6 Vitamins in milk and dairy products

6.1 Introduction

Vitamins are organic chemicals required by the body in trace amounts but which cannot be synthesized by the body. The vitamins required for growth and maintenance of health differ between species; compounds regarded as vitamins for one species may be synthesized at adequate rates by other species. For example, only primates and the guinea-pig require ascorbic acid (vitamin C; section 6.4) from their diet; other species possess the enzyme gluconolactone oxidase which is necessary for the synthesis of vitamin C from D-glucose or D-galactose. The chemical structures of the vitamins have no relationship with each other. The principal classification of vitamins is based on their solubility in water. Water-soluble vitamins are the B group (thiamin, riboflavin, niacin, biotin, panthothenate, folate, pyridoxine (and related substances, vitamin B_6) and cobalamin (and its derivatives, vitamin B_{12})) and ascorbic acid (vitamin C) while the fat-soluble vitamins are retinol (vitamin A), calciferols (vitamin D), tocopherols (and related compounds, vitamin E) and phylloquinone (and related compounds, vitamin K). The water-soluble vitamins and vitamin K function as co-enzymes while vitamin A is important in the vision process, vitamin D functions like a hormone and vitamin E is primarily an antioxidant.

Milk is the only source of nutrients for the neonatal mammal during the early stage of life until weaning. Thus, in addition to providing macronutrients (protein, carbohydrate and lipid) and water, milk must also supply sufficient vitamins and minerals to support the growth of the neonate. Human beings continue to consume milk into adulthood and thus milk and dairy products continue to be important sources of nutrients in the diet of many peoples worldwide. The concentrations of macronutrients and minerals in milk have been discussed in Chapters 1 and 5; vitamin levels in milk and dairy products will be considered here. Milk is normally processed to a lesser or greater extent before consumption. Thus it is important to consider the influence of processing on the vitamin status of milk and dairy products.
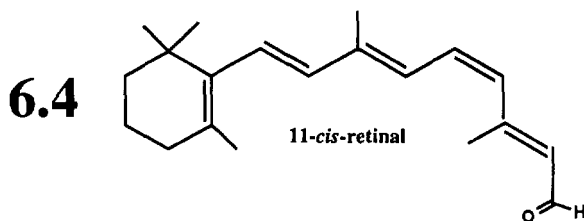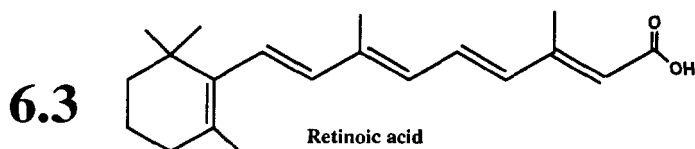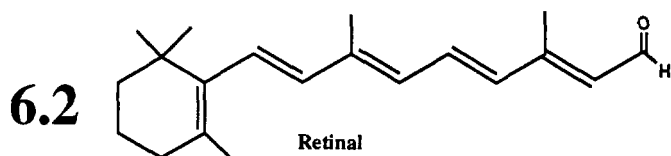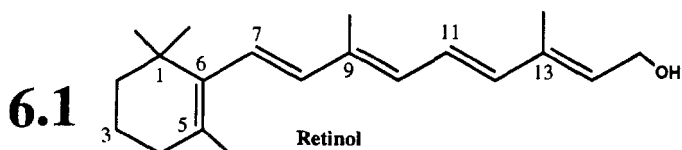
Recommended dietary allowances (RDA) for vitamins are recommended intake of various vitamin to ensure the good health of a high proportion of the human population. The RDA values quoted below refer to the United States population (Whitney and Rolfes, 1996). Reference nutrient intake

(RNI) is the quantity of a nutrient sufficient to meet the needs of 97% of the population. Nutrient intakes equal to the RNI thus pose only a very small risk of deficiency. United Kingdom RNI values (Department of Health, 1991) are also quoted below.
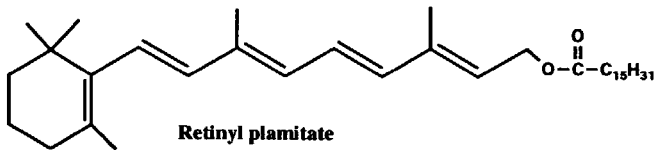
6.2   Fat-soluble vitamins

6.2.1   Retinol (vitamin A)

Vitamin A (retinol, **6.1**) is the parent of a range of compounds known as retinoids, which possess the biological activity of vitamin A. In general, animal foods provide preformed vitamin A as retinyl esters (e.g. **6.5**, which are easily hydrolysed in the gastrointestinal tract) while plant foods provide precursors of vitamin A, i.e. carotenoids. Only carotenoids with a β-ionone ring (e.g. β-carotene) can serve as vitamin A precursors. β-Carotene (**6.6**)

6.1

Retinol

6.2

Retinal

6.3

Retinoic acid

6.4

11-cis-retinal

6.5

Retinyl plamitate

6.6

β-Carotene

Cleavage at this point results
in two molecules of Vitamin A

15

15'
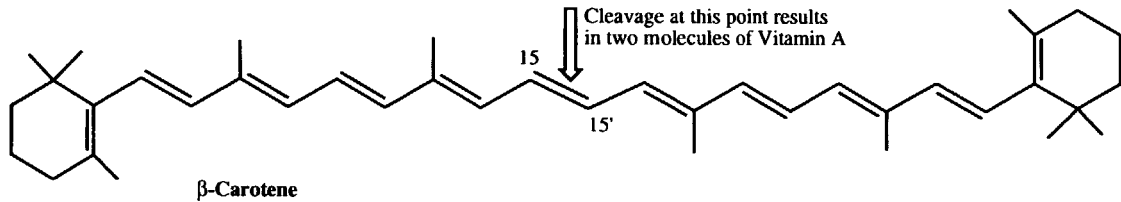
may be cleavaged at its centre by the enzyme, β-carotene-15,15'-oxygenase (present in the intestinal mucosa), to yield 2 mol retinol per mol. However, cleavage of other bonds results in the formation of only 1 molecule of retinol per molecule of β-carotene. In practice, 6 μg β-carotene will yield only 1 μg of retinol. Likewise, 12 μg other carotenes which are vitamin A precursors (i.e. which contain one β-ionone ring) are required to yield 1 μg of retinol. Thus, 1 retinol equivalent (RE) is defined as 1 μg retinol, 6 μg β-carotene or 12 μg of other precursor carotenes.

Retinol can be oxidized to retinal (**6.2**) and further to retinoic acid (**6.3**). *Cis–trans* isomerization can also occur, e.g. the conversion of all *trans*-retinal to 11-*cis*-retinal (**6.4**), which is important for vision.

Vitamin A has a number of roles in the body: it is involved in the vision process, in cell differentiation, in growth and bone remodelling and in the immune system. US RDAs for vitamin A are 1000 μg RE day^{-1} for men and 800 μg RE day^{-1} for women. UK RNI values for vitamin A are 700 and 600 μg RE day^{-1} for adult men and women, respectively. The body will tolerate a wide range of vitamin A intakes (500–15 000 μg RE day^{-1}) but insufficient or excessive intakes result in illness. Vitamin A deficiency (< 500 μg RE day^{-1}) results in night blindness, xerophthalmia (progressive blindness caused by drying of the cornea of the eye), keratinization (accumulation of keratin in digestive, respiratory and urogenital tract tissues) and finally exhaustion and death. At excessive intake levels (> 15 000 μg RE day^{-1}), vitamin A is toxic. Symptoms of hypervitaminosis A include skin rashes, hair loss, haemorrhages, bone abnormalities and fractures, and in extreme cases, liver failure and death.

The major dietary sources of retinol are dairy products, eggs and liver, while important sources of β-carotene are spinach and other dark-green leafy vegetables, deep orange fruits (apricots, cantaloupe) and vegetables (squash, carrots, sweet potatoes, pumpkin). The richest natural sources of vitamin A are fish liver oils, particularly halibut and shark.

Vitamin A activity is present in milk as retinol, retinyl esters and as carotenes. Whole cows' milk contains an average of 52 μg retinol and 21 μg carotene per 100 g. The concentration of retinol in raw sheep's and pasteurized goats' milks is 83 and 44 μg per 100 g, respectively, although milks of these species are reported (Holland *et al.*, 1991) to contain only trace amounts of carotenes. Human milk and colostrum contain an average of 58 and 155 μg retinol per 100 g, respectively. In addition to their role as provitamin A, the carotenoids in milk are reponsible for the colour of milk fat (Chapter 11).

The concentration of vitamin A and carotenoids in milk is strongly influenced by the carotenoid content of the feed. Milk from animals fed on pasture contains higher levels of carotenes than that from animals fed on concentrate feeds. There is also a large seasonal variation in vitamin A concentration; summer milk contains an average of 62 μg retinol and 31 μg carotene per 100 g while the values for winter milk are 41 and 11 μg per

100 g, respectively. The breed of cow also has an influence on the concentration of vitamin A in milk: milk from Channel Islands breeds typically contains 65 μg and 27 μg retinol per 100 g in summer and winter, respectively, and 115 and 27 μg carotene per 100 g in summer and winter, respectively.

Other dairy products are also important sources of vitamin A (Appendix 6A). Whipping cream (39% fat) contains about 565 μg retinol and 265 μg carotene per 100 g. The level of vitamin A in cheese varies with the fat content (Appendix 6A). Camembert (23.7% fat) contains 230 μg retinol and 315 μg carotene per 100 g, while Cheddar (34.4% fat) contains 325 μg retinol and 225 μg carotene per 100 g. Whole-milk yogurt (3% fat; unflavoured) contains roughly 28 μg retinol and 21 μg carotene per 100 g while the corresponding values for ice-cream (9.8% fat) are 115 and 195 μg per 100 g, respectively.

Vitamin A is relatively stable to most dairy processing operations. In general, vitamin A activity is reduced by oxidation and exposure to light. Heating below 100°C (e.g. pasteurization) has little effect on the vitamin A content of milk, although some loss may occur at temperatures above 100°C (e.g. when frying using butter). Losses of vitamin A can occur in UHT milk during its long shelf-life at ambient temperatures. Vitamin A is stable in pasteurized milk at refrigeration temperatures provided the milk is protected from light, but substantial losses can occur in milk packaged in translucent bottles. Low-fat milks are often fortified with vitamin A for nutritional reasons. Added vitamin A is less stable to light than the indigenous vitamin. The composition of the lipid used as a carrier for the exogenous vitamin influences its stability. Protective compounds (e.g. ascorbyl palmitate or β-carotene) will reduce the rate at which exogenous vitamin A is lost during exposure to light. Yogurts containing fruit often contain higher concentrations of vitamin A precursor carotenoids than natural yogurts. The manufacture of dairy products which involves concentration of the milk fat (e.g. cheese, butter) results in a *pro rata* increase in the concentration of vitamin A. The increased surface area of dried milk products accelerates the loss of vitamin A; supplementation of milk powders with vitamin A and storage at low temperatures minimizes these losses.

6.2.2   Calciferols (vitamin D)

Unlike other vitamins, cholecalciferol (vitamin D_3) can be formed from a steroid precursor, 7-dehydrocholesterol (6.7), by the skin when exposed to sunlight; with sufficient exposure to the sun, no preformed vitamin D is required from the diet.

UV light (280–320 nm) causes the photoconversion of 7-dehydrocholesterol to pre-vitamin D_3. This pre-vitamin can undergo further photoconversion to tachysterol and lumisterol or can undergo a temperature-dependent isomerization to cholecalciferol (vitamin D_3, 6.8). At body temperature, this
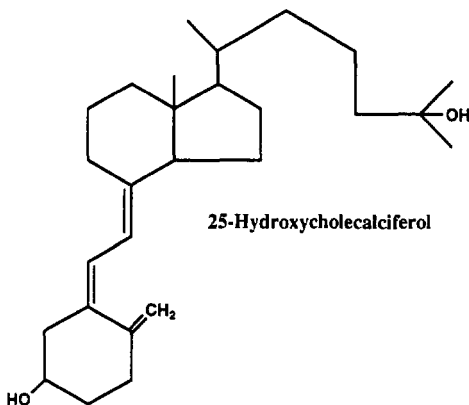
6.7

7-Dehydrocholesterol

6.8

Cholecalciferol, Vitamin D_3

6.9

25-Hydroxycholecalciferol
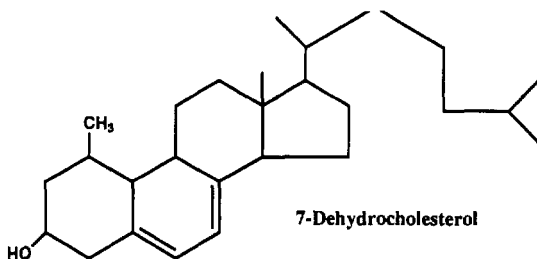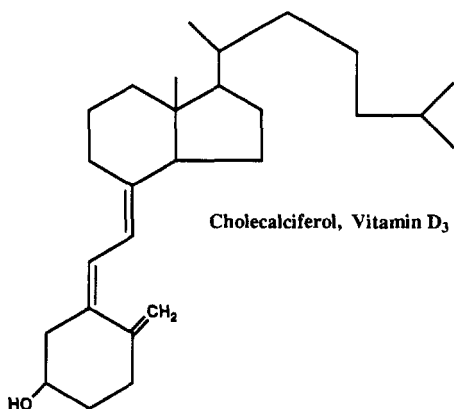
conversion requires about 28 h to convert 50% of previtamin D_3 to vitamin D_3. Thus, production of vitamin D_3 in the skin can take a number of days. Preformed vitamin D_3 is obtained from the diet. Vitamin D_3 is stored in various fat deposits around the body. Regardless of the source of vitamin D_3, it must undergo two hydroxylations to become fully active. Vitamin D_3 is transported by a specific binding protein through the circulatory system to the liver where the enzyme, 25-hydroxylase, converts it to 25-hydroxy-

6.10

1, 25-Dihydroxycholecalciferol
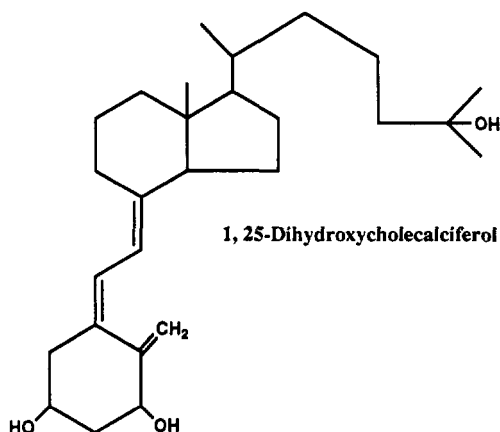
cholecalciferol (25(OH)D$_3$; **6.9**) which is converted to 1,25-dihydroxy-cholecalciferol (1,25(OH)$_2$D$_3$; **6.10**) by the enzyme, 1-hydroxylase, in the kidney. Alternatively, 25(OH)D$_3$ can be hydroxylated at position 24 to form 24,25-dihydroxycholecalciferol (24,25(OH)$_2$D$_3$). At least 37 metabolites of vitamin D$_3$ have been identified, but only 3,25(OH)$_2$D$_3$, 24,25(OH)$_2$D$_3$ and 1,25(OH)$_2$D$_3$ have significant biological activity; 1,25(OH)$_2$D$_3$ is the most biologically active metabolite of vitamin D$_3$.

Vitamin D$_2$ (ergocalciferol) is formed by the photoconversion of ergosterol, a sterol present in certin fungi and yeasts, and differs from cholecalciferol in having an extra methyl group at carbon 24 and an extra double bond between C$_{22}$ and C$_{23}$. Ergocalciferol was widely used for many years as a therapeutic agent.

The principal physiological role of vitamin D in the body is to maintain plasma calcium by stimulating its absorption from the gastrointestinal tract, its retention by the kidney and by promoting its transfer from bone to the blood. Vitamin D acts in association with other vitamins, hormones and nutrients in the bone mineralization process. In addition, vitamin D has a wider physiological role in other tissues in the body, including the brain and nervous system, muscles and cartilage, pancreas, skin, reproductive organs and immune cells.

The RDA for vitamin D is 10 and 5 μg day^{-1} for persons aged 19–24 years or over 25 years, respectively. RNI values for vitamin D are 10 μg day^{-1} for persons over 65 years and for pregnant or lactating women. With the exception of these and other at-risk groups, the RNI value for dietary vitamin D is 0 μg day^{-1}. The classical syndrome of vitamin D deficency is rickets, in which bone is inadequately mineralized, resulting in growth retardation and skeletal abnormalities. Adult rickets or osteomalacia occurs most commonly in women who have low calcium intakes and little exposure to sunlight and have had repeated pregnancies or periods

of lactation. Hypervitaminosis D (excess intake of vitamin D) is character-
ized by enhanced absorption of calcium and transfer of calcium from bone
to the blood. These cause excessively high concentrations of serum calcium
which can precipitate at various locations in the body, causing kidney stones
or calcification of the arteries. Vitamin D can exert these toxic effects if
consumed continuously at only relatively small amounts in excess of the
RDA.

Relatively few foods contain significant amounts of vitamin D. In
addition to conversion *in situ* by the body, the principal sources of vitamin
D are foods derived from animal sources, including egg yolk, fatty fish and
liver. Unfortified cows' milk is not an important source of vitamin D.

The major form of vitamin D in both cows' and human milk is
$25(OH)D_3$. This compound is reported to be responsible for most of the
vitamin D in the blood serum of exclusively breast-fed infants. Whole cows'
milk contains only about $0.03\,\mu g$ vitamin D per $100\,g$ and 1 litre of milk per
day will supply only 10–20% of the RDA. Therefore, milk is often fortified
(at the level of $c.\ 1-10\,\mu g\,l^{-1}$) with vitamin D. Fortified milk, dairy products
or margarine are important dietary sources of vitamin D. The concentration
of vitamin D in unfortified dairy products is usually quite low. Vitamin D
levels in milk vary with exposure to sunlight.

As with other fat-soluble vitamins, the concentration of vitamin D in
dairy products is increased *pro rata* by concentration of the fat (e.g. in the
production of butter or cheese). Vitamin D is relatively stable during storage
and to most dairy processing operations. Studies on the degradation of
vitamin D in fortified milk have shown that the vitamin may be degraded
by exposure to light. However, the conditions necessary to cause significant
losses are unlikely to be encountered in practice. Extended exposure to light
and oxygen are needed to cause significant losses of vitamin D.

6.2.3   Tocopherols and related compounds (vitamin E)

Eight compounds have vitamin E activity, four of which are derivatives of
tocopherol (**6.11**) and four of tocotrienol (**6.12**); all are derivatives of
6-chromanol. Tocotrienols differ from tocopherols in having three carbon–
carbon double bonds in their hydrocarbon side chain. α-, β-, γ- or δ-
tocopherols and tocotrienols differ with respect to number and position of
methyl groups on the chromanol ring. The biological activity of the different
forms of the tocopherols and tocotrienols varies with their structure. D- and
L-enantiomers of vitamin E also occur; the biological activity of the D-form
is higher than that of the L-isomer. Vitamin E activity can be expressed as
tocopherol equivalents (TE), where 1 TE is equivalent to the vitamin E
activity of $1\,mg$ α-D-tocopherol. The biological activity of β- and γ-
tocopherols and α-tocotrienol is 50, 10 and 33% of the activity of α-D-
tocopherol, respectively.

6.11

Tocopherols

6.12

Tocotrienols
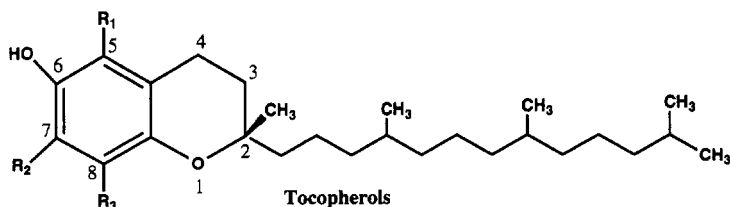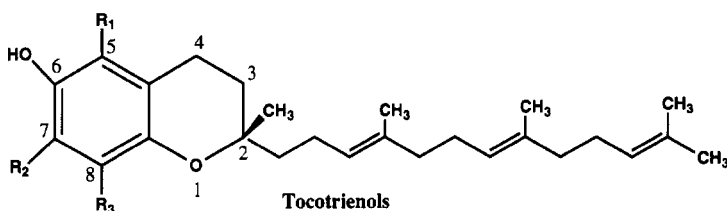
α– $R_1 = CH_3, R_2 = CH_3, R_3 = CH_3$
β– $R_1 = CH_3, R_2 = H, R_3 = CH_3$
γ– $R_1 = H, R_2 = CH_3, R_3 = CH_3$
δ– $R_1 = H, R_2 = H, R_3 = CH_3$

Vitamin E is a very effective antioxidant. It can easily donate a hydrogen from the phenolic -OH group on the chromanol ring to free radicals. The resulting vitamin E radical is quite unreactive as it is stabilized by delocalization of its unpaired electron into the aromatic ring. Vitamin E thus protects the lipids (particularly polyunsaturated fatty acids) and membranes in the body against damage caused by free radicals. The role of vitamin E is of particular importance in the lungs where exposure of cells to oxygen is greatest. Vitamin E also exerts a protective effect on red and white blood cells. It has been suggested that the body has a system to regenerate active vitamin E (perhaps involving vitamin C) once it has acted as an antioxidant.

Vitamin E deficiency is normally associated with diseases of fat malabsorption and is rare in humans. Deficiency is characterized by erythrocyte haemolysis and prolonged deficiency can cause neuromuscular dysfunction. Hypervitaminosis E is not common, despite an increased intake of vitamin E supplements. Extremely high doses of the vitamin may interfere with the blood clotting process.

The RDAs for vitamin E are 10 mg and 8 mg α-TE day^{-1} for men and women, respectively. UK RNI values have not been established for vitamin E since its requirement is largely dependent on the content of polyunsaturated lipids in the diet. However, the Department of Health (1991) suggested that 4 and 3 mg α-TE day^{-1} are adequate for men and women, respectively. The major food sources of vitamin E are polyunsaturated vegetable oils and products derived therefrom (e.g. maragrine, salad dressings), green and leafy

vegetables, wheat germ, whole-grain cereal products, liver, egg yolk, nuts and seeds.
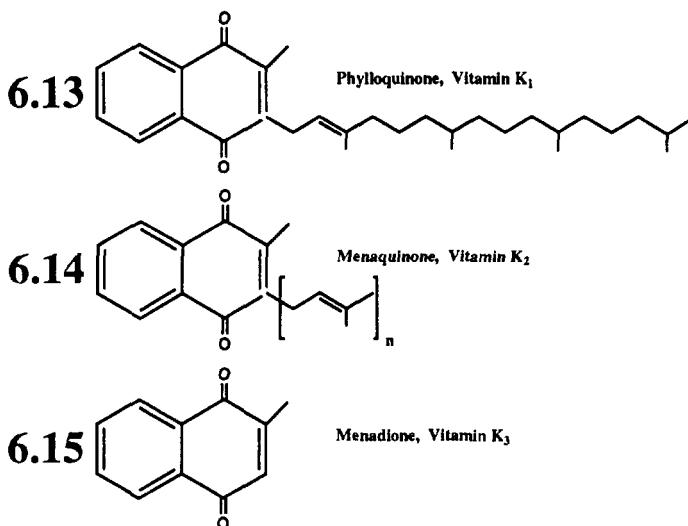
The concentration of vitamin E in cows' milk is quite low (0.09 mg per 100 g) and is higher in summer than in winter milks. Human milk and colostrum contain somewhat higher concentrations (~ 0.3 and ~ 1.3 mg per 100 g, respectively). Most dairy products contain low levels of vitamin E (Appendix 6A) and thus are not important sources of this nutrient. However, levels are higher in dairy products supplemented with vegetable fat (e.g. some ice-creams, imitation creams, fat-filled dried skim milk). Like other fat-soluble vitamins, the concentration of vitamin E in dairy products is increased *pro rata* with fat content. Vitamin E is relatively stable below 100°C but is destroyed at higher temperatures (e.g. deep-fat frying). The vitamin may also be lost through oxidation during processing. Oxidative losses are increased by exposure to light, heat or alkaline pH, and are promoted by the presence of pro-oxidants, lipoxygenase or catalytic trace elements (e.g. Fe^{3+}, Cu^{2+}). Pro-oxidants increase the production of free radicals and thus accelerate the oxidation of vitamin E. Exogenous vitamin E in milk powders supplemented with this nutrient appears to be stable for long storage periods if the powders are held at or below room temperaure. The potential of feed supplemented with vitamin E to increase the oxidative stability of milk has been investigated, as has the potential use of exogenous tocopherols added directly to the milk fat.

6.2.4   *Phylloquinone and related compounds (vitamin K)*

The structure of vitamin K is characterized by methylnaphthoquinone rings with a side chain at position 3. It exists naturally in two forms: phylloquinone (vitamin K_1; **6.13**) occurs only in plants, while menaquinones (vitamin K_2; **6.14**) are a family of compounds with a side chain consisting of between 1 and 14 isoprene units. Menaquinones are synthesized only by bacteria (which inhabit the human gastrointestinal tract and thus provide some of the vitamin K required by the body). Menadione (vitamin K_3; **6.15**) is a synthetic compound with vitamin K activity. Unlike K_1 and K_2, menadione is water soluble and is not active until it is alkylated *in vivo*.

The physiological role of vitamin K is in blood clotting and is essential for the synthesis of at least four of the proteins (including prothrombin) involved in this process. Vitamin K also plays a role in the synthesis of a protein (osteocalcin) in bone. Vitamin K deficiency is rare but can result from impaired absorption of fat. Vitamin K levels in the body are also reduced if the intestinal flora is killed (e.g. by antibiotics). Vitamin K toxicity is rare but can be caused by excessive intake of vitamin K supplements. Symptoms include erythrocyte haemolysis, jaundice, brain damage and reduced effectiveness of anticoagulants.

The RDAs for vitamin K for people aged 19–24 years are 70 μg and 60 μg day^{-1} for men and women, respectively. Corresponding values for

6.13 Phylloquinone, Vitamin K_1

6.14 Menaquinone, Vitamin K_2

6.15 Menadione, Vitamin K_3

adults aged 25 years and over are 80 and 65 μg day^{-1}. The Department of Health (1991) suggested that a vitamin K intake of 1 μg per kg body weight per day is safe and adequate. The principal food sources of vitamin K are liver, green leafy vegetables and milk.

Whole cows' milk contains 0.4–1.8 μg vitamin K per 100 g while human milk contains about 0.2 μg per 100 g. Human colostrum contains higher concentrations of vitamin K, which are necessary since bacteria capable of synthesizing vitamin K take time to become established in the intestine of the neonate. Irradiation under anerobic and apolar conditions can result in *cis–trans* isomerization, resulting in loss of activity since only the *trans* isomer has vitamin K activity. However, unit operations in dairy processing are unlikely to have an effect on the stability of this nutrient.

6.3 B-group vitamins

The B-group is a heterogeneous collection of water-soluble vitamins, most of which function as co-enzymes or are precursors of co-enzymes. The B-group vitamins are thiamin, riboflavin, niacin, biotin, pantothenic acid, pyridoxine (and related substances, vitamin B_6), folate and cobalamin (and its derivatives, vitamin B_{12}).
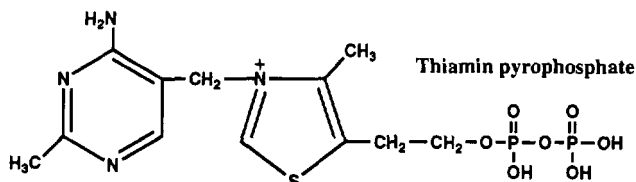
6.3.1   Thiamin (vitamin B_1)

Thiamin (vitamin B_1; **6.16**) consists of two heterocyclic rings (substitued pyrimidine and substituted thiazole), linked by a methylene bridge. Thiamin acts as a co-enzyme in the form of thiamin pyrophosphate (TPP; **6.17**)

6.16

Thiamin (Vitamin B$_1$)

6.17

Thiamin pyrophosphate
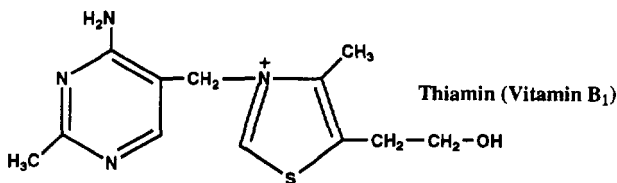
which is an essential co-factor for many enzyme-catalysed reactions in carbohydrate metabolism. TPP-dependent pyruvate dehydrogenase catalyses the conversion of pyruvate ($CH_3COCOOH$) to acetyl CoA (CH_3CO-CoA) in mitochondria. The acetyl CoA produced in this reaction enters the Krebs cycle and also serves as a substrate for the synthesis of lipids and acetylcholine (and thus is important for the normal functioning of the nervous system). TPP is necessary in Krebs cycle for the oxidative decarboxylation of α-ketoglutarate ($HOOCCH_2CH_2COCOOH$) to succinyl CoA ($HOOCCH_2CH_2CO$-CoA) by the α-ketoglutarate dehydrogenase complex. TPP also functions in reactions involving the decarboxylation of ketoacids derived from branched-chain amino acids and in transketolase reactions in the hexose monophosphate pathway for glucose metabolism.

The characteristic disease caused by prolonged thiamin deficiency is beriberi, the symptoms of which include oedema, enlarged heart, abnormal heart rhythms, heart failure, wasting, weakness, muscular problems, mental confusion and paralysis.

Thiamin is widespread in many nutritious foods but pig meat, liver, whole-grain cereals, legumes and nuts are particularly rich sources. Because of its importance in energy metabolism, the RDA for thiamin is quoted in terms of energy intake (0.12 mg kJ^{-1} day^{-1}; 1 mg day^{-1} minimum). This is approximately equivalent to 1.5 mg and 1.1 mg per day for men and women, respectively. The RNI value for thiamin is 0.4 mg per 1000 kcal (4186 kJ) per day for adults.

Milk contains, on average, 0.03 mg thiamin per 100 g. Most (50–70%) of the thiamin in bovine milk is in the free form; lesser amounts are phosphorylated (18–45%) or protein-bound (7–17%). The concentration in mature human milk is somewhat lower (c. 0.02 mg per 100 g). Human colostrum contains only trace amounts of thiamin which increase during lactation. Pasteurized milk from goats and Channel Island breeds of cow contain about 0.04 mg per 100 g, while values for raw sheep's milk are somewhat higher, with an average of 0.08 mg per 100 g. Most of the thiamin

in bovine milk is produced by micro-organisms in the rumen and, therefore, feed, breed of the cow or season have relatively little effect on its concentration in milk.

Thiamin levels in milk products (Appendix 6A) are generally 0.02–0.05 mg per 100 g. As a result of the growth of the *Penicillium* mould, the rind of Brie and Camembert cheese is relatively rich in thiamin (0.5 and 0.4 mg per 100 g, respectively).

Thiamin is relatively unstable and is easily cleaved by a nucleophilic displacement reaction at its methylene carbon. The hydroxide ion (OH^-) is a common nucelophile which can cause this reaction in foods. Thiamin is thus more stable under slightly acid conditions. Thiamin is reported to be relatively stable to pasteurization and UHT heat treatment ($\leqslant 10\%$ losses) and during the storage of pasteurized milk, but losses of 20–40% have been reported for UHT milks stored for long periods of time (1–2 years). The light sensitivity of thiamin is less than that of other light-sensitive vitamins.

6.3.2 Riboflavin (vitamin B₂)

Riboflavin (vitamin B_2; **6.18**) consists of an isoalloxazine ring linked to an alcohol derived from ribose. The ribose side chain of riboflavin can be modified by the formation of a phosphoester (forming flavin mononucleotide, FMN, **6.19**). FMN can be joined to adenine monophosphate to form flavin adenine dinucleotide (FAD, **6.20**). FMN and FAD act as co-enzymes by accepting or donating two hydrogen atoms and thus are involved in redox reactions. Flavoprotein enzymes are involved in many metabolic pathways. Riboflavin is a yellow-green fluorescent compound and, in addition to its role as a vitamin, it is responsible for the colour of milk serum (Chapter 11).

Symptoms of riboflavin deficiency include cheilosis (cracks and redness at the corners of the mouth), glossitis (painful, smooth tongue), inflamed eyelids, sensitivity of the eyes to light, reddening of the cornea and skin rash. The US RDA for riboflavin is expessed in terms of energy intake ($c.$ 0.14 mg kJ^{-1} day^{-1}, equivalent to about 1.7 and 1.3 mg day^{-1} for men and women, respectively). Corresponding UK RNI values are 1.3 and 1.1 mg day^{-1} for adult men and women, respectively. Important dietary sources of riboflavin include milk and dairy products, meat and leafy green vegetables. Cereals are poor sources of riboflavin, unless fortified. There is no evidence for riboflavin toxicity.

Milk is a good source of riboflavin; whole milk contains about 0.17 mg per 100 g. Most (65–95%) of the riboflavin in milk is present in the free form; the remainder is present as FMN or FAD. Milk also contains small amounts (about 11% of total flavins) of a related compound, 10-(2'-hydroxyethyl) flavin, which acts as an antivitamin. The concentration of this compound must be considered when evaluating the riboflavin activity in milk. The concentration of riboflavin in milk is influenced by the breed of

6.18

Riboflavin

6.19

Flavin mononucleotide

6.20

Flavin adenine dinucleotide
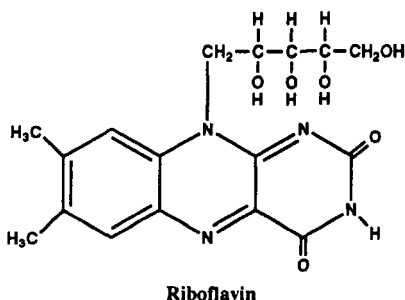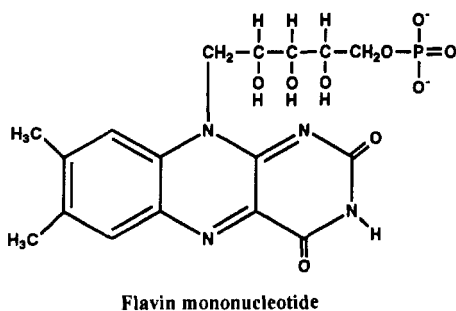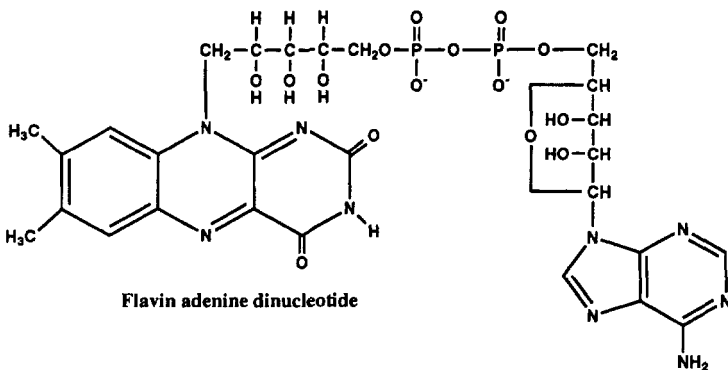
cow (milk from Jersey and Guernsey cows contains more riboflavin than Holstein milk). Summer milk generally contains slighly higher levels of riboflavin than winter milk. Interspecies variations in concentration are also apparent. Raw sheep's milk contains about 0.32 mg per 100 g while the mean value for pasteurized goats' milk (0.13 mg per 100 g) is lower; human milk contains 0.03 mg per 100 g. Dairy products also contain significant amounts

6.21
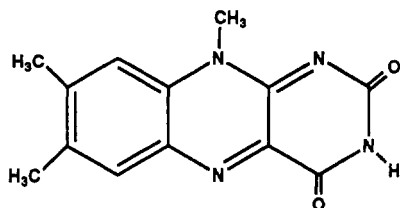
Lumiflavin

of riboflavin (Appendix 6A). Cheese contains 0.3–0.5 mg per 100 g and yogurt about 0.3 mg per 100 g. The whey protein fraction of milk contains a riboflavin-binding protein (RfBP) which probably originates from blood plasma, although its function in milk is unclear.

Riboflavin is stable in the presence of oxygen, heat and at acid pH. However, it is labile to thermal decomposition under alkaline conditions. The concentration of riboflavin in milk is unaffected by pasteurization and little loss is reported for UHT-treated milks. The most important parameter affecting the stability of riboflavin in dairy products is exposure to light (particularly wavelengths in the range 415–455 nm). At alkaline pH, irradiation cleaves the ribitol portion of the molecule, leaving a strong oxidizing agent, lumiflavin (**6.21**). Irradiation under acidic conditions results in the formation of lumiflavin and a blue fluorescent compound, lumichrome. Lumiflavin is capable of oxidizing other vitamins, particularly ascorbate (section 6.4 and Chapter 11). Loss of riboflavin in milk packaged in materials that do not protect against light can be caused by either sunlight or by lights in retail outlets. Packaging in paperboard containers is the most efficient method for minimizing this loss, although glass containing a suitable pigment has also been used. Riboflavin is more stable in high-fat than in low-fat or skim milk, presumably as a result of the presence of antioxidants (e.g. vitamin E) in the milk fat which protect riboflavin against photo-oxidation.

6.3.3   Niacin

Niacin is a generic term which refers to two related chemical compounds, nicotinic acid (**6.22**) and its amide, nicotinamide (**6.23**); both are derivatives of pyridine. Nicotinic acid is synthesized chemically and can be easily converted to the amide in which form it is found in the body. Niacin is obtained from food or can be synthesized from tryptophan (60 mg of dietary tryptophan has the same metabolic effect as 1 mg niacin). Niacin forms part of two important co-enzymes, nicotinamide adenine dinucleotide (NAD) and nicotinamide adenine dinucleotide phosphate (NADP), which are co-factors for many enzymes that participate in various metabolic pathways and function in electron transport.

6.22

Nicotinic acid

6.23

Nicotinamide
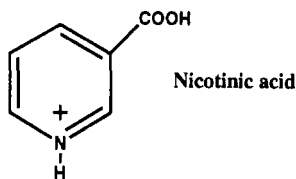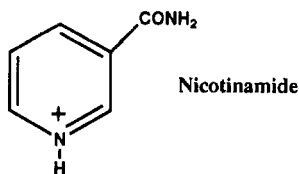
The classical niacin deficiency disease is pellagra, which is characterized by symptoms including diarrhoea, dermatitis, dementia and eventually death. High-protein diets are rarely deficient in niacin since, in addition to the preformed vitamin, such diets supply sufficient tryptophan to meet dietary requirements. Large doses of niacin can cause the dilation of capillaries, resulting in a painful tingling sensation.

The RDA for niacin is expressed in terms of energy intake; 6.6 mg niacin equivalent (NE, 1 mg niacin or 60 mg tryptophan) per 1000 kcal (4186 kJ) per day is recommended (13 NE day^{-1} minimum). This is approximately equivalent to 19 and 15 mg NE day^{-1} for men and women, respectively. The UK RNI value for niacin is 6.6 mg NE per 1000 kcal (4186 kJ) per day for adults. The richest dietary sources of niacin are meat, poultry, fish and whole-grain cereals.

Milk contains about 0.1 mg niacin per 100 g and thus is not a rich source of the preformed vitamin. Tryptophan contributes roughly 0.7 mg NE per 100 g milk. In milk, niacin exists primarily as nicotinamide and its concentration does not appear to be affected greatly by breed of cow, feed, season or stage of lactation. Pasteurized goats' (0.3 mg niacin and 0.7 mg NE from tryptophan per 100 g) and raw sheep's (0.4 mg niacin and 1.3 mg NE from tryptophan per 100 g) milk are somewhat richer than cows' milk. Niacin levels in human milk are 0.2 mg niacin and 0.5 mg NE from tryptophan per 100 g. The concentration of niacin in most dairy products is low (Appendix 6A) but is compensated somewhat by tryptophan released on hydrolysis of the proteins.

Niacin is relatively stable to most food-processing operations. It is stable to exposure to air and resistant to autoclaving (and is therefore stable to pasteurization and UHT treatments). The amide linkage of nicotinamide can be hydrolysed to the free carboxylic acid (nicotinic acid) by treament with acid but the vitamin activity is unaffected. Like other water-soluble vitamins, niacin can be lost by leaching.

6.3.4   Biotin

Biotin (6.24) consists of an imidazole ring fused to a tetrahydrothiophene ring with a valeric acid side chain. Biotin acts as a co-enzyme for carboxylases involved in the synthesis and catabolism of fatty acids and for branched-chain amino acids and gluconeogenesis.

6.24
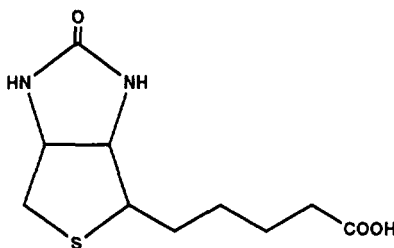
Biotin

Biotin deficiency is rare but under laboratory conditions it can be induced by feeding subjects with large amounts of raw egg white which contains the protein, avidin, which has a binding site for the imidazole moiety of biotin, thus making it unavailable. Avidin is denatured by heat and, therefore, biotin binding occurs only in raw egg albumen. Symptoms of biotin deficiency include scaly dermatitis, hair loss, loss of appetite, nausea, hallucinations and depression.

Biotin is widespread in foods, although its availability is affected somewhat by the presence of binding proteins. Biotin is required in only small amounts. Although US RDA values have not been established, the estimated safe and adequate intake of biotin is $30-100\,\mu g\,day^{-1}$ for adults. The Department of Health (1991) suggested that biotin intakes between 10 and $200\,\mu g\,day^{-1}$ are safe and adequate. Biotin is reported to be non-toxic in amounts up to at least $10\,mg\,day^{-1}$.

Milk contains about $1.9\,\mu g$ biotin per $100\,g$, apparently in the free form. Pasteurized caprine, raw ovine and human milks contain 3.0, 2.5 and $0.7\,\mu g$ per $100\,g$, respectively. The concentration of biotin in cheese ranges from 1.4 (Gouda) to 7.6 (Camembert) μg per $100\,g$ (Appendix 6A). Skim-milk powder contains high levels of biotin ($c.\,20\,\mu g$ per $100\,g$) owing to the concentration of the aqueous phase of milk during its manufacture. Biotin is stable during food processing and storage and is unaffected by pasteurization.

6.3.5   Panthothenic acid

Pantothenic acid (6.25) is a dimethyl derivative of butyric acid linked to β-alanine. Pantothenate is part of the structure of co-enzyme A (CoA), and

6.25
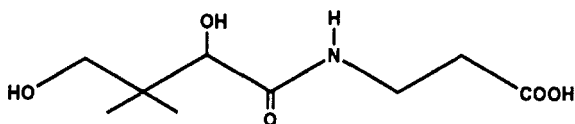
Pantothenic acid

as such is vital as a co-factor for numerous enzyme-catalysed reactions in lipid and carbohydrate metabolism.

Pantothenate deficiency is rare, occurring only in cases of severe malnutrition; characteristic symptoms include vomiting, intestinal distress, insomnia, fatigue and occasional diarrhoea. Pantothenate is widespread in foods; meat, fish, poulty, whole-grain cereals and legumes are particularly good sources. Although no RDA or RNI value has been established for panthothenate, safe and adequate intake of this vitamin for adults is estimated to be $3-7\,\text{mg}\,\text{day}^{-1}$. Pantothenate is non-toxic at doses up to $10\,\text{g}\,\text{day}^{-1}$.

Milk contains, on average, $0.35\,\text{mg}$ panthothenate per $100\,\text{g}$. Pantothenate exists partly free and partly bound in milk and its concentration is influenced by breed, feed and season. Raw ovine and pasteurized caprine milks contain slightly higher concentrations of this nutrient (averaging 0.45 and $0.41\,\text{mg}$ per $100\,\text{g}$, respectively). The values for pantothenate in human milk vary widely; values ranging from 0.2 to $0.7\,\text{mg}$ per $100\,\text{g}$ have been reported. Mean concentrations of pantothenate in cheese vary from about 0.3 (cream cheese, Gouda) to 0.7 (Stilton) mg per $100\,\text{g}$ (Appendix 6A). Pantothenate is stable at neutral pH but is easily hydrolysed by acid or alkali at high temperatures. Pantothenate is reported to be stable to pasteurization.

6.3.6 *Pyridoxine and related compounds (vitamin B_6)*

Vitamin B_6 occurs naturally in three related forms: pyridoxine (**6.26**; the alcohol form), pyridoxal (**6.27**; aldehyde) and pyridoxamine (**6.28**; amine). All are structurally related to pyridine. The active co-enzyme form of this vitamin is pyridoxal phosphate (PLP; **6.29**), which is a co-factor for transaminases which catalyse the transfer of amino groups (**6.29**). PLP is also important for amino acid decarboxylases and functions in the metabolism of glycogen and the synthesis of sphingolipids in the nervous system. In addition, PLP is involved in the formation of niacin from tryptophan (section 6.3.3) and in the initial synthesis of haem.

Deficiency of vitamin B_6 is characterized by weakness, irritability and insomnia and later by convulsions and impairment of growth, motor

6.26

Pyridoxine

6.27

Pyridoxal

6.28

Pyridoxamine

6.29

Pyridoxal phosphate   Pyridoxamine phosphate
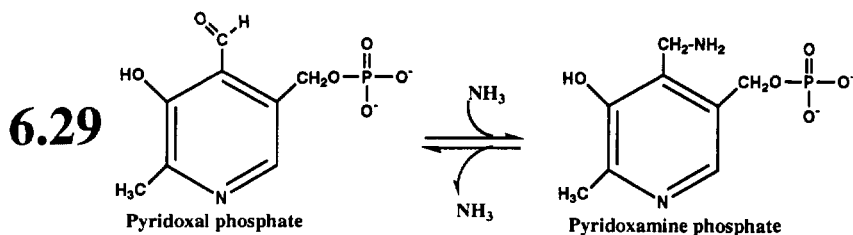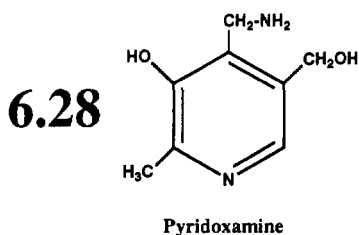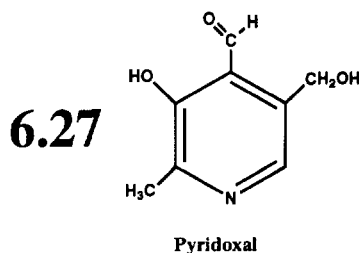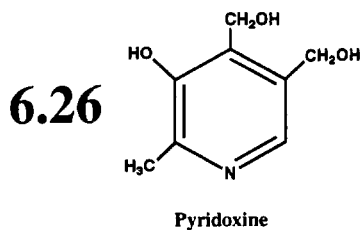
functions and immune response. High doses of vitamin B_6, often associated with excessive intake of supplements, are toxic and can cause bloating, depression, fatigue, irritability, headaches and nerve damage.

Since vitamin B_6 is essential for amino acid (and hence protein) metabolism, its RDA is quoted in terms of protein intake (0.016 mg per g protein per day, equivalent to about 2.0 and 1.6 mg day^{-1} for men and women,

respectively). The corresponding UK RNI value for B_6 is $15\,\mu g\,g^{-1}$ protein for adults. Important sources of B_6 include green, leafy vegetables, meat, fish and poultry, shellfish, legumes, fruits and whole grains.

Whole milk contains, on average, $0.06\,mg\ B_6$ per $100\,g$, mainly in the form of pyridoxal (80%); the balance is mainly pyridoxamine (20%), with trace amounts of pyridoxamine phosphate. Concentrations in raw ovine and pasteurized caprine milks are similar to those in cows' milk (0.08 and $0.06\,mg$ per $100\,g$, respectively). The concentration of B_6 varies during lactation; colostum contains lower levels than mature milk. Seasonal variation in the concentration of vitamin B_6 has been reported in Finnish milk; levels were higher (14%) when cattle were fed outdoors than when they were fed indoors. Mature human milk contains about $0.01\,mg\ B_6$ per $100\,g$.

In general, dairy products are not major sources of B_6 in the diet. Concentrations in cheeses and related products vary from about 0.04 (fromage frais, cream cheese) to 0.22 (Camembert) mg per $100\,g$ (Appendix 6A). Whole-milk yogurt contains roughly 0.1 mg per $100\,g$ and the concentration in skim-milk powder is $c.\ 0.6\,mg$ per $100\,g$.

All forms of B_6 are sensitive to UV light and may be decomposed to biologically inactive compounds. Vitamin B_6 may also be decomposed by heat. Losses of 45% and 20–30% can occur on cooking meat and vegetables, respectively. The aldehyde group of pyridoxal and the amine group of pyridoxamine show some reactivity under conditions that may be encountered during milk processing. An outbreak of B_6 deficiency in 1952 was attributed to the consumption of heated milk products. Pyridoxal and/or its phosphate can react directly with the sulphydryl group of cysteine residues in proteins, forming an inactive thiazolidine derivative (**6.30**). Losses during pasteurization and UHT treatments are relatively small, although losses of up to 50% can occur in UHT milk during its shelf-life.

6.30
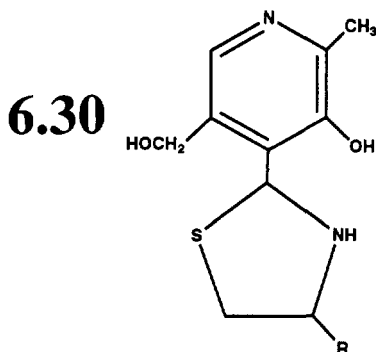
Thiazolidine derivative of pyridoxal

6.3.7 Folate

Folate consists of a substituted pteridine ring linked through a methylene bridge to p-aminobenzoic acid and glutamic acid (**6.31**). Up to seven glutamic acid residues can be attached by γ-carboxyl linkages, producing polyglutamyl folate (**6.31**) which is the major dietary and intracellular form of the vitamin. Reductions and substitutions on the pteridine ring result in tetrahydrofolate (H_4 folate; **6.32**) and 5-methyl tetrahydrofolate (5-methyl-H_4 folate; **6.33**). Folate is a co-factor in the enzyme-catalysed transfer of single carbon atoms in many metabolic pathways, including the biosynthesis of purines and pyramidines (essential for DNA and RNA) and interconversions of amino acids. Folate interacts with vitamin B_{12} (section 6.3.8) in the enzyme-catalysed synthesis of methionine and in the activation of 5-methyl-H_4 folate to H_4 folate. H_4 Folate is involved in a complex and inter-linked series of metabolic reactions (Garrow and James, 1993).

6.31

Folate

6.32

Tetrahydrofolate

6.33
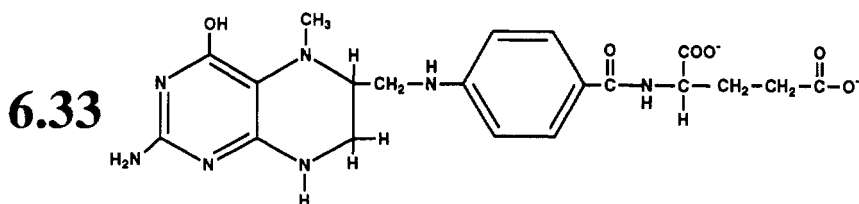
5-methyl tetrahydrofolate

Folate deficiency impairs cell division and protein synthesis; symptoms include megaloblastic anaemia, digestive system problems (heartburn, diarrhoea, constipation), suppression of the immune system, glossitis and problems with the nervous system (depression, fainting, fatigue, mental confusion). The RDA for folate is $3 \mu g$ per kg body weight per day (equivalent to $c.\ 200$ and $180 \mu g\,day^{-1}$ for men and women, respectively). The RNI value for adults is $200 \mu g\,day^{-1}$. Higher intakes of folate have been suggested for women of child-bearing age to prevent the development of neural tube defects in the developing foetus.

Rich dietary sources of folate include leafy green vegetables, legumes, seeds and liver. Milk contains about $6 \mu g$ folate per 100 g. The dominant form of folate in milk is 5-methyl-H_4 folate. Folate in milk is mainly bound to folate-binding proteins and about 40% occurs as conjugated polyglutamate forms. The folate binding proteins of milks of various species have been characterized (Fox and Flynn, 1992). It has been suggested that protein binding increases the bioavailability of folate. Winter milk is reported to contain higher concentrations of folate than summer milk (7 and $4 \mu g$ per 100 g, respectively). Raw sheep's milk contains, on average, $5 \mu g$ per 100 g while the value for pasteurized goats' milk is $1 \mu g$ per 100 g. Folate levels in human milk increase from 2 to $5 \mu g$ per 100 g as colostrum changes to mature milk. Folate levels in some dairy products are shown in Appendix 6A. Whipping cream contains about $7 \mu g$ per 100 g while the value for cheese varies widely from $30-40 \mu g$ per 100 g (Edam, Cheddar) to greater than $100 \mu g$ per 100 g (Camembert); the high concentration found in mould-ripened varieties presumably reflects biosynthesis of folate by the mould. The concentration of folate in yogurt is about $18 \mu g$ per 100 g, principally in the form of formyl folate. The higher level of folate in yogurt is due to biosynthesis, particularly by *Streptococcus salivarius* subsp. *thermophilus*, and perhaps to some added ingredients.

Folate is a relatively unstable nutrient; processing and storage conditions that promote oxidation are of particular concern since some of the forms of folate found in foods are easily oxidized. The reduced forms of folate (dihydro- and tetrahydrofolate) are oxidized to p-aminobenzoylglutamic acid and pterin-6-carboxylic acid, with a concomitant loss in vitamin activity. 5-Methyl-H_4 folate can also be oxidized. Antioxidants (particularly ascorbic acid in the context of milk) can protect folate against destruction. The rate of the oxidative degradation of folate in foods depends on the derivative present and the food itself, particularly its pH, buffering capacity and concentration of catalytic trace elements and antioxidants.

Folate is sensitive to light and may be subject to photodecomposition. Heat treatment influences folate levels in milk. Pasteurization and the storage of pasteurized milks have relatively little effect on the stability of folate but UHT treatments can cause substantial losses. The concentration of oxygen in UHT milk (from the headspace above the milk or by diffusion

through the packaging material) has an important influence on the stability of folate during the storage of UHT milk, as have the concentrations of ascorbate in the milk and of O_2 in the milk prior to heat treatment. Folate and ascorbic acid (section 6.4) are the least stable vitamins in powdered milks.

The heat stability of folate-binding proteins in milk should also be considered in the context of folate in dairy foods. Breast-fed babies require less dietary folate (55 μg folate day^{-1} to maintain their folate status) than bottle-fed infants (78 μg day^{-1}). The difference has been attributed to the presence of active folate-binding proteins in breast milk; folate-binding proteins originally present in milk formulae are heat-denatured during processing. However, a study involving feeding radiolabelled folate to rats together with dried milks prepared using different heat treatments showed no differences in folate bioavailability (Öste, Jägerstad and Andersson, 1997).

6.3.8   Cobalamin and its derivatives (vitamin B₁₂)

Vitamin B_{12} consists of a porphyrin-like ring structure, with an atom of Co chelated at its centre, linked to a nucleotide base, ribose and phosphoric acid (**6.34**). A number of different groups can be attached to the free ligand site on the cobalt. Cyanocobalamin has -CN at this position and is the commercial and therapeutic form of the vitamin, although the principal dietary forms of B_{12} are 5'-deoxyadenosylcobalamin (with 5'-deoxyadenosine at the R position), methylcobalamin (-CH$_3$) and hydroxocobalamin (-OH). Vitamin B_{12} acts as a co-factor for methionine synthetase and methylmalonyl CoA mutase. The former enzyme catalyses the transfer of the methyl group of 5-methyl-H$_4$ folate to cobalamin and thence to homocysteine, forming methionine. Methylmalonyl CoA mutase catalyses the conversion of methylmalonyl CoA to succinyl CoA in the mitochondrion.

Vitamin B_{12} deficiency normally results from indequate absorption rather than inadequate dietary intake. Pernicious anaemia is caused by vitamin B_{12} deficiency; symptoms include anaemia, glossitis, fatigue and degeneration of the peripheral nervous system and hypersensitivity of the skin. The adult RDA and RNI for B_{12} are 2 and 1.5 μg day^{-1}, respectively. Unlike other vitamins, B_{12} is obtained exclusively from animal food sources, such as meat, fish, poultry, eggs, shellfish, milk, cheese and eggs. Vitamin B_{12} in these foods is protein-bound and released by the action of HCl and pepsin in the stomach.

Bovine milk contains, on average, 0.4 μg B_{12} per 100 g. The predominant form is hydroxycobalamin and more than 95% of this nutrient is protein bound. The concentration of B_{12} in milk is influenced by the Co intake of the cow. The predominant source of B_{12} for the cow, and hence the ultimate origin of B_{12} in milk, is biosynthesis in the rumen. Therefore, its concentra-

6.34
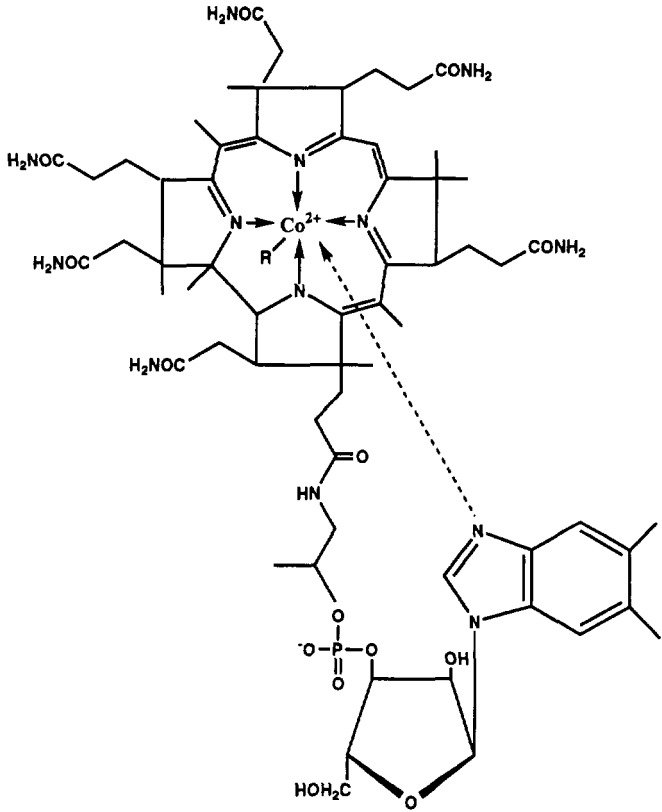
Vitamin B_{12}

tion in milk is not influenced greatly by feed, breed or season. Higher concentrations are found in colostrum than in mature milk.

The B_{12}-binding proteins of human milk have been studied in detail. The principal binding protein (R-type B_{12}-binding protein) has a molecular mass of c. 63 kDa and contains about 35% carbohydrate. Most or all of the B_{12} in human milk is bound to this protein. A second protein, transcobalamin II, is present at low concentrations.

Raw ovine and pasteurized caprine milks contain 0.6 and 0.1 µg B_{12} per 100 g, respectively. Human colostrum contains 0.1 µg per 100 g but the mature milk contains only traces of B_{12}. Concentrations of B_{12} in dairy products (Appendix 6A) include about 0.3 µg per 100 g for cream and 1 µg per 100 g for many cheese varieties. Yogurt contains roughly 0.2 µg per 100 g of this nutrient.

Vitamin B_{12} is stable to pasteurization and storage of pasteurized milks (< 10% loss). UHT heat treatment, and in particular storage of UHT milk, causes greater losses. Storage temperature has a major influence on the

stability of B_{12} in UHT milk. Losses during storage at 7°C are minimal for up to 6 months but at room temperature (the normal storage conditions for UHT milk), losses can be significant after only a few weeks. Oxygen levels in UHT milk do not appear to influence the stability of B_{12}.

6.4 Ascorbic acid (vitamin C)

Ascorbic acid (6.35) is a carbohydrate which can be synthesized from D-glucose or D-galactose by most species with the exception of primates, guinea-pigs, an Indian fruit bat and certain birds. Ascorbate can be oxidized reversibly to dehydroascorbate (6.36) in the presence of transition metal ions, heat, light or mildly alkaline conditions without loss of vitamin activity. Dehydroascorbate can be oxidized irreversibly to 2,3-diketogulonic acid (6.37) with loss of activity. 2,3-Diketogulonic acid can be broken down to oxalic and L-threonic acids and ultimately to brown pigments.

6.35

Ascorbic acid

6.36
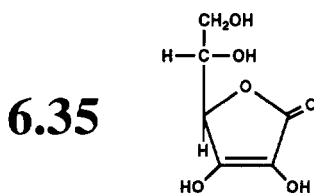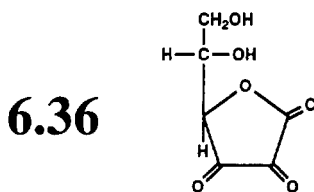
Dehydroascorbic acid

6.37
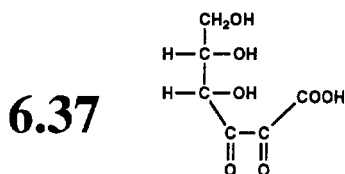
2, 3-Diketogulonic acid

Ascorbic acid is a strong reducing agent and therefore is an important antioxidant in many biological systems. It is also necessary for the activity of the hydroxylase that catalyses the post-translational conversion of proline to hydroxyproline and lysine to hydroxylysine. This post-translational hydroxylation is vital for the formation of collagen, the principal protein in connective tissue. Ascorbate functions to maintain iron in its correct oxidation state and aids in its absorption. Vitamin C also functions in amino acid metabolism, in the absorption of iron and increases resistance to infection. The classical vitamin C deficiency syndrome is scurvy, the symptoms of which include microcytic anaemia, bleeding gums, loose teeth, frequent infections, failure of wounds to heal, muscle degeneration, rough skin, hysteria and depression. The popular scientific literature has suggested major health benefits associated with ascorbate intakes far in excess of the RDA. While many of these claims are spurious, they have led to the widespread use of vitamin C supplements. Toxic effects of vitamin C have been reported and include nausea, abdominal cramps, diarrhoea, urinary tract problems and kidney stones. The RDA and RNI for vitamin C are 60 and 40 mg day^{-1}, respectively. However, ascorbate requirements vary with sex, physical stress and perhaps with age. The richest sources of ascorbic acid are fruits and vegetables; milk is a poor source. Milk contains about 1 mg ascorbate per 100 g, although reported values range from about 0.85 to 2.75 mg per 100 g. These differences reflect the fact that ascorbate levels can be reduced markedly during the handling and storage of milk. A ratio of ascorbate to dehydroascorbate in milk of 4 : 1 has been reported, although this ratio is greatly influenced by oxidation. Some authors have reported seasonal differences in the concentration of vitamin C in milk (highest in winter milk) but the influence of this factor is unclear.

Human milk and colostrum contain about 4 and 7 mg ascorbate per 100 g, respectively. Raw sheep's milk contains more ascorbate (c. 5 mg per 100 g) than bovine milk, although reported values for pasteurized caprine milk are similar to those for cow's milk. Ascorbate is readily oxidized at the pH of milk. The rate of oxidation is influenced by factors including temperature, light, the concentration of oxygen and the presence of catalytic trace elements. Ascorbic acid is of great importance in establishing and maintaining redox equilibria in milk (as discussed in detail in Chapter 11), the protection of folate (section 6.3.7) and in the prevention of oxidized flavour development in milk. The photochemical degradation of riboflavin (section 6.3.2) catalyses the oxidation of ascorbate.

At least 75% of the vitamin C in milk survives pasteurization, and losses during storage of pasteurized milk are usually minimal. However, considerable losses of vitamin C have been reported in milk packaged in transparent containers. The extent of losses during UHT treatment depends on the amount of oxygen present during heat treatment and subsequent storage, and on storage temperature. The concentration of ascorbate in creams and

yogurts is similar to, or a little lower than, that in milk (Appendix 6A); cheese contains only trace amounts.

References

Department of Health (1991) *Dietary Reference Values for Food Energy and Nutrients for the United Kingdom*, Report on Health and Social Subjects No. 40, HMSO, London.

Fox, P.F. and Flynn, A. (1992) Biological properties of milk proteins, in *Advanced Dairy Chemistry*, Vol. 1: *Proteins* (ed. P.F. Fox), Elsevier Applied Science, London, pp. 255–84.

Garrow, J.S. and James, W.P.T. (1993) *Human Nutrition and Dietetics*, Churchill Livingstone, Edinburgh.

Holland, B., Welch, A.A., Unmin, I.D. *et al.* (1991) *McCance and Widdowson's The Composition of Foods*, 5th edn, Royal Society of Chemistry and Ministry of Agriculture, Fisheries and Food, Cambridge and London.

Öste, R., Jägerstad, M. and Andersson I. (1997) Vitamins in milk and milk products, in *Advanced Dairy Chemistry*, Vol. 3: *Lactose, Water, Salts and Vitamins* (ed. P.F. Fox), Chapman & Hall, London, pp. 347–402.

Whitney, E.N. and Rolfes, S.R. (1996) *Understanding Nutrition*, West Publishing, St. Paul.

Suggested reading

Belitz, H.-D. and Grosch, W. (1987) *Food Chemistry*, Springer-Verlag, New York.

Garrow, J.S. and James, W.P.T. (1993) *Human Nutrition and Dietetics*, Churchill Livingstone, Edinburgh.

Jensen, R.G. (ed.) (1995) *Handbook of Milk Composition*, Academic Press, San Diego.

Öste, R., Jägerstad, M. and Andersson I. (1997) Vitamins in milk and milk products, in *Advanced Dairy Chemistry*, Vol. 3: *Lactose, Water, Salts and Vitamins*, (ed. P.F. Fox), Chapman & Hall, London, pp. 347–402.

Whitney, E.N. and Rolfes, S.R. (1996). *Understanding Nutrition*, West Publishing, St. Paul.

Appendices

Appendix 6A   Vitamin and vitamin precursor concentrations (per 100 g) in dairy products (modified from Holland et al., *1991)*

Product	Retinol (μg)	Carotone (μg)	Vitamin D (μg)	Vitamin E (mg)	Thiamin (mg)	Ribo-flavin (mg)	Niacin (mg)	Trp ÷ 60 (mg)	Vitamin B_6 (mg)	Vitamin B_{12} (μg)	Folate (μg)	Panto-thenate (mg)	Biotin (μg)	Vitamin C (mg)
Skimmed milk														
pasteurized	1	Tr	Tr	Tr	0.04	0.18	0.1	0.8	0.06	0.4	6	0.32	2.0	1
UHT, fortified	61	18	0.1	0.02	0.04	0.18	0.1	0.8	0.05	Tr	4	0.33	1.5	35[a]
Whole milk														
pasteurized	52	21	0.03	0.09	0.04	0.17	0.1	0.7	0.06	0.4	6	0.35	1.9	1
summer	62	31	0.03	0.10	0.04	0.17	0.1	0.7	0.06	0.4	4	0.35	1.9	1
winter	41	11	0.03	0.07	0.04	0.17	0.1	0.7	0.06	0.4	7	0.35	1.9	1
sterilized, in														
container	52	21	0.03	0.09	0.03	0.14	0.1	0.8	0.04	0.1	Tr	0.28	1.8	Tr
Channel Island milk														
whole,														
pasteurized	46	71	0.03	0.11	0.04	0.19	0.1	0.9	0.06	0.4	6	0.36	1.9	1
summer	65	115	0.04	0.13	0.04	0.19	0.1	0.9	0.06	0.4	5	0.36	1.9	1
winter	27	27	0.03	0.09	0.04	0.19	0.1	0.9	0.06	0.4	7	0.36	1.9	1
semi-skimmed,														
UHT	14	22	0.01	0.04	0.04	0.19	0.1	0.9	0.05	0.2	1	0.34	1.5	Tr
Dried skimmed														
milk[b] (fortified)	350	5	2.10	0.27	0.38	1.63	1.0	8.5	0.60	2.6	51	3.28	20.1	13
with vegetable														
fat (fortified)	395	15	10.50	1.32	0.23	1.20	0.6	5.5	0.35	2.3	36	2.15	15.0	11
Evaporated milk,														
whole	105	100	3.95[c]	0.19	0.07	0.42	0.2	2.0	0.07	0.1	11	0.75	4.0	1
Goat's milk,														
pasteurized	44	Tr	0.11	0.03	0.04	0.13	0.3	0.7	0.06	0.1	1	0.41	3.0	1
Human milk,														
colostrum	155	(135)	N	1.3	Tr	0.03	0.1	0.7	Tr	0.1	2	0.12	Tr	7
transitional	85	(37)	N	0.48	0.01	0.03	0.1	0.5	Tr	Tr	3	0.20	0.2	6
mature	58	(24)	0.04	0.34	0.02	0.03	0.2	0.5	0.01	Tr	5	0.25	0.7	4
Sheep's milk, raw	83	Tr	0.18	0.11	0.08	0.32	0.4	1.3	0.08	0.6	5	0.45	2.5	5
Fresh whipping														
cream,														
pasteurized														
(39.3% fat)	565	265	0.22	0.86	0.02	0.17	Tr	0.5	0.04	0.2	7	0.22	1.4	1
Cheeses														
Brie	285	210	0.20	0.84	0.04[d]	0.43	0.4	4.5	0.15	1.2	58	0.35	5.6	Tr
Camembert	230	315	(0.18)	0.65	0.05[e]	0.52	1.0	4.9	0.22	1.1	102	0.36	7.6	Tr
Cheddar,														
average	325	225	0.26	0.53	0.03	0.40	0.1	6.0	0.10	1.1	33	0.36	3.0	Tr

Food														
Cheddar-type (15% fat)	165	100	0.11	0.39	0.03	0.53	0.1	7.4	0.13	1.3	56	0.51	3.8	Tr
Cheese spread, plain	275	105	0.17	0.24	0.05	0.36	0.1	3.2	0.08	0.6	19	0.51	3.6	Tr
Cottage cheese plain	44	10	0.03	0.08	0.03	0.26	0.1	3.2	0.08	0.7	27	0.40	3.0	Tr
reduced fat (1.4% fat)	16	4	0.01	0.03	(0.03)	(0.26)	(0.1)	3.1	(0.08)	(0.7)	(27)	(0.40)	(3.0)	Tr
Cream cheese	385	220	0.27	1.00	0.03	0.13	0.1	0.7	0.04	0.3	11	0.27	1.6	Tr
Danish blue	280	250	(0.23)	0.76	0.03	0.41	0.5	4.7	0.12	1.0	50	0.53	2.7	Tr
Edam	175	150	(0.19)	0.48	0.03	0.35	0.1	6.1	0.09	2.1	40	0.38	1.8	Tr
Feta	220	33	0.50	0.37	0.04	0.21	0.2	3.5	0.07	1.1	23	0.36	2.4	Tr
Fromage frais fruit	82	N	0.04	(0.01)	0.02	0.35	0.1	1.6	0.04	1.4	15	N	N	Tr
plain	100	Tr	0.05	0.02	0.04	0.40	0.1	1.6	0.10	1.4	15	N	N	Tr
very low fat (0.2% fat)	3	N	Tr	Tr	(0.03)	(0.37)	(0.1)	1.8	(0.07)	(1.4)	(15)	N	N	Tr
Gouda	245	145	(0.24)	0.53	0.03	0.30	0.1	5.6	0.08	1.7	43	0.32	1.4	Tr
Parmesan	345	210	(0.25)	0.70	0.03	0.44	0.1	9.3	0.13	1.9	12	0.43	3.3	Tr
Processed cheese, plain	270	95	0.21	0.55	0.03	0.28	0.1	4.9	0.08	0.9	18	0.31	2.3	Tr
Stilton, blue	355	185	0.27	0.61	0.03	0.43	0.5	5.3	0.16	1.0	77	0.71	3.6	Tr
Drinking yogurt, UHT	Tr	Tr	Tr	Tr	0.03	0.16	0.1	0.7	0.05	0.2	12	0.19	0.9	0
Low-fat yogurt, plain	8	5	0.01	0.01	0.05	0.25	0.1	1.2	0.09	0.2	17	0.45	2.9	1
Whole-milk yogurt plain	28	21	0.04	0.05	0.06	0.27	0.2	1.3	0.10	0.2	18	0.50	2.6	1
fruit	39	16	(0.04)	(0.05)	0.06	0.30	0.1	1.3	0.07	0.1	10	0.30	2.0	1
Ice-cream dairy, vanilla	115	195	0.12	0.21	0.04	0.25	0.1	0.8	0.08	0.4	7	0.44	2.5	1
non-dairy, vanilla	1	6	Tr	0.84	0.04	0.24	0.1	0.7	0.07	0.5	8	0.43	3.0	1

Tr, Trace; N, nutrient present in significant quantities but there is no reliable information on amount; ( ), estimated value.

[a] Unfortified milk would contain only traces of vitamin C.

[b] Unfortified skimmed milk powder contains approximately 8 μg retinol, 3 μg carotene, Tr vitamin D and 0.01 mg vitamin E per 100 g. Some brands contain as much as 755 μg retinol 10 μg carotene and 4.6 μg vitamin D per 100 g.

[c] This is for fortified product. Unfortified evaported milk contains approximately 0.09 μg vitamin D per 100 g.

[d] The rind alone contains 0.5 mg thiamin per 100 g.

[e] The rind alone contains 0.4 mg thiamin per 100 g.