Human biochemistry

JAMES M. ORTEN, Ph.D.
Professor of Biochemistry and Director of Graduate Affairs, Wayne State University School of Medicine, Detroit, Michigan

OTTO W. NEUHAUS, Ph.D.
Professor and Chairman of Biochemistry, University of South Dakota, School of Medicine, Vermillion, South Dakota

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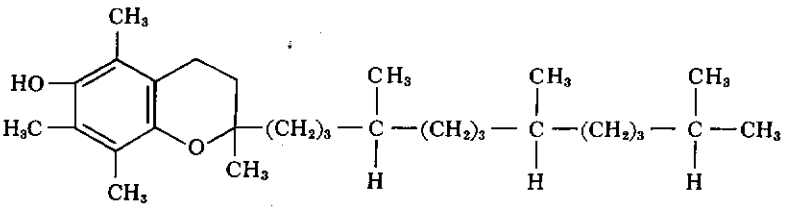
results. The threshold of toxicity seems to be about 20,000 to 25,000 units per kilogram of body weight per day. Such doses are not ordinarily employed.

The reason for the toxicity of vitamin D is the difficulty of excretion of this vitamin, rather than its storage in the liver. Any excretion is gradual, by way of the bile. Excess cholecalciferol injected into animals remains in the circulation for several months. In contrast, the *water-soluble* vitamins, if given in excess, are excreted promptly in the urine and are therefore relatively nontoxic.

α-Tocopherol (vitamin E)

The possibility that reproductivity might be dependent on a vitamin was first suggested by Mattill, in 1920; such a vitamin was discovered 2 years later by Evans and by Sure. They fed rats a diet of purified foodstuffs plus cod-liver oil and yeast. Such a diet was assumed to contain all the vitamins necessary for the rat. However, although the animals grew at a normal rate, they did not bear young. Addition of a variety of vegetable oils rectified this condition. The factor contained in these foods was termed vitamin E, the *antisterility* vitamin or fertility factor. Numerous subsequent investigations resulted in the isolation of vitamin E in crystalline form, the determination of its structure, and finally its synthesis. It was given the name tocopherol (from Greek, meaning 'child-bearing', plus *-ol*, for 'an alcohol').

Properties and structure. Vitamin E is a fat-soluble, water-insoluble, light yellow oil, stable to heat and acids, rather unstable to alkalies, that is slowly oxidized. It is found in the nonsaponifiable fraction of the vegetable oils.



α-Tocopherol

Like vitamins A and D, there is more than one form of vitamin E. We now distinguish several different tocopherols. α-Tocopherol is the most potent, has been synthesized, and is commercially available. Its structure is shown above. The other tocopherols differ in the number and position of the methyl groups attached to the benzene ring.

Occurrence. The tocopherols are widely distributed in plant and animal tissues and differ from vitamin A in not being concentrated chiefly in the liver. Particularly good sources are cottonseed oil, corn oil, peanut oil, and wheat germ oil, but not olive oil. Green lettuce leaves and orange peels also have a high content, and nearly all green-leaved plants have some of this vitamin. It is also present in meat, butter, milk, eggs, and fish-liver oils. Vitamin E activity is displayed by many other organic compounds, some of them quite unrelated structurally to the vitamin. A number of phenols, quinones, coumarins, etc. show some vitamin E action. Slight changes in the structure of the active tocopherols, e.g., shortening the side chain, may reduce or even abolish the physiologic effects of these compounds.

Absorption. Absorption of the tocopherols is not efficiently accomplished

but is is believed to occur similarly to that of the other fat-soluble vitamins. Bile salts and the presence of fats are believed to be useful if not entirely essential. However, rancid fats destroy this vitamin by oxidation (p. 903).

If the mother is fed an adequate diet, the fetus absorbs through the placenta sufficient tocopherol for its needs but not enough for storage. This must be supplied to the young animal (and presumably to the infant) by the milk. Storage occurs in various tissues but chiefly in adipose tissue.

Water-soluble forms are available. The water-soluble disodium phosphate ester of α-tocopherol, when administered to rabbits intramuscularly, has a more rapid and constant effect than the oil-soluble vitamin administered orally.

Most of the plasma vitamin E is transported by lipoproteins, which also account for most of the blood lipids (p. 256).

Effects of deficiency. In rats, a lack of α-tocopherol results in damage to the reproductive system of both males and females. There is a degeneration of the germinal epithelium that cannot be remedied, after it is once established, by feeding the vitamin. If the female fed a vitamin E–free diet does become pregnant, the embryo dies and is resorbed.

At the present time, it cannot be definitely stated that man requires vitamin E for reproduction. Many clinical investigations have been reported in which vitamin E concentrates were used to attempt to remedy sterility, habitual abortion, and various abnormalities of premature infants, but results are doubtful.

Besides having effects on the reproductive system, vitamin E is also necessary for the structural and functional maintenance of skeletal muscle, cardiac muscle, smooth muscle, and the peripheral vascular system in a variety of laboratory animals. Indeed, the present opinion is that the effects on muscle are of greater importance than are the effects on fertility. Muscular dystrophy and morphologic changes in various tissues are caused by a vitamin E deficit and are accompanied by increased oxygen consumption of the muscle and by alterations in chemical composition and functional behavior. Creatine elimination then is increased (p. 717). This latter effect is believed to be the result of an inability of the skeletal muscle to utilize creatine.

A type of muscular dystrophy can be produced in lambs by the feeding of a raw kidney bean–hay ration. The condition can be prevented by administering α-tocopherol *plus selenium* (p. 551). The unheated beans are believed to contain a heat-labile antivitamin E or antiselenium factor, or both.

Probably the factors responsible for fertility and muscular dystrophy are somewhat different. α-Tocopherylhydroquinone has no antisterility activity but does have a curative effect on muscular dystrophy in rabbits. The hydroquinone is an antidystrophic factor, whereas the tocopherol has chiefly an antisterility action. In chicks, a vitamin E deficiency results in injury to the nervous system from an impairment of the blood vessels in the brain. In all these conditions, there seems to be no comparable effect on human beings.

In rabbits, vitamin E deficiency causes derangement of nucleic acid metabolism. This is shown by a higher output of allantoin and a change in the content of tissue nucleic acids.

Vitamin E has a sparing action on vitamin A and carotene; e.g., vitamin A

and carotene are more effective in curing their deficiency symptoms if vitamin E is administered at the same time. Ingestion of extra amounts of α-tocopherol increases the storage of vitamin A in the liver of rats, and many other examples of the close connection between these two vitamins could be cited. This biologic relation undoubtedly has a chemical basis. Vitamin E is an antioxidant; i.e., it can prevent the oxidation of various other easily oxidized substances, notably fats (p. 903) and vitamin A. For this reason it is often added to foods to prevent oxidation. Possibly this protection is effective even within the cells. The antioxidant properties of E are enhanced by certain other substances, many of which are also antioxidants. Phenols and ascorbic acid are notable examples.

Other work suggests the involvement of vitamin E in combating certain human nutritional anemias. Serum α-tocopherol levels have been found to be low in some types of anemia, particularly in infants. The anemia is a macrocytic type and responds favorably to vitamin E therapy. The erythrocytes appear to be less resistant to hemolysis, resulting in a hemolytic type of anemia. The vitamin E may protect unsaturated fatty acids in the erythrocyte cell membrane from oxidative destruction, and hence prevent hemolysis of the cell. There is some current evidence that α-tocopherol may be involved in the biosynthesis of heme. In vitamin E deficiency there appears to be an impairment in the induction of the enzymes, δ-aminolevulinic acid synthetase and possibly also dehydratase (p. 782). Further studies into the biochemical nature of the effect are needed, however.

Mechanism of action. Although the mechanism of action of vitamin E has not been clearly elucidated, there is some evidence that the vitamin may serve as a cofactor in the electron-transfer system, probably functioning between cytochromes b and c. α-Tocopherol most likely acts as an antioxidant for preserving the physiologic lipid configuration of cell membranes, protecting especially linoleic acid and perhaps other unsaturated fatty acids.

Human requirements. The Food and Nutrition Board of the National Research Council again has included vitamin E in its 1974 revision of recommended daily allowances (p. 510). The recommended allowance for adults is 15 international units for men and 12 IU for women. The amount for women is increased to 15 IU during pregnancy and lactation. For infants and children, 4 to 15 IU are recommended, depending on age and sex. The amounts of vitamin E needed apparently increase when the amounts of dietary polyunsaturated fatty acids increase.

One international unit (IU) of *dl*-α-tocopherol is defined as the biologic activity of 1.1 mg. of the pure compound (or 0.67 mg. *d*-α-tocopherol).

Nutritional status with respect to vitamin E in man is usually estimated from the plasma (or serum) concentration, which varies from 0.5 to 1.2 mg. per 100 ml. of total tocopherol–normal adult populations. Values below 0.5 mg are undesirable.

Toxicity. Tocopherols are considered to be relatively nontoxic in man. However, according to recent studies large doses are definitely toxic in animals. Therefore, as in the case of vitamins A and D, caution should be exercised in the use of large doses for any prolonged period of time.

Phylloquinone (vitamin K)

Dam, a Danish investigator, in 1919, discovered a hemorrhagic disease in chicks attributable to the lack of a food factor that he later (1934) called in Ger-

cobalamin. *p*-Aminobenzoic acid, inositol, α-lipoic acid, and choline are frequently included in this list, but many nutritionists believe that these are not true vitamins, although deficiencies of them in the diet of experimental animals cause characteristic symptoms to develop. None of them, with the possible exception of α-lipoic acid, appears to be a coenzyme or a part of one.

Ascorbic acid (vitamin C)

Scurvy was probably the first disease to be definitely associated with a food deficiency. It was common in Europe in the fifteenth century and must have been known long before that. It frequently occurred among sailors on long voyages when fresh food was not available (p. 556). The essential role of vitamin C (ascorbic acid) in the prevention and cure of scurvy both in man and in certain experimental animals is now well established.

Properties. Ascorbic acid is water soluble and insoluble in fats and oils. It is very sensitive to oxidation, particularly in the presence of copper but not of aluminum. Therefore, foods prepared in copper vessels lose ascorbic acid quickly. This factor is also rapidly destroyed by alkalies but is fairly stable in weakly acid solutions. Consequently, baking soda has a harmful effect, but cooking in steam has little destructive action on the ascorbic acid of foods, if they are neutral or slightly acid. Drying vegetables usually results in a loss of ascorbic acid, but many attempts have been made to provide desiccated foods containing all the vitamins, including ascorbic acid, unchanged. Freezing has no deleterious effect on the vitamin. Because it is so easily oxidized, ascorbic acid is a strong reducing agent.

Occurrence. From a nutritional standpoint, the citrus fruits and tomatoes are the best sources of ascorbic acid. Other natural sources may be richer in it, but they are either inedible or are not consumed in considerable amounts. For example, both green peppers and parsley are richer than oranges in this vitamin, but they do not enter into the diet to any great extent. Spinach and other greens are good sources of it also, but they lose their vitamin C content progressively on storage at room temperature. Citrus fruit juices and tomato juice may be canned with but slight loss of the antiscorbutic factor. However, they should not be permitted to be in contact with air for a long period of time because of loss by oxidation. Cantaloupes, strawberries, cabbage, and turnips, when raw, are all about equivalent to tomatoes, but the two latter lose some vitamin C in cooking. Potatoes, fresh peas, asparagus, and lettuce are good sources also.

Plant polyphenols apparently play an important part as antioxidants in protecting ascorbic acid from oxidative destruction. A number of polyphenols, especially rutin, quercetin, and related flavonols, have this property. The effect is believed to be indirect, because of the chelation of heavy metal ions (Cu^{++}, etc.) that catalyze the oxidative degradation of ascorbic acid. The vitamin C–like action of the so-called bioflavonoids is attributed to this protective action. Bioflavonoids thus decrease oxidative losses of ascorbic acid from foods, etc. during storage or in the intestinal tract, especially in individuals in achlorhydria or hypochlorhydria.

Ascorbic acid occurs to some extent in animal tissues. In 1928, Szent-Györgyi found a "hexuronic acid" with high reducing power in the adrenal cortex and later showed that it had antiscorbutic properties. However, the adrenal