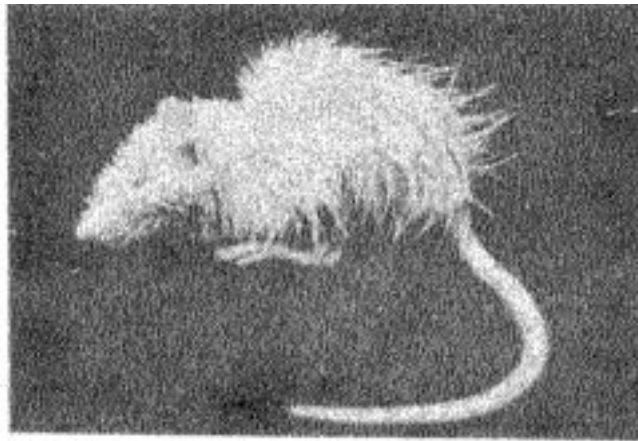




- a) dry beriberi, in which nervous symptoms or polyneuritis predominate,
- b) wet beriberi, in which the symptoms are also associated with edema and serous effusions,
- c) acute pernicious beriberi in which the heart is involved, and
- d) mixed beriberi, where all these symptoms are seen together.



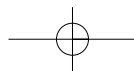
**Fig. 17.4** Normal albino rat

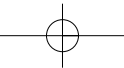


**Fig. 17.5** Vitamin B<sub>1</sub>-deficient albino rat of same age (Courtesy: Department of Biochemistry, Jawaharlal Institute, Pondicherry)

*Requirements* The requirement of thiamine is dependent upon the carbohydrate intake; the more the carbohydrate content of the diet, the greater the thiamine requirement. The addition of fat in the diet decreases the thiamine requirement. The recommended allowance for thiamine is 1.5–2 mg daily for men and 1–1.2 mg for women. Although thiamine is ordinarily non-toxic, it has occasionally produced anaphylactic shock after repeated intravenous injections.

Some of the antivitamins for thiamine are pyrithiamine and oxythiamine.

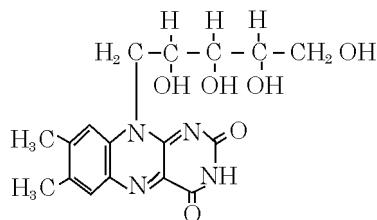




*Estimation* With alkaline potassium ferricyanide, it is oxidised to a blue compound, thiochrome, which is estimated fluorimetrically.

### Riboflavin (Vitamin B<sub>2</sub>, Lactoflavin)

*Chemistry* Riboflavin is a yellow orange compound which is soluble in water to a limited extent. Its aqueous solutions exhibit greenish-yellow fluorescence. Riboflavin is heat-stable but alkali-labile and photo-labile. Exposure to sunlight destroys this vitamin. It is stable towards acids and oxidising agents. Chemically, it contains the 6,7-dimethyl isoalloxazine ring which is linked by the central nitrogen atom to a D-ribose moiety.



*Riboflavin*

*Sources* Riboflavin is widely distributed in plant and animal sources. Milk, liver, kidney and eggs are the best sources of this vitamin. Vegetables, fruits and roots also contain moderate quantities of this vitamin. Cereals, grain and seeds do not contain much riboflavin.

*Biochemical functions* In combination with proteins, riboflavin coenzymes, like flavin mononucleotide (FMN) and flavin adenine dinucleotide (FAD), function in various dehydrogenation reactions. Many of these flavoproteins or yellow enzymes are distributed widely and take part in tissue respiration, where FAD and FMN act as components of the electron transport system. FAD is the coenzyme for diaphorase, D-amino acid dehydrogenase, glycine oxidase, xanthine dehydrogenase and fatty acyl CoA dehydrogenase, while FMN is a constituent of cytochrome C-reductase and L-amino acid dehydrogenase. The flavoproteins are therefore called flavo-enzymes which are oxidation-reduction enzymes. FMN and FAD are tightly but not covalently bound to proteins and hence function as prosthetic groups. Many flavoproteins contain metal atoms also and are actually metalloflavoproteins.

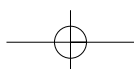
*Effects of deficiency* Pellagra is associated with a lack of niacin and sometimes riboflavin. Many of the symptoms like dermatitis could be cured by combined therapy with niacin and riboflavin.

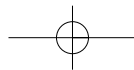
In man, cheilosis (fissures at the angles of the mouth or angular stomatitis (Fig. 17.6), glossitis, inflammation of cornea, bloodshot eyes, dimness of vision, photophobia, itching, burning and dryness of eyes and redness of the conjunctiva are reported. The increase in the blood supply to the eye (increased vascularisation of the cornea) is intended to furnish more oxygen to avascular tissues due to diminution of oxidations in such a deficient state.

*Requirements* For children and adults, the recommended daily allowance of riboflavin is 1.6–2 mg.

Some of the antivitamins are galactoflavin and diethyl derivative of riboflavin.

Estimation is done either by using a fluorimeter, taking advantage of the fluorescence of riboflavin solution, or by microbiological assay using *Lactobacillus casei*.



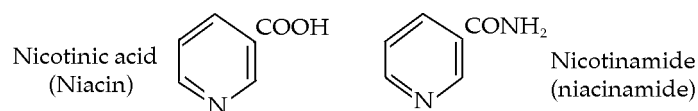


**Fig. 17.6** Angular stomatitis and angular conjunctivitis in Vitamin B<sub>2</sub> deficiency  
(Courtesy: Padmashri Prof. G. Venkataswamy, Professor of Ophthalmology and Vice Dean, Madurai Medical College (Director, Aravind Eye Hospital, Madurai))

### Niacin and niacinamide

Niacin is the pellagra-preventing factor, which is now recognised as the constituent of two of the important coenzymes, NAD<sup>+</sup> and NADP<sup>+</sup>, concerned with a cellular oxidation reactions.

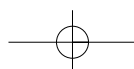
*Chemistry* Niacin is nicotinic acid and niacinamide is nicotinamide. Both these compounds contain a pyridine ring system. Originally, niacin or nicotinic acid was obtained by the oxidation of nicotine, the alkaloid present in tobacco leaves. Niacin and niacinamide are white crystalline, water-soluble compounds.

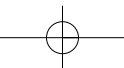


*Niacin and niacinamide*

*Sources* Liver, fish, yeast, beans and peanuts are the best sources for niacin, in which it is present mostly as niacinamide. Wheat and rice bran as well as vegetables are moderate sources but milling of grains and loss of bran results in loss of this vitamin. Milk and fish contain only a low amount of niacin. However, supplementing the diet with these foods helps to provide an ample amount of tryptophan which is readily converted to niacin in the liver. (For a full discussion on the conversion of tryptophan to niacin, see Chapter 14.)

Corn is poor in tryptophan and hence in the available niacin.





**Biochemical functions** Niacin in the form of niacinamide is a part of the coenzymes, NAD<sup>+</sup> (DPN<sup>+</sup>, coenzyme I) and NADP<sup>+</sup> (TPN<sup>+</sup>, coenzyme II). NAD<sup>+</sup> is concerned with cellular respiration as a component of the electron transport system. Many dehydrogenases use NAD<sup>+</sup>, some use NADP<sup>+</sup>, and a few enzymes can use either. NAD<sup>+</sup> and NADP<sup>+</sup> function as 'electron-sinks' in biological oxidation. They are loosely bound to the enzymes, dialysable and hence typical coenzymes.

NADPH is required for the synthesis of fatty acids and cholesterol and to reduce oxidised glutathione to glutathione.

**Effects of deficiency** Niacin deficiency symptoms are exhibited usually only by humans and dogs among the mammals. The deficiency of this vitamin causes 'pellagra' in man and 'canine black tongue' in dogs. Pellagra (rough skin) is a disease characterised by bronzing and thickening of the skin leading to inflammation, this being found to occur on the back of the hands, forearms and the neck. The dermatitis of pellagra can be prevented by the administration of niacin, the response being spectacular. Other manifestations of pellagra are diarrhea, angular stomatitis, glossitis and dementia. The three 'd's associated with symptoms of pellagra are dermatitis, diarrhea and dementia.

**Requirements** The recommended allowance for children is 6–14 mg per day and for adults 17–20 mg. The requirement is increased in women during pregnancy and lactation. The niacin requirement is influenced by the protein content of the diet, since niacin can be synthesised in the liver from the amino acid tryptophan. It has been calculated that 1 g of good quality protein contains about 60 mg of tryptophan from which 1 mg of niacin can be synthesised.

Hypervitaminosis leads to flushing due to vasodilatation and G.I. distress. Blood pressure is reduced. Heart patients with already low blood pressure therefore should not be given niacin though it offers beneficial effects in atherosclerosis, as it lowers serum cholesterol, while the amide does not.

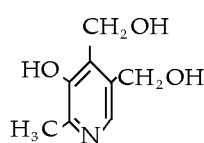
Some of the antivitamins are pyridine 3-sulphonic acid and 3-acetyl pyridine.

**Estimation** is done by: 1) treating with cyanogen bromide and coupling with aniline, and 2) by microbiological assay using *Lactobacillus arabinosus*.

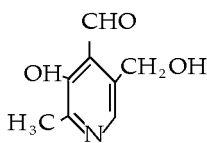
**Detoxication** Excess of niacin is detoxified by methylation of its pyridine nitrogen (protective synthesis) and excretion.

### Pyridoxine (Vitamin B<sub>6</sub>)

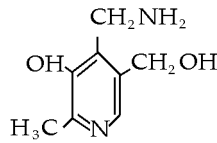
**Chemistry** Pyridoxine, pyridoxal and pyridoxamine, are all interconvertible in the body and can act as vitamin B<sub>6</sub>. Their structures are given below:



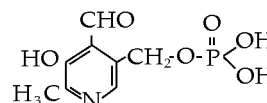
pyridoxine



pyridoxal

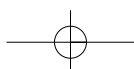


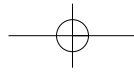
pyridoxamine



pyridoxal phosphate (coenzyme)

Pyridoxine, pyridoxal and pyridoxamine are all water soluble. They are also soluble in alcohol and slightly soluble in certain fat solvents. Pyridoxine is resistant to heat, but sensitive to light, ultraviolet rays and alkali. Pyridoxal and pyridoxamine are less stable in heat and are liable to be destroyed if heated to high temperatures.





*Sources* Egg yolk, fish, milk, and meat are the richest sources of vitamin B<sub>6</sub>. Vegetables like cabbage and legumes and foods such as whole grains and crude molasses contain moderate amounts of this vitamin.

*Biochemical functions* Pyridoxine, pyridoxal and pyridoxamine are interconvertible in the body. They all function as their phosphorylated derivatives as coenzymes for certain enzyme systems, the most important of them being the transaminases. The latter enzymes are important in the conversion of amino acids to ketoacids and vice versa and in integrating protein and carbohydrate metabolisms. Biosynthesis of several physiologically important compounds like porphyrins from succinyl CoA and glycine, nicotinic acid from tryptophan and gamma aminobutyric acid (GABA) from glutamic acid requires pyridoxal phosphate. In the case of metabolism of tryptophan, its conversion to nicotinic acid does not take place in pyridoxine deficiency. Xanthurenic acid, however, appears in increased amounts in urine in such cases. Pyridoxal phosphate also acts as co-decarboxylase for decarboxylation of certain amino acids and also as a coenzyme for desulhydration and transulphuration of reactions in cysteine metabolism. Dehydrases (serine and threonine) also require this vitamin. Pyridoxal phosphate is a component of phosphorylases.

There is a strong relationship between unsaturated fatty acids and pyridoxine. The symptoms seen in essential fatty acid deficiency resemble those seen in pyridoxine deficiency, and one has a sparing action on the other. It is thought that pyridoxine can help in the utilisation of the unsaturated fatty acids with great economy.

*Effects of deficiency* In many species, pyridoxine deficiency causes a hypochromic microcytic anemia and retardation of growth.

In humans pyridoxine deficiency gives rise to certain symptoms gradually. The first effect is a fall in hemoglobin level and anemia, followed by lethargy, depression and mental confusion. Unlike adults, the deficiency symptoms appear quickly in infants, when pyridoxine is lacking in their milk. General irritability, vomiting, diarrhea and convulsions are observed. In children B<sub>6</sub> deficiency leads to predominantly neurological symptoms and convulsive seizures but anemia is not very marked. But in adults the main symptoms are those of anemia. This may be due to influence of B<sub>6</sub> dependent coenzyme on the apoenzyme of the decarboxylase as age advances.

The symptoms of pyridoxine deficiency have been observed in patients receiving isonicotinic acid hydrazide (INH) in the treatment of tuberculosis. This is now explained to be due to the antagonistic action of INH to pyridoxine, as they are structural analogues. A slow removal of INH by the liver in some individuals (slow acetylators) results in a chemical reaction of INH with pyridoxal derivatives and their depletion. Pellagra is a frequent accompaniment of pyridoxine deficiency.

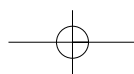
*Requirements* The adult requirement of this vitamin is between 2–3 mg per day and in the case of pregnant women 6–7 mg per day.

Some antivitamins of pyridoxine are deoxy pyridoxine and isoniazid.

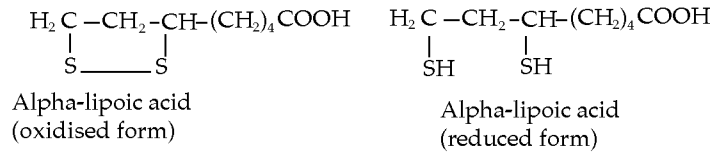
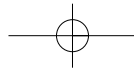
*Estimation* is done by bioassay using rat and microbiological assay, using *Sacharomyces carlsbergensis* (*L. casei* for pyridoxal only).

### **Pantothenic acid**

*Chemistry* Pantothenic (Greek, everywhere) acid is a yellow viscous oil; it is destroyed by heat in dry conditions but not in moist conditions, unlike thiamine. Oxidising and reducing agents do not destroy this vitamin. However, it is unstable in the presence of strong alkalis and acids. Pantothenic acid can be considered a compound of pantoic acid with beta alanine connected by a peptide linkage.







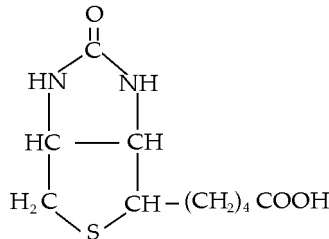
*Lipoic acid – oxidised and reduced forms*

**Biochemical function** Attempts to produce lipoic acid deficiency in animals have not been successful. However, many of the physiological functions of lipoic acid are known, especially those concerned with the oxidative decarboxylation of pyruvate and alpha ketoglutarate, producing acetyl CoA and succinyl CoA, respectively. The reactions take place in conjunction with TPP.

### **Biotin (anti-egg white injury factor)**

Biotin was first recognised as an essential growth factor for microorganisms. Later on, it was observed that experimental biotin deficiency could be produced by feeding raw egg white, which contains avidin as an antagonist or antivitamin to biotin.

**Chemistry** Biotin is a heat stable, crystalline substance soluble in water and ethyl alcohol. It contains a sulphur atom in its molecule. However, if the sulphur is replaced by oxygen, as in oxybiotin, it is active in curing biotin deficiency. Biotin occurs both in a free and in a combined state in natural foods. In combination with lysine it may occur as biocytin.

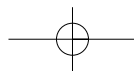


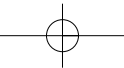
*Biotin*

**Sources** Biotin is widely distributed in both animal and plant sources. Animal foods like liver, kidneys and milk are very good sources of this vitamin. Vegetables contain moderate amounts, tomatoes and yeast being good sources of this vitamin. A large amount of biotin is supplied by intestinal bacteria.

**Biochemical functions** The multi subunit enzymes which are involved in carboxylation reactions include pyruvate carboxylase and acetyl CoA carboxylase. Biotin is linked to the epsilon-NH<sub>2</sub> of lysine of the apoenzyme. CO<sub>2</sub> is then added to the biotin nitrogen of the enzyme as a whole. The functions of biotin in the form of its coenzyme, N-carboxy biotin, in various carboxylation reactions have been established. Carbon dioxide fixation results in the biosynthesis of compounds incorporating one more carbon atom and is one of the first steps in the synthesis of fatty acids by the extramitochondrial system. The N-carboxybiotin enzyme complex transfers its 'active' CO<sub>2</sub> to compounds like acetyl CoA to give malonyl CoA. The biotin enzyme plays an active role in the formation of oxaloacetate from pyruvate and CO<sub>2</sub>.

**Effects of deficiency** When rats are fed on a diet containing raw egg-white in large quantities, characteristic symptoms like dermatitis and nervous manifestations start appearing. From regions around





the eyes, the hair falls in a fashion resulting in the 'spectacled-eye syndrome'. All these symptoms can be avoided by supplementing the diet with biotin or by feeding cooked egg-white. The egg-injury factor is now identified as a heat labile protein known as avidin and is believed to exert its effect by combining with biotin and including deficiency of the latter by hampering absorption.

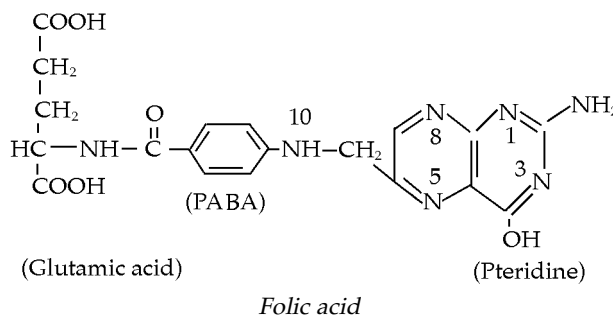
In human volunteers, biotin deficiency resulted in symptoms like scaly desquamation of skin, muscular pain, loss of appetite, anemia and lethargy.

Children develop dermatitis, alopecia, and loss of muscular control, and have retarded growth in biotin deficiency.

**Requirements** The biotin requirement of humans is uncertain, although an intake of 150–300  $\mu$ g per day has been suggested. The intestinal biosynthesis of biotin by microflora contributes to some extent to the requirement of this vitamin. Even in deficient states, more excretion of biotin occurs in urine than what can be accounted for by the dietary content.

### Folic acid (Pteroyl glutamic acid)

**Chemistry** Folic acid can be considered as consisting of three important portions: the pteridine nucleus, p-amino-benzoic acid (PABA) and glutamic acid. The number of glutamic acid residues may be one, three or seven, in different natural forms of folic acid. In plants, it is one of seven glutamates while liver was shown to contain penta glutamate.

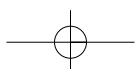


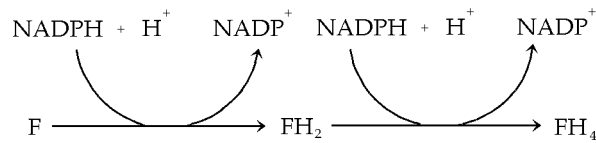
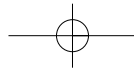
The simplest one is pteroyl monoglutamic acid (PGA) shown to be necessary for the growth of *Lactobacillus casei*. It is the monoglutamate that can be absorbed. Intestinal enzymes hydrolyse polyglutamate to monoglutamate. A major portion is reduced to tetra hydrofolate and methylated as N<sup>5</sup> methyl FH<sub>4</sub> within the intestinal cell as an integral part of the absorption process. Folate absorption is impaired in idiopathic steatorrhea, tropical sprue and in various other diseases of the small intestines.

In blood, approximately two-thirds of folate is bound to proteins.

**Sources** Folic acid is widely distributed in nature and is abundantly present in the green foliage of plants (hence the name 'folic'). It is a yellow substance, slightly soluble in water. Liver, kidneys, yeast, cauliflower and cabbage are the best sources.

**Biochemical functions** Folic acid functions in the body as folate coenzymes. Folic acid is converted into tetra hydrofolic acid with the aid of NADPH catalysed by folate reductase. Folic acid antagonists like amethopterin (methotrexate) block the reduction of FH<sub>2</sub> to FH<sub>4</sub> at the dihydrofolate reductase step. (Hence they are used for producing remissions in leukemias.)





*Reduction of folic acid to dihydro and tetrahydro folic acids*

The tetrahydro folic acid then acts as the one carbon acceptor and forms derivatives like formyl tetrahydro folic acid. The one carbon moiety may be —CHO (formyl), —CH<sub>2</sub> OH (hydroxy methyl), formate (H—COO—), —HC \ NH (formimino) or methyl (—CH<sub>3</sub>). The complexes are interconvertible due to a NADP<sup>+</sup> dependent hydroxy methyl dehydrogenase system and thus any of these groups can be added to or removed from compounds, in the various reactions involved in one carbon metabolism. N<sup>5</sup> formyl FH<sub>4</sub> is called folinic acid. N<sup>5</sup> methyl FH<sub>4</sub> is the major form of folate derivative in blood. Folic acid has its biochemical role in the metabolism of glycine, serine, glutamic acid, histidine, betaine and choline. It is also involved in several biosynthetic processes, in the incorporation of formyl carbon into the purine skeleton and for the synthesis of thymine. Folic acid is required for the formation of N-formyl methionine required to initiate biosynthesis of proteins in lower organisms. In fact, the macrocytic anemia in folic acid deficiency may be attributed to the decreased formation of new red blood cells due to interference with purine and thymine synthesis.

*Effect of deficiency* The deficiency of this vitamin results in macrocytic anemia. Many cases of macrocytic anemia in infants, nutritional macrocytic anemia in adults with megaloblastic marrow, have all been effectively cured by treatment with folic acid. It also has a favourable effect on hematopoiesis in pernicious anemia having the same effect on blood formation as vitamin B<sub>12</sub>. But, far less vitamin B<sub>12</sub> is needed than folic acid. While a small dose of 300–500 µg of folate per day evokes a positive hematological response in folate deficiency anemia, there is no response in pernicious anemia with such a small dose.

*Requirement* A hematologic response is seen in anemic patients (suffering from folic acid deficiency) with a dose of 300–500 µg of this vitamin daily. The effect is more marked with a lower dose in the presence of vitamin B<sub>12</sub>. For a normal adult, a daily intake of 500 µg is available in the ordinary diet. In case of macrocytic anemias, a daily dose of 200 mg of this vitamin by mouth or 10–20 mg intravenously is given.

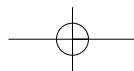
Some of the antivitamins are aminopterin and amethopterin (methotrexate).

*Estimation* is done by microbiological assay using *Streptococcus lactus* and estimation of 'figlu' (formimino glutamic acid) in urine. In folic acid deficiency there is increased excretion of 'figlu' in urine on a loading dose of histidine.

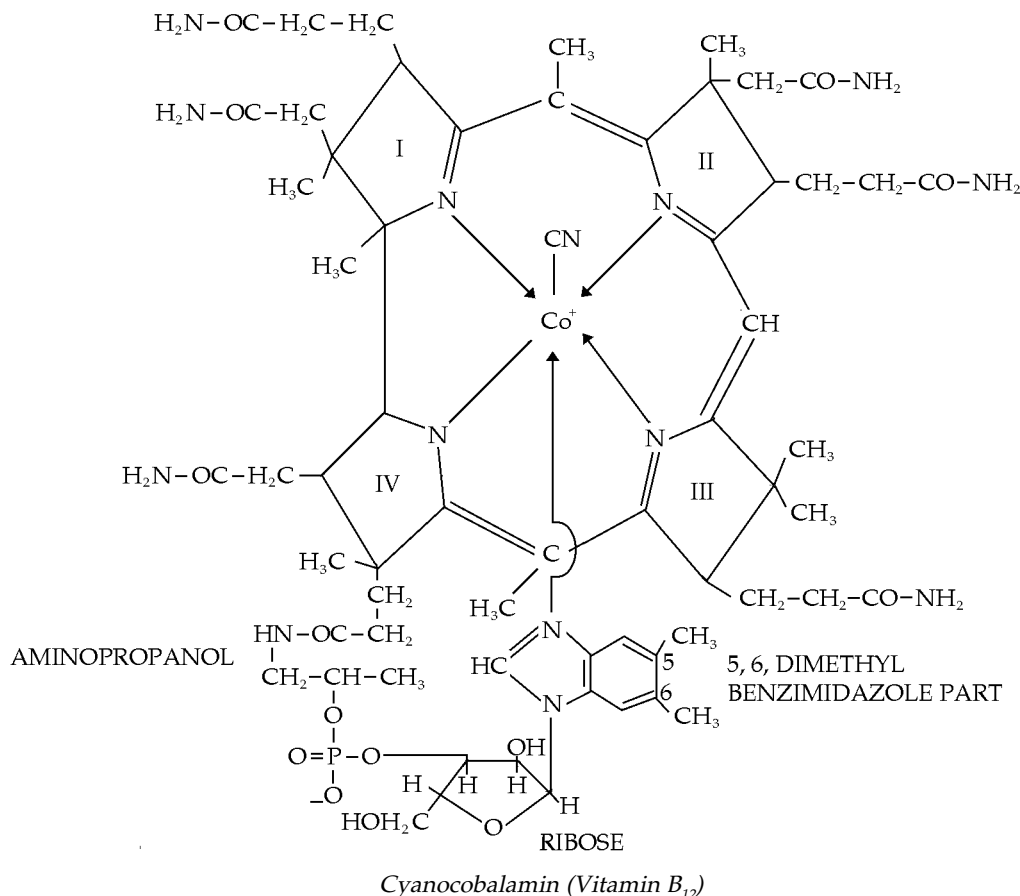
### **Cobalamins, Vitamin B<sub>12</sub> (Cyanocobalamin)**

*Chemistry* Vitamin B<sub>12</sub> or the antipernicious anemia factor (extrinsic factor of Castle) is a red crystalline compound containing cobalt and phosphorus.

The central portion of the molecule consists of the corrin ring system consisting of four pyrrole rings surrounding a cobalt atom. The pyrrole rings are connected to each other through methene bridges at three places only while the rings I and IV are directly connected. A 5,6-dimethyl benzimidazole moiety is connected to the cobalt atom of the corrin ring on one side and to the ribose moiety at the other. The ribose in turn is connected to one of the pyrrole rings through aminopropanol and phosphate moieties. A cyanide group is coordinately linked to the cobalt atom in cyanocobalamin, but may be replaced by —NO<sub>2</sub> or —OH groups in nitro cobalamin and hydroxy cobalamin

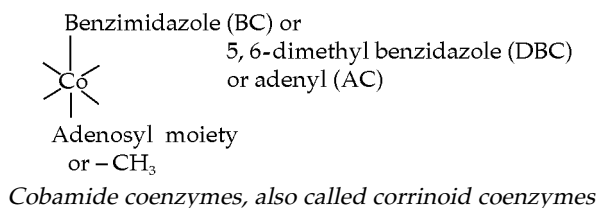


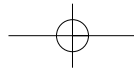
respectively. Hydroxycobalamin has therapeutic use, as it binds strongly with plasma proteins and is retained longer in the system.



Vitamin B<sub>12</sub> is cobalamin containing cyanide, but in its coenzyme forms the cyanide is substituted by adenosyl moiety or CH<sub>3</sub> group. So far, four cobamide coenzymes are known. The coenzymes are 1) 5,6-dimethyl benzimidazole cobamide (DBC), 2) benzimidazole cobamide (BC), 3) adenyl cobamide (AC), and 4) a coenzyme in which methyl group is attached to cobalt instead of the adenosyl moiety. These coenzymes do not contain the cyanide group and are called corrinoid coenzymes.

Cobalamins are water soluble, heat stable and stable in the presence of dilute acids at pH 4.0.





*Sources* Vitamin B<sub>12</sub> is almost absent in plant material. It is produced by the Streptomyces group of organisms and is a by-product in the manufacture of Streptomycin. Animal tissues are good sources of this vitamin. Egg yolk and milk contain adequate amounts.

*Biochemical function* Cobalamin-binding proteins, known collectively as 'R-proteins', are secreted by the salivary glands and the stomach and bind the cobalamins at the acid pH. The 'R-proteins' are normally degraded by pancreatic proteases. Subsequently, the cobalamins bind to the intrinsic factor of Castle. Therefore, the absorption of vitamin B<sub>12</sub> by the ileum requires previous release of the bound vitamin B<sub>12</sub> in the stomach in the presence of HCl. A low gastric acidity interferes with the availability of B<sub>12</sub> for absorption. In pancreatic insufficiency, cobalamin molecules are not released from the 'R-proteins'.

The intrinsic factor of Castle is a low molecular weight specific glycoprotein and is not protease-sensitive. It is secreted by the parietal cells of the gastric mucosa of the cardia and the fundus of the stomach. After release from the R-proteins, the cobalamins bind to the intrinsic factor. The complex crosses the ileal mucosa. The intrinsic factor is released and the vitamin is transferred to a plasma transport protein, transcobalamin II. In addition there is transcobalamin I which exists in the liver and plasma. This protein functions as the storage form of cobalamin, a unique situation for the storage of water-soluble vitamin.

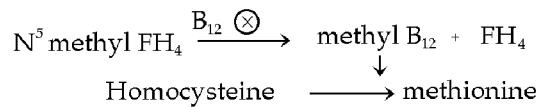
The major circulating vitamin is methyl cobalamin. Cobalamin binds to the receptors on the plasma membrane and gets internalised. Once inside, it is converted to hydroxycobalamin and then methyl cobalamin.

In the absence of the intrinsic factor, there is no absorption of vitamin B<sub>12</sub>. Hence, very often, B<sub>12</sub> is parenterally administered for treatment.

B<sub>12</sub> functions as cobamide coenzyme and is involved in many biochemical processes. One of the reactions catalysed in the presence of B<sub>12</sub> coenzyme is the enzymatic conversion of methyl malonyl CoA to succinyl CoA in animal tissues. The enzyme is L-methyl malonyl CoA mutase and the coenzyme, 5-deoxy adenosyl cobalamin. The reaction takes place in the mitochondria. Methyl malonic acid appears in excess in the urine of patients with pernicious anemia but disappears on treatment with B<sub>12</sub>. The neurological symptoms of pernicious anemia due to B<sub>12</sub> deficiency are attributed to the accumulation of methyl malonic acid.

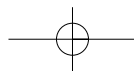
In addition, the neurological manifestations are also due to a deficiency of methionine required for transmethylation for the metabolism in the myelin sheath and deranged fatty acid metabolism, chiefly propionic acid metabolism as B<sub>12</sub> is required for the conversion of propionic acid to succinic acid. As homocysteine is not effectively converted to methionine for want of methyl B<sub>12</sub>, there is deficiency of methionine and excretion of taurine. Taurine is a metabolite of homocysteine.

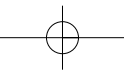
The other important reaction of vitamin B<sub>12</sub> is to release the folate trap and the methylation of homocysteine to methionine. This takes place in the cytosol. Only vitamin B<sub>12</sub> can convert N<sup>5</sup>-methyl FH<sub>4</sub> to FH<sub>4</sub> and in this process, methyl B<sub>12</sub> is formed.



⊗ Methyl transferase apoenzyme binds cobalamin and N<sup>5</sup>-methyl FH<sub>4</sub>, and effects the transfer of the methyl group.

*Release of folate trapped as N<sup>5</sup> methyl FH<sub>4</sub> and methylation of homocysteine*





In  $B_{12}$  deficiency, the above reactions do not take place, and folate is permanently trapped as  $N^5$ -methyl  $FH_4$  (folate trap) and is therefore not available for one carbon transfers. This would result in diminished synthesis of thymidylate and DNA, hence, the macrocytic megaloblastic anemia in  $B_{12}$  deficiency. It is not a direct manifestation of  $B_{12}$  deficiency but an accompanying deficiency of folate. The hematological manifestations of pernicious anemia are thus not the primary effect of  $B_{12}$  but of secondary deficiency of  $FH_4$  and therefore cured with folate. The neurological manifestations can, however be cured only with  $B_{12}$ . Vitamin  $B_{12}$  is required to convert D-ribonucleotides to deoxy D-ribonucleotides functioning in a specific DBC dependent ribonucleotide reductase in prokaryotes. Thus it helps in the formation of DNA, and, thereby, proteins.

The mechanism involved in erythropoiesis and increase in the number of erythrocytes after vitamin  $B_{12}$  therapy is believed to be due to its effect on the DNA synthesis by releasing the folate from the folate trap. The megaloblastic and macrocytic anemia seen in  $B_{12}$  deficiency disappears on treatment with this vitamin.

*Effects of deficiency* Vitamin  $B_{12}$  is the antipernicious anemia factor and is found to dramatically cure the hematological defects (megaloblastic and macrocytic anemia) and the neurological manifestations of pernicious anemia. It is recognised as the extrinsic factor of Castle required for hemopoiesis. Manifestations of  $B_{12}$  deficiency include loss of appetite and failure of growth, macrocytic and megaloblastic anemia, pernicious anemia (involving sub-acute combined degeneration of the cord) and deranged metabolism in nervous tissue. In most cases, the deficiency of  $B_{12}$  is due to deficiency of the intrinsic factor rather than the vitamin itself.

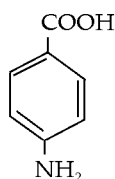
*Requirements* The average requirement for adults is 1–2  $\mu\text{g}$  of vitamin  $B_{12}$  per day. Vegetarians do not get the required amount through their diet, unless they include milk in it. Some of the fermented foods contain vitamin  $B_{12}$  in adequate amounts. Natural drinking water from the rivers and wells may contain the required amounts of this vitamin.

*Estimation* is by microbiological assay using *L. lactis*.

### Para-aminobenzoic acid

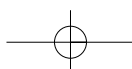
It was observed that a diet containing all the known vitamins like thiamine, riboflavin, niacin, pyridoxine and pantothenic acid was still lacking in a factor concerned with lactation in rats. The hair of black rats on such a diet turned grey and this factor was called 'anti-grey hair' factor.

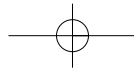
This is now identified as para-aminobenzoic acid. This compound is already present as a constituent part of folic acid and is believed to be necessary for the biosynthesis of folic acid by microor-



*Para-aminobenzoic acid*

ganisms in the intestines. PABA by itself has been recognised as a growth factor in rats. PABA is a structural analogue of sulphanilamide and the bacteriostatic effect of the sulpha drug is explained



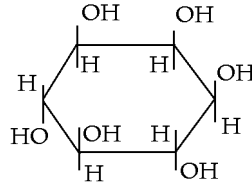


to be due to its inhibition (competitive inhibition) of the phenolase system which is required for the life of certain microorganisms.

Some authors do not consider PABA a vitamin although it is a substance which cannot be biosynthesised in the human body. Perhaps there is no need for this compound in the body except for the intestinal flora.

### Inositol

*Chemistry* This compound exists in the natural form as mesoinositol (myoinositol) which is biologically effective as a growth factor. There are nine stereo-isomers of which the biologically active form is the optically inactive form.



*myoinositol*

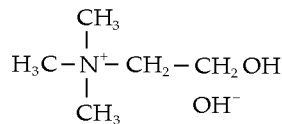
*Biochemical functions* It acts as a lipotropic agent along with choline in experimental animals. It converts neutral triacyl glycerols and phosphatidic acids to inositol phosphatides (lipositols). Some hormones use the latter as their second messengers to release  $\text{Ca}^{2+}$ . Deficiency of inositol results in alopecia and failure of growth. Inositol is required for the formation of inositol phospholipids in the brain. Inositol probably forms a complex with tocopherols needed for the proper storage of creatine in the muscle. It is widely distributed in most natural foodstuffs of plant and animal origin. Yeast, milk, nuts and fruits are the best sources.

Phytic acid is inositol hexaphosphoric acid. The calcium and magnesium phytins are present in corn.

### Choline

*Chemistry* Choline is trimethyl-hydroxyethyl-ammonium hydroxide. It is synthesised in the body from glycine, and is related to one carbon metabolism and folic acid. Some authors do not include choline under vitamins as it could be synthesised by the human system from serine through ethanolamine in required amounts.

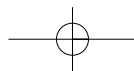
*Sources* Meat, egg, yolk, bread, cereals, beans and peanuts are good sources.



Choline  
(Trimethyl N-ethanolamine hydroxide)

*Biochemical functions* Choline as a lipotropic substance: Choline is a constituent of various phospholipids like the lecithins and sphingomyelins which have important physiological functions of the body. Choline helps in the formation of phospholipids in the liver, and thereby the disposal of triacyl glycerols as lipoprotein complexes, and preventing of fatty infiltration of liver. It is also reported to enhance oxidation of fatty acids in the liver. For these reasons choline is a potent lipotropic substance.

Choline after being oxidised to betaine is a methylating agent (see chapter 14).



Choline is also required for the formation of acetyl choline, one of the chemical mediations of nerve activity.

*Effects of deficiency* Choline deficiency results in fatty infiltration into the liver. On a low choline diet, puppies develop lack of appetite, failure of growth and fatty liver. Rats maintained on low choline diet develop fatty liver, cirrhosis of the liver and hemorrhages in the kidneys and eyes. In chicks and turkeys, choline deficiency causes perosis or slipped tendon disease.

*Requirement* The human requirement of choline has not been established. The presence of adequate amounts of methionine in the diet has a sparing action on the exogenous requirement of choline.

### Vitamin P

The colouring pigments in plants like rutin and hesperidin are referred to as vitamin P. They are bioflavonoids. They influence capillary permeability and potentiate vitamin C. They are present in orange and lemon peel.

### Antivitamins (Vitamin antagonists)

Antivitamins are synthetic compounds which are either structural analogues of the vitamins (in many cases having opposite effects physiologically) or compounds which increase the requirement of a vitamin thereby inducing an artificial deficiency. Pyriothiamine and oxythiamine are antagonists to thiamine while isoriboflavin is the antivitamin for riboflavin. Niacin is antagonised by pyridine sulphonic acid and pantothenic acid by pantooyltaurine. Isonicotinic hydrazide (Isoniazid), a potent drug used in the treatment of tuberculosis, is antagonistic to pyridoxine and overdosage of this drug produces convulsive episodes characteristic of pyridoxine deficiency. Aminopterin and amethopterin (methotrexate) are antivitamins for folic acid are used in the treatment of cancer. The sulphur drugs antagonise para-aminobenzoic acid and folic acid. Vitamin K is antagonised by dicoumarol.

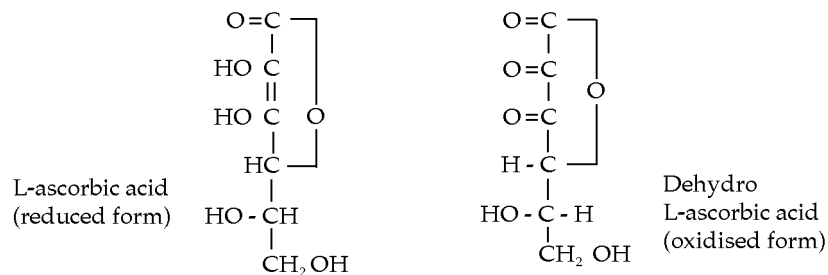
Most of these antivitamins functions by inhibiting the enzyme systems requiring the vitamin as a coenzyme, this being a type of competitive inhibition. Such inhibition can be overcome by using higher doses of the vitamin.

### Vitamin C (L-ascorbic acid)

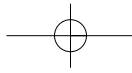
The water soluble vitamins also include vitamin C, the antiscorbutic vitamin.

*Chemistry* L-ascorbic acid is related chemically to glucose and glucuronic acid. It is highly soluble in water, has a sour taste and is easily oxidised in aqueous solution by atmospheric oxygen and oxidising agents. The oxidation is enhanced in the presence of traces of copper. It is the least stable of all the water-soluble vitamins. Alkalis completely decompose it, while in acidic solution it is quite stable. Metaphosphoric acid and trichloro-acetic acid act as negative catalysts and depress the oxidative decomposition of ascorbic acid in vitro.

On oxidation under mild conditions, ascorbic acid is converted to dehydro-ascorbic acid.



L-ascorbic acid – reduced and oxidised forms



*Sources* Ascorbic acid is distributed widely in many plant and animal foods. Fruits and vegetables are excellent sources of this vitamin. Citrus fruits, guava, gooseberry and green peppers are the richest sources, while cabbage and spinach are reasonably good sources. Among animal tissues, the highest amounts are present in the adrenal cortex, gonads and glandular organs. Cow's milk does not contain appreciable amounts and has no value as an antiscorbutic food.

*Biochemical functions* Ascorbic acid has been recognised as an important substance in the body involved in the redox mechanisms. In the presence of glutathione, the ascorbic acid can be converted from its oxidised form to the reduced form easily and is believed to play a role in many of the respiratory processes. Cytochrome C, pyridine and flavin nucleotides may need vitamin C.

The presence of a high concentration of ascorbic acid in the gonads and adrenal cortex shows that it plays a role in the production of the steroid hormones from cholesterol, by activating certain enzyme systems involved in the process. A biochemical reaction to stress is depletion of ascorbic acid from the adrenal cortex.

The formation of bile pigments from the breakdown of hemoglobin also requires vitamin C.

The metabolism of phenyl alanine and tyrosine requires vitamin C. The excretion of certain phenyl pyruvic acid derivatives in infants is abolished by vitamin C therapy.

The absorption of iron from the intestines is facilitated by vitamin C which converts the ferric form to the ferrous form. Interconversion of ferrous to ferric forms later on in the tissues may also be helped by the redox systems in the body and vitamin C may play a role in that. The mobilisation of iron from the bones is also augmented.

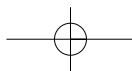
Vitamin C is involved in the formation of the intercellular cementing substances and collagen. In its deficiency, although the organic matrix and inorganic calcium phosphate are available, the cementing substance is lacking and this leads to bone disorders. This also affects the formation of dentine and is needed for the maintenance of teeth. Collagen contains hydroxy proline to the extent of 12 per cent which is formed from the amino acid proline. It also contains hydroxy lysine. The conversion of proline to hydroxy proline and lysine to hydroxy lysine requires vitamin C. In the absence of this vitamin, an abnormal collagen is produced and causes lesions like spongy gums, hemorrhages, fracture of bones, faulty cartilage formation leading to pain in joints, etc.

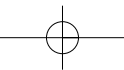
The major excretory products of vitamin C are ascorbic acid itself and dehydro-ascorbic acid with a small amount of oxalic acid.

*Effects of deficiency* Ascorbic acid is biosynthesised in many animal species except in humans, monkeys and guinea-pigs. When guinea pigs are maintained on a scorbutic diet, growth ceases in two weeks and symptoms start appearing. The joints become tender, and swollen and the animal lies on its back with its legs kept sprawled (scurvy position). It winces when pressed because of pain in the joints due to subcutaneous and subperiosteal hemorrhages. The gums become tender and show hemorrhages. The teeth and nails become loose. In humans, ascorbic acid deficiency may occur due to continued sub-normal intake of this vitamin.

Decreased resistance to infection, slow healing of wound and union of fractures, hemorrhages inside the muscles and under the skin (petechial hemorrhages) occur as the deficiency progresses. Subcutaneous bleeding results in the formation of red patches under the skin; many endocrine functions become sluggish on a low vitamin C intake especially in the gonads and adrenal cortex.

Deficiency of vitamin C causes a reduction in the amount of intercellular substance and weakness in the endothelial wall of the capillaries. In scurvy, due to the deficiency of vitamin C, there is anemia, pains at the joints and hemorrhages from the mucous membranes of the mouth and gastrointestinal tract. There is swelling and bleeding of gums with ulceration and even gangrene.





There are no known toxic effects of vitamin C. Potential complications of chronic massive overdose include calcium oxalate stones and detrimental effects of vitamin C on the absorption of other vitamins like vitamin B<sub>12</sub> and some drugs.

*Clinical importance* In febrile conditions, infections and stress, vitamin C is lost heavily from the body. Hence, adequate quantities of this vitamin should be taken in such conditions.

The usefulness of this vitamin in many pathological conditions like atherosclerosis, deep vein thrombosis, diabetes mellitus, cancer, skin diseases, frostbite and in common cold and maintenance of youthfulness makes one call it the 'versatile' vitamin.

*Requirements* On an average, about 60 mg of ascorbic acid per day is required in the diet by normal adults. The requirement is increased during infection, fevers and toxic conditions. Presently some workers in this field consider that 60 mg per day is inadequate for many beneficial effects of this vitamin, and that about 1–2 g should be taken daily.

*Ascorbic acid saturation test* The determination of merely the plasma levels or urinary levels of ascorbic acid may not always be indicative of the ascorbic acid status of an individual. A satisfactory method for assessing it is the ascorbic acid saturation test. Orally or intravenously administered ascorbic acid will be taken up by the tissues which get saturated with the vitamin. The excretion of this vitamin in urine is then determined after four hours and six hours, and again the next day. On administering 700 mg of ascorbic acid in the test, at least 200 mg will be excreted in the urine within four hours in normal subjects. On the subsequent day, not less than 50 mg should be present in the urine. In ascorbic acid deficient states, less amounts are excreted. This test is useful in screening subclinical scorbutic states.

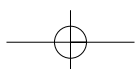
*Estimation* Estimation is done by titration with 2, 6-dichlorophenol indophenol which is reduced by this vitamin, and colorimetrically by treatment of the dehydro form with 2, 4-dinitrophenyl hydrazine.

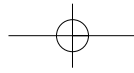
## SUMMARY

Vitamins are organic molecules not usually synthesised in the living systems and hence need to be taken in the diet. They are accessory food factors required in small quantities for metabolism, growth and warding off diseases. Water-soluble vitamins are B complex vitamins like thiamine, riboflavin, niacin, folic acid, vitamin B<sub>12</sub> as well as vitamin C. Fat soluble vitamins are A, D, E and K.

**Vitamin A** exists in the body as retinol, retinal and retinoic acid collectively called retinoids. Beta carotenes of carrots are precursors of retinol. Retinol supports normal functions of reproduction in males and females and retinal is involved in Wald's visual cycle. It is needed for the synthesis of rhodopsin. Retinoic acid is required for synthesis of glycoproteins as carrier of oligosaccharides. Deleterious manifestations of vitamin A deficiency are nyctalopia (night-blindness), xerophthalmia, keratomalacia, and drying of mucous membrane. The daily recommended dosage is 5000 IU. The Carr–Price reaction with antimony trichloride or UV spectrophotometry is employed for estimation of vit A.

**Vitamin D** occurs in fish oils in significant amounts. Its absence in diet causes rickets in children and osteomalacia in adults. Vitamin D proper has no biological effect. It should be converted to 1,25-dihydroxy cholecalciferol (calcitriol) in the liver (25 hydroxylation) and kidneys (1-hydroxylation). Calcitriol is needed for promoting cross-linking of bone collagen and increasing the synthesis of osteocalcin of the bone. Thus, it causes bone accretion (formation) of new bones. In old bones, it causes bone-demineralisation which is its intrinsic property. The required daily dose of vitamin D is 400 IU for children.





**Vitamin E** is tocopherol, good sources of which are wheat germ and cotton seed oil. All vegetable oils and animal fat contain some amount of Vitamin E. By virtue of its having phenolic hydroxyl group, Vitamin E is a good electron-sink and a powerful antioxidant. Being lipid soluble, it has the enviable role of functioning as antioxidant of membrane proteins and lipids while water soluble antioxidants like vitamin C do not have such a privilege. However, the vitamin E free radical formed when vitamin E acts as an antioxidant is quenched by vitamin C. In Vitamin E deficiency, erythrocytes become fragile. Daily requirement is 15–30 mg.

**Vitamin K:** Compounds which have vitamin K<sub>2</sub> activity are naphthoquinone derivatives. Vitamin K<sub>3</sub> is menadione. Green leafy vegetables and spinach have this vitamin. Vitamin K is very necessary for normal blood clotting through its role in Gamma carboxylation and formation of GLA residues in clotting factors prothrombin, VII, IX and X. In its deficiency, there will be a tendency to bleed profusely. Dicoumarol, an antivitamin to K prolongs clotting time. The daily requirement of vitamin K is about 2 mg.

**Thiamine (B<sub>1</sub>)** is a water soluble vitamin and is thermolabile. Its main sources are rice and wheat bran. As thiamine pyrophosphate (TPP), it functions as coenzyme for oxidative decarboxylation of ketoacids like pyruvate. It is also part of transaldolase and transketolase. Dietary deficiency of B<sub>1</sub> causes loss of appetite (anorexia) and gradual loss of weight. Neuronal function is disturbed, leading to polyneuritis and beriberi. The daily dose is 1–2 mg.

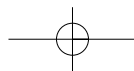
**Riboflavin (B<sub>2</sub>)** is a yellow water-soluble vitamin, heat-stable but photolabile. Milk is a good source. The coenzymes of B<sub>2</sub> are FMN and FAD involved in electron-transport chain. Dietary deficiency causes cheilosis, glossitis, inflammation of cornea and redness of the conjunctiva. Angular stomatitis and angular conjunctivitis are two glaring manifestations of ariboflavinosis. Daily dosage is 1.6–2 mg.

**Niacin and niacinamide (B<sub>3</sub>)** is a water-soluble vitamin occurring in significant amounts in fish, yeast, beans and peanuts. Coenzymes of niacin are NAD<sup>1</sup> and NADP<sup>1</sup> and their reduced products NADH and NADPH. NAD<sup>1</sup> is involved in electron transport chain. Deficiency of niacin causes the disease pellagra (rough skin) characterised by bronzing and thickening of the skin leading to inflammation. There will be diarrhea, dementia and dermatitis (3 'ds'). The daily dosage is 17–20 mg. Hypervitaminosis leads to flushing due to vasodilatation. Blood pressure is reduced. Patients with low blood pressure should not take niacin indiscriminately. Niacin brings down blood cholesterol and is beneficial in atherosclerosis.

**Pyridoxin (B<sub>6</sub>)** is a water-soluble vitamin. Egg yolk, milk, fish and meat are good sources. Its coenzyme forms are pyridoxal phosphate and pyridoxamine phosphate. They are essential for transamination and decarboxylation reactions. Synthesis of several important biochemicals like heme, GABA and conversion of tryptophan to niacin require B<sub>6</sub>. B<sub>6</sub> improves neuronal function. Deficiency in children causes convulsion and hypochromic microcytic anemia in adults. Tuberculosis patients receiving isonicotinic acid hydrazide (INH) may develop B<sub>6</sub> deficiency. With B<sub>1</sub> and B<sub>12</sub>, B<sub>6</sub> is prescribed as neurobion for the improvement of nerve function. The daily requirement is 2–3 mg.

**Pantothenic acid** is an yellow viscous oil unstable in presence of acids and alkalis. The best sources are eggs and milk. Coenzyme form is CoASH, needed for carbohydrate and lipid metabolisms – TCA cycle, de novo lipogenesis and beta oxidation. As succinyl CoA, it is required to convert aceto acetate to acetyl CoA in extra hepatic tissues for getting energy. Acyl carrier protein required for extra mitochondrial lipogenesis contains pantothenic acid moiety. CoASH with carnitine is required for translocation of fatty acids from crystal into the matrix of mitochondria. As pantothenic acid is universally present in all types of food stuffs, deficiency of this vitamin is rare. The daily dose is about 5 mg.

**Biotin** is water-soluble sulphur-containing vitamin. Milk and tomatoes contain this vitamin. The protein avidin in raw egg is an antivitamin of biotin. Biotin is needed for fixation of carbon dioxide in carboxylation reactions. Biotin is linked to epsilon amino group of lysine of the apoenzyme. CO<sub>2</sub> is



then added to biotin nitrogen in the biotin enzyme complex. Biotin deficiency causes scaly desquamation of skin, muscular pain and loss of appetite. Daily requirement is 150–300 µg.

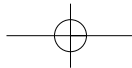
**Folic acid** is a yellow water-soluble substance. Yeast, cauliflower and cabbage are the best sources. The coenzyme of folic acid is tetrahydro folate (FH<sub>4</sub>). It participates extensively in one carbon metabolism. One carbon moieties received and transferred by FH<sub>4</sub> are formyl, hydroxy methyl, methylene, methenyl and formimino groups. Under special circumstances, methyl group also is transferred, as for example N<sup>5</sup> methyl FH<sub>4</sub> reacting with B<sub>12</sub> to form methyl B<sub>12</sub>. By decreasing the bioavailability of folate, synthesis of thymidylate and thereby DNA will decrease. This is the basis for use of amethopterin (methotrexate) and ameenopterin for producing remissions in leukemias as they inhibit the action of dihydrofolate reductase in the formation of FH<sub>4</sub>. A deficiency of folate leads to deficiency of DNA and cells. Thus macrocytic anemia with megaloblastic marrow is encountered in folate deficiency. Folate is usually given with B<sub>12</sub> because the pernicious anemia of B<sub>12</sub> deficiency is due to secondary deficiency of folate from folate trap. The hematological pathology is exactly the same in folate as well as B<sub>12</sub> deficiency. Daily requirement is 500 µg, while in macrocytic anemia, a daily dose of 200 mg is recommended.

**Vitamin B<sub>12</sub>** is a red crystalline water-soluble compound. As it has a cyanide group coordinately linked to cobalt, it is called cyanocobalamin. Hydroxycobalamin in which CN is replaced by OH has greater therapeutic value. The coenzyme for B<sub>12</sub> is called cobamide coenzyme and corrinoid coenzyme. Dietary vitamin B<sub>12</sub>, the extrinsic factor of Castle binds to 'R' proteins at acid pH. Pancreatic proteases digest 'R' proteins and the vitamin is taken up by the intrinsic factor of Castle, a glycoprotein necessary for intestinal absorption of B<sub>12</sub>. B<sub>12</sub> is needed for the formation of methyl B<sub>12</sub> from N<sup>5</sup> methyl FH<sub>4</sub>. In the absence of B<sub>12</sub>, folate is 'trapped' as N<sup>5</sup> methyl FH<sub>4</sub> and not available for metabolism. So a primary deficiency of B<sub>12</sub> will cause a secondary deficiency of folate. Therefore in pernicious anemia, the hematological pathology is the same as in the deficiency of folate, that is, macrocytic anemia with megaloblastic marrow. B<sub>12</sub> coenzyme is also required in propionic and metabolism to convert methyl malonyl CoA to succinyl CoA. In pernicious anemia due to B<sub>12</sub> deficiency, methyl malonic acid accumulates in blood and urine.

Methyl malonic acid is toxic to the brain. As homocysteine is not converted to methionine for want of methyl B<sub>12</sub>, a deficiency of methionine sets in. This affects transmethylation in myelin sheath. There are neurological manifestations in pernicious anemia, which appear later. One should not be duped by early hematological manifestations and treat pernicious anemia by folate alone. No doubt there will be correction of altered blood picture, but the underlying neurological defects might not get corrected and will have serious consequences. It is always preferable to give folate and B<sub>12</sub> for macrocytic megaloblastic anemia and B<sub>1</sub>, B<sub>6</sub> and B<sub>12</sub> (neurobion) for neurological disorders.

DBC cobamide is needed for the activity of ribonucleotide reductase for the conversion of D-ribonucleotides to deoxy D-ribonucleotides for DNA synthesis in prokaryotes. The average daily requirement is 1–2 µg per day. Milk is the only vegetarian source.

**Vitamin C** is a water-soluble vitamin destroyed by oxidising substances and is easily oxidised to dehydro L-ascorbic acid. It is a good antioxidant and rehabilitates vitamin E free radical. It reduces ferric to ferrous required for intestinal absorption. Proline and lysine incorporated in collagen are hydroxylated with the help of vitamin C. Deficiency leads to scurvy, decreased resistance to infection, slow healing of wounds, and delayed union of fractures, hemorrhages inside the muscle and under the skin, bleeding of the gums and ulcers in the mouth. Though the recommended dose is only 60 mg per day, a mega dose of 1–2 g a day is beneficial in atherosclerosis, arteriosclerosis, deep vein thrombosis, frostbite, diabetes mellitus, cancer and common cold and helps to keep people young by maintaining intercellular integrity through its role in collagen metabolism.

**QUESTIONS**

- What are the chemistry, physiological functions, biochemical effects, deficiency manifestations and daily dosage of vitamin A?
- What are the chemistry, physiological functions, biochemical effects, deficiency manifestations and daily dosage of vitamin B<sub>1</sub>?
- What are the chemistry, physiological functions, biochemical effects, deficiency manifestations and daily dosage of vitamin B<sub>2</sub>?
- What are the chemistry, physiological functions, biochemical effects, deficiency manifestations and daily dosage of Niacin?
- What are the chemistry, physiological functions, biochemical effects, deficiency manifestations and daily dosage of vitamin C?
- Write notes on Wald's visual cycle.
- Vitamin D can be considered a hormone rather than a vitamin. Explain.
- Write notes on (i) antioxidant role of vitamin E (ii) GLA residues.
- What are the coenzyme forms of thiamine, riboflavin, niacin, pantothenic acid, pyridoxin, folic acid and cyanocobalamin? List the functions.
- Name the deficiency diseases of B<sub>1</sub>, niacin, pyridoxin, vitamin D, folate, B<sub>12</sub> and L-ascorbic acid. Explain the biochemical cause for the same.
- Name three B-complex vitamins required for neuronal function and their mode of action in the brain.
- Write notes on angular stomatitis, pellagra, beriberi, nyctalopia, keratomalacia, scurvy, xerophthalmia, renal rickets.
- Write notes on INH and slow acetylators, Gopalan's syndrome, amethopterin, DBC (cobamide), methyl cobamide and dicoumarol.
- Why should folic acid and B<sub>12</sub> be given for macrocytic megaloblastic anemia ?
- Explain the absorption of extrinsic factor of Castle.
- Explain the hematological and neurological manifestations of vitamin B<sub>12</sub>.
- Describe the role of vitamin C in intestinal absorption and collagen synthesis. What are the advantages of a mega dose of vitamin C in nutrition ?
- Explain the formation of one fat soluble and one water soluble vitamin in the body from cholesterol and aminoacid respectively.

