Folate

Contents

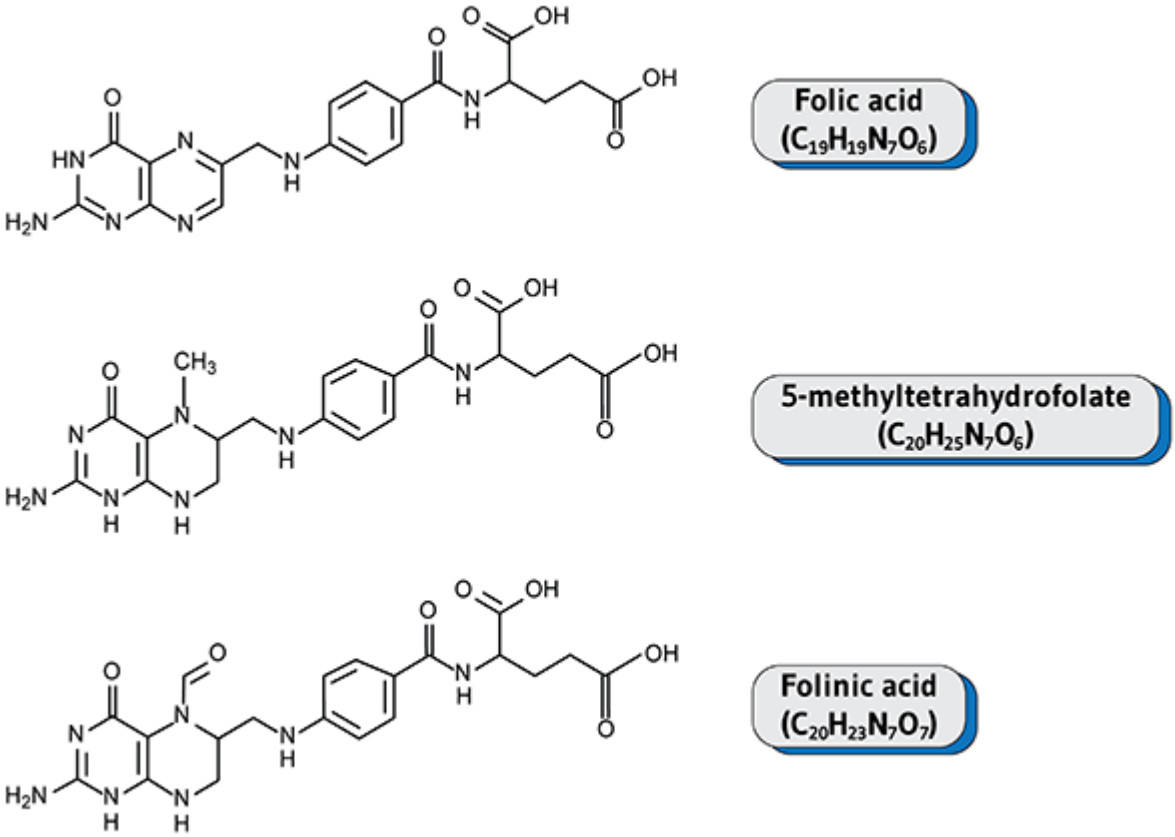
Español

Summary

- Folate is a generic term referring to both natural folates in food and folic acid, the synthetic form used in supplements and fortified food. Folate is critical in the metabolism of nucleic acid precursors and several amino acids, as well as in methylation reactions. *(More information)*
- Severe deficiency in either folate or vitamin B_{12} can lead to megaloblastic anemia, which causes fatigue, weakness, and shortness of breath. Improper treatment of vitamin B_{12}-dependent megaloblastic anemia with high dose supplemental folic acid can potentially delay the diagnosis of vitamin B_{12} deficiency and thus leave the individual at risk of developing irreversible brain damage. *(More information)*
- Folate status is influenced by the presence of genetic variations in folate metabolism, particularly those found in the 5,10-methylenetetrahydrofolate reductase (*MTHFR*) gene. *(More information)*
- Inadequate folate status during early pregnancy increases the risk of congenital anomalies. The introduction of mandatory folic acid fortification of refined grain products in the US in 1998 has reduced the prevalence of neural tube defects (NTDs) in newborns. Yet, folate status is considered inadequate in a majority of women of childbearing age worldwide. Moreover, genetic factors might modify the risk of NTDs by increasing the susceptibility to folate deficiency during pregnancy. Several studies are currently investigating the role of folic acid supplementation in the prevention of congenital anomalies other than NTDs. *(More information)*
- Folate deficiency and elevated concentrations of homocysteine in the blood are associated with increased risk of cardiovascular disease (CVD). Although folic acid supplementation has been proven effective to control circulating homocysteine concentrations, the effect of homocysteine lowering on the incidence of CVD is still debated. *(More information)*
- Low folate status has been linked to increased cancer risk. However, intervention trials with high doses of folic acid have not generally shown any benefit on cancer incidence. *(More information)*
- Prospective cohort studies have reported an inverse association between folate status and colorectal cancer (CRC) risk, especially among men. The relationship between folate status and cancer risk is however complex and requires further research. *(More information)*
- Folate is essential for brain development and function. Low folate status and/or high homocysteine concentrations are associated with cognitive dysfunction in aging (from mild impairments to dementia). Whether supplemental B-vitamins, including folic acid, will have long-term benefits in maintaining cognitive health is not yet known. *(More information)*
- Several autosomal recessive disorders affecting folate transport and metabolism can be treated with high doses of folinic acid, a folate derivative. *(More information)*

Folate is a water-soluble B-vitamin, which is also known as vitamin B_9 or folacin. Naturally occurring folates exist in many chemical forms; folates are found in food, as well as in metabolically active forms in the human body. Folic acid is the major synthetic form found in fortified foods and vitamin supplements. Other synthetic forms include folinic acid (**Figure 1**) and levomefolic acid. Folic acid has no biological activity unless converted into folates (1). In the following discussion, forms found in food or the body are referred to as "folates," while the form found in supplements or fortified food is referred to as "folic acid."

Figure 1. Chemical Structures

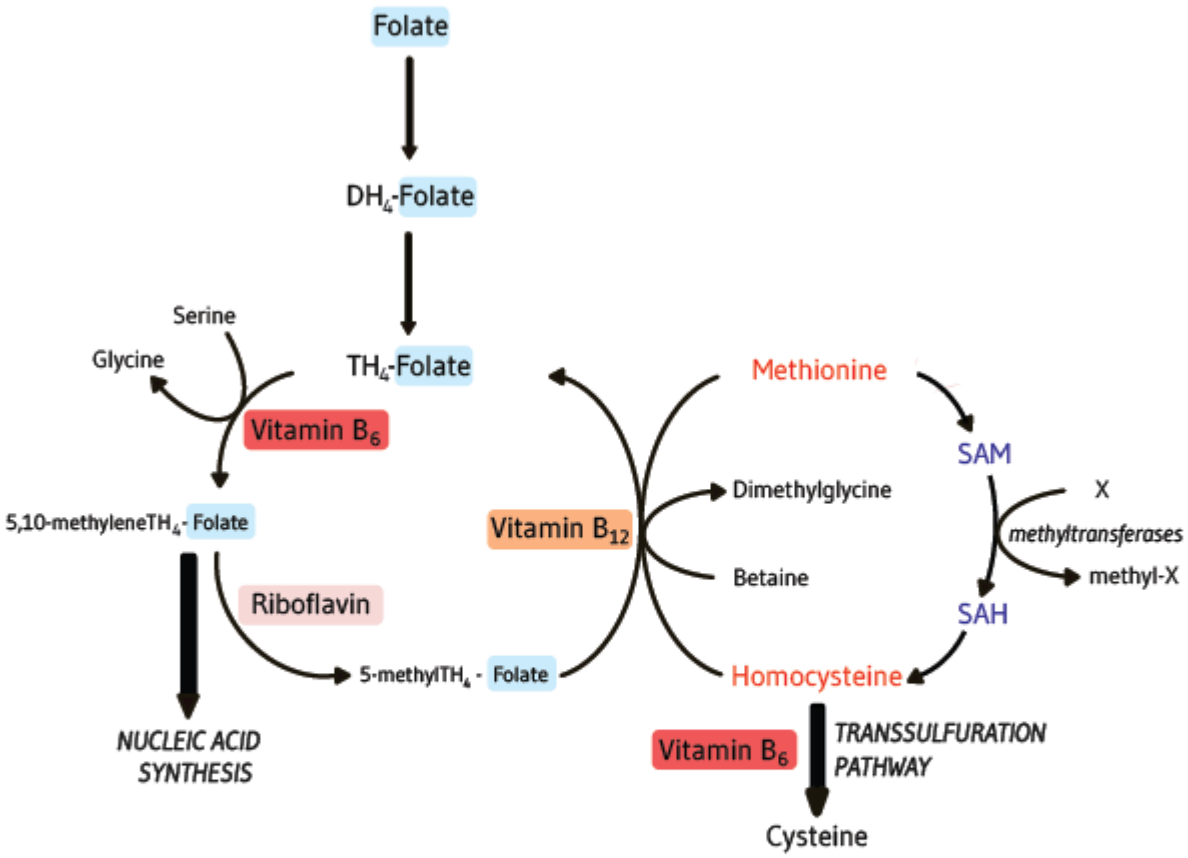


Function

One-carbon metabolism

The only function of folate coenzymes in the body appears to be in mediating the transfer of one-carbon units (2). Folate coenzymes act as acceptors and donors of one-carbon units in a variety of reactions critical to the metabolism of nucleic acids and amino acids (**Figure 2**) (3).

Figure 2. Overview of One-carbon Metabolism



5,10-methylenetetrahydrofolate is required for the synthesis of nucleic acids, and 5-methyltetrahydrofolate is required for the formation of methionine from homocysteine. Methionine, in the form of methyl donor S-adenosylmethionine (SAM), is essential to many biological methylation reactions, including DNA methylation. Methylenetetrahydrofolate reductase (MTHFR) is a riboflavin (FAD)-dependent enzyme that catalyzes the reduction of 5,10-methylenetetrahydrofolate to 5-methyltetrahydrofolate. *SAM, S-adenosylmethionine; SAH, S-adenosylhomocysteine; TH_4-Folate, Tetrahydrofolate.*

Nucleic acid metabolism

Folate coenzymes play a vital role in DNA metabolism through two different pathways. (1) The synthesis of DNA from its precursors (thymidine and purines) is dependent on folate coenzymes. (2) A folate coenzyme is required for the synthesis of methionine from homocysteine, and methionine is required for the synthesis of S-adenosylmethionine (SAM). SAM is a methyl group (one-carbon unit) donor used in most biological methylation reactions, including the methylation of a number of sites within DNA, RNA, proteins, and phospholipids. The methylation of DNA plays a role in controlling gene expression and is critical during cell differentiation. Aberrations in DNA methylation have been linked to the development of cancer (see Cancer).

Amino acid metabolism

Folate coenzymes are required for the metabolism of several important amino acids, namely methionine, cysteine, serine, glycine, and histidine. The synthesis of methionine from homocysteine is catalyzed by methionine synthase, an enzyme that requires not only folate (as 5-methyltetrahydrofolate) but also vitamin

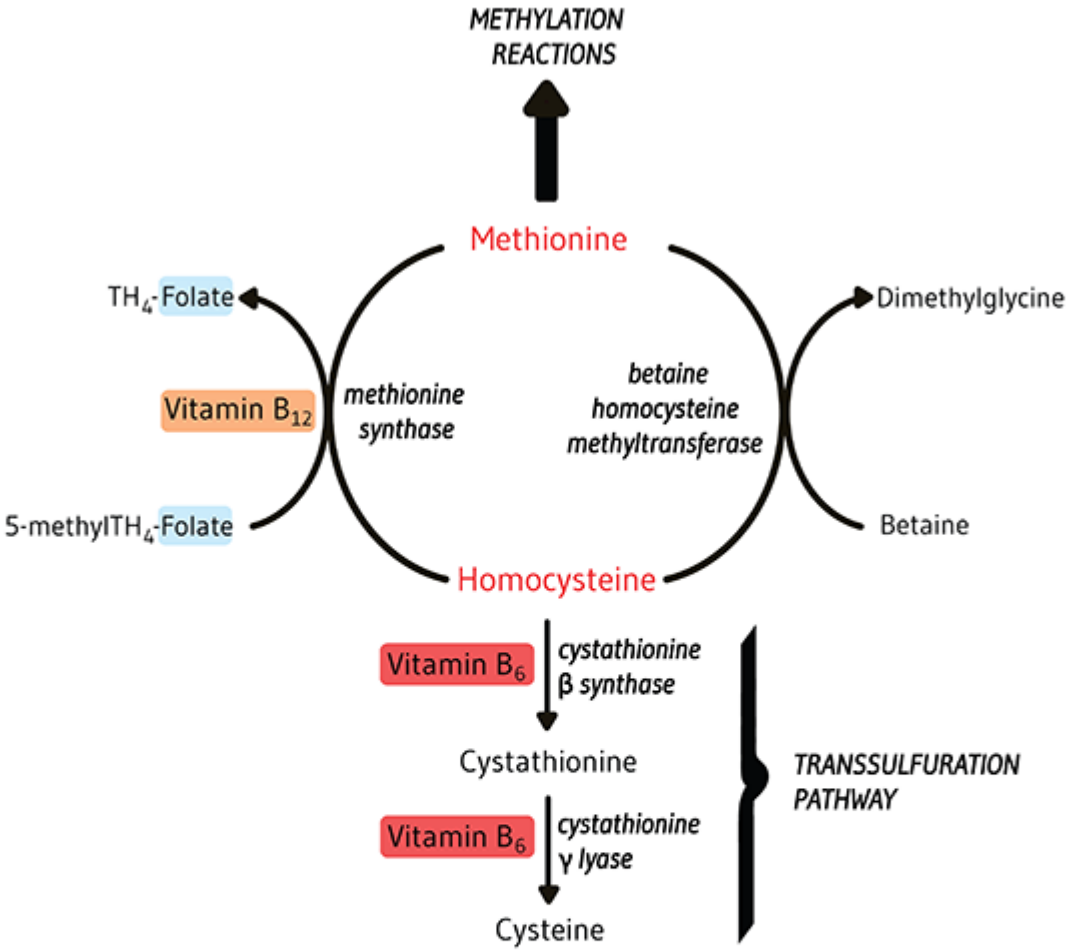
B_{12}. Thus, folate (and/or vitamin B_{12}) deficiency can result in decreased synthesis of methionine and an accumulation of homocysteine. Elevated blood concentrations of homocysteine have been considered for many years to be a risk factor for some chronic diseases, including cardiovascular disease and dementia (see Disease Prevention).

Nutrient interactions

Vitamin B_{12} and vitamin B_6

The metabolism of homocysteine, an intermediate in the metabolism of sulfur-containing amino acids, provides an example of the interrelationships among nutrients necessary for optimal physiological function and health. Healthy individuals utilize two different pathways to metabolize homocysteine (**Figure 3**). One pathway (methionine synthase) synthesizes methionine from homocysteine and is dependent on both folate and vitamin B_{12} as cofactors. The other pathway converts homocysteine to another amino acid, cysteine, and requires two vitamin B_6-dependent enzymes. Thus, the concentration of homocysteine in the blood is regulated by three B-vitamins: folate, vitamin B_{12}, and vitamin B_6 (4). In some individuals, riboflavin (vitamin B_2) is also involved in the regulation of homocysteine concentrations (see the article on Riboflavin).

Figure 3. Homocysteine Metabolism



Homocysteine is methylated to form the essential amino acid methionine in two pathways. The reaction of homocysteine remethylation catalyzed by the vitamin B_{12}-dependent methionine synthase captures a methyl group from the folate-dependent one-carbon pool (5-methyltetrahydrofolate). A second pathway requires betaine (N,N,N-trimethylglycine) as a methyl donor for the methylation of homocysteine catalyzed by betaine homocysteine methyltransferase. The catabolic pathway of homocysteine, known as the transsulfuration pathway, converts homocysteine to the amino acid cysteine via two vitamin B_6 (PLP)-dependent enzymes. Cystathionine β synthase catalyzes the condensation of homocysteine with serine to form cystathionine, and cystathionine is then converted to cysteine, α-ketobutyrate, and ammonia by cystathionine γ lyase. *TH_4-Folate, Tetrahydrofolate.*

Riboflavin

Although less well recognized, folate has an important metabolic interaction with riboflavin. Riboflavin is a precursor of flavin adenine dinucleotide (FAD), a coenzyme required for the activity of the folate-metabolizing enzyme, 5,10-methylenetetrahydrofolate reductase (MTHFR). FAD-dependent MTHFR in turn catalyzes the reaction that generates 5-methyltetrahydrofolate (see **Figure 2** above). This active form of folate is required to form methionine from homocysteine. Along with other B-vitamins, higher riboflavin intakes have been associated with decreased plasma homocysteine concentrations (5). The effects of riboflavin on folate metabolism appear to be greatest in individuals homozygous for the common c.677C>T polymorphism (i.e., TT genotype) in the *MTHFR* gene (see Genetic variation in folate requirements) (6). These individuals (about 10% of adults worldwide) typically present with low folate status, along with elevated homocysteine

concentrations, particularly when folate and/or riboflavin intake is suboptimal. The elevated homocysteine concentration in these individuals, however, is highly responsive to lowering with riboflavin supplementation, confirming the importance of the riboflavin-*MTHFR* interaction (7).

Vitamin C

Vitamin C may limit degradation of natural folate coenzymes and supplemental folic acid in the stomach and thus improve folate bioavailability. A cross-over trial in nine healthy men found that oral co-administration of 5-methyltetrahydrofolic acid (343 µg) and vitamin C (289 mg or 974 mg) was associated with higher concentrations of serum folate compared to 5-methyltetrahydrofolic acid alone (8). Moreover, a recent study suggested that several genetic variations of folate metabolism might influence the effect of vitamin C on folate metabolism (9).

Bioavailability

Dietary folates exist predominantly in the polyglutamyl form (containing several glutamate residues), whereas folic acid—the synthetic vitamin form—is a monoglutamate, containing just one glutamate moiety. In addition, natural folates are reduced molecules, whereas folic acid is fully oxidized. These chemical differences have major implications for the bioavailability of the vitamin such that folic acid is considerably more bioavailable than naturally occurring food folates at equivalent intake levels.

The intestinal absorption of dietary folates is a two-step process that involves the hydrolysis of folate polyglutamates to the corresponding monoglutamyl derivatives, followed by their transport into intestinal cells. There, folic acid is converted into a naturally occurring folate, namely 5-methyltetrahydrofolate, which is the major circulating form of folate in the human body (see **Figure 1** above).

The bioavailability of naturally occurring folates is inherently limited and variable. There is much variability in the ease with which folates are released from different food matrices, and the polyglutamyl "tail" is removed (de-conjugation) before uptake by intestinal cells. Also, other dietary constituents can contribute to instability of labile folates during the processes of digestion. As a result, naturally occurring folates show incomplete bioavailability compared with folic acid. The bioavailability of folic acid, in contrast, is assumed to be 100% when ingested as a supplement, while folic acid in fortified food is estimated to have about 85% the bioavailability of supplemental folic acid.

Of note, folate recommendations in the US and certain other countries are now expressed as Dietary Folate Equivalents (DFEs), a calculation that was devised to take into account the greater bioavailability of folic acid compared to naturally occurring dietary folates (see The Recommended Dietary Allowance).

Transport

Folate and its coenzymes require transporters to cross cell membranes. Folate transporters include the reduced folate carrier (RFC), the proton-coupled folate transporter (PCFT), and the folate receptor proteins, FRα and FRβ. Folate homeostasis is supported by the ubiquitous distribution of folate transporters, although abundance and importance vary among tissues (10). PCFT plays a major role in folate intestinal transport since mutations affecting the gene encoding PCFT cause hereditary folate malabsorption. Defective PCFT also leads to impaired folate transport into the brain (see Disease Treatment). FRα and RFC are also critical for folate transport across the blood-brain barrier when extracellular folate is either low or high, respectively. Folate is essential for the proper development of the embryo and the fetus. The placenta is known to concentrate folate to the fetal circulation, leading to higher folate concentrations in the fetus compared to those found in the pregnant woman. All three types of receptors have been associated with folate transport across the placenta during pregnancy (11).

Deficiency

Causes

Folate deficiency is most often caused by a dietary insufficiency; however, folate deficiency can also occur in a number of other situations. For example, chronic and heavy alcohol consumption is associated with diminished absorption of folate (in addition to low dietary intake), which can lead to folate deficiency (12). Smoking is also associated with low folate status. In one study, folate concentrations in blood were about 15% lower in smokers compared to nonsmokers (13). Additionally, impaired folate transport to the fetus has been described in pregnant women who either smoked or abused alcohol during their pregnancy (14, 15).

Pregnancy is a time when the folate requirement is greatly increased to sustain the demand for rapid cell replication and growth of fetal, placental, and maternal tissue. Conditions such as cancer or inflammation can also result in increased rates of cell division and metabolism, causing an increase in the body's demand for folate (16). Moreover, folate deficiency can result from some malabsorptive conditions, including inflammatory bowel diseases (Crohn's disease and ulcerative colitis) and celiac disease (17). Several medications may also contribute to folate deficiency (see Drug interactions). Finally, a number of genetic diseases affecting folate absorption, transport, or metabolism can cause folate deficiency or impede its metabolic functions (see Disease Treatment).

Symptoms

Clinical folate deficiency leads to megaloblastic anemia, which is reversible with folic acid treatment. Rapidly dividing cells like those derived from bone marrow are most vulnerable to the effects of folate deficiency since DNA synthesis and cell division are dependent on folate coenzymes. When folate supply to the rapidly dividing cells of the bone marrow is inadequate, blood cell division is reduced, resulting in fewer but larger red blood cells. This type of anemia is called megaloblastic or macrocytic anemia, referring to the enlarged, immature red blood cells. Neutrophils, a type of white blood cell, become hypersegmented, a change that can be found by examining a blood sample microscopically. Because normal red blood cells have a lifetime in the circulation of approximately four months, it can take months for folate-deficient individuals to develop the characteristic megaloblastic anemia. Progression of such an anemia leads to a decreased oxygen carrying capacity of the blood and may ultimately result in symptoms of fatigue, weakness, and shortness of breath (1). It is important to point out that megaloblastic anemia resulting from folate deficiency is identical to the megaloblastic anemia resulting from vitamin B_{12} deficiency, and further clinical testing is required to diagnose the true cause of megaloblastic anemia (see Toxicity).

Individuals in the early stages of folate deficiency may not show obvious symptoms, but blood concentrations of homocysteine may increase (see Disease Prevention). Yet, the concentration of circulating homocysteine is not a specific indicator of folate status, as elevated homocysteine can be the result of vitamin B_{12} and other B-vitamin deficiencies, lifestyle factors, and renal insufficiency. Subclinical deficiency is typically detected by measurement of folate concentrations in serum/plasma or in red blood cells.

The Recommended Dietary Allowance (RDA)

Determination of the RDA

Traditionally, the dietary folate requirement was defined as the amount needed to prevent a deficiency severe enough to cause symptoms like anemia. The most recent RDA (1998; **Table 1**) was based primarily on the adequacy of red blood cell folate concentrations at different levels of folate intake, as judged by the absence of abnormal hematological indicators. Red cell folate has been shown to correlate with liver folate stores and is used as an indicator of long-term folate status. Plasma folate reflects recent folate intake and is not a reliable biomarker for folate status. Maintenance of normal blood homocysteine concentrations, an indicator of one-carbon metabolism, was considered only as an ancillary indicator of adequate folate intake.

Because pregnancy is associated with a significant increase in cell division and other metabolic processes that require folate coenzymes, the RDA for pregnant women is considerably higher than for women who are not pregnant (3). However, the prevention of neural tube defects (NTDs) was not considered when setting the RDA for pregnant women. Rather, reducing the risk of NTDs was considered in a separate recommendation for women capable of becoming pregnant (see Disease Prevention), because the crucial events in the development of the neural tube occur before many women are aware that they are pregnant (18).

Dietary Folate Equivalents (DFEs)

When the Food and Nutrition Board of the US Institute of Medicine set the new dietary recommendation for folate, they introduced a new unit, the Dietary Folate Equivalent (DFE) (1). Use of the DFE reflects the higher bioavailability of synthetic folic acid found in supplements and fortified food compared to that of naturally occurring food folates (18).

- 1 microgram (µg) of food folate provides 1 µg of DFEs
- 1 µg of folic acid taken with meals or as fortified food provides 1.7 µg of DFEs
- 1 µg of folic acid (supplement) taken on an empty stomach provides 2 µg of DFEs

For example, a serving of food containing 60 µg of folate would provide 60 µg of DFEs, while a serving of pasta fortified with 60 µg of folic acid would provide 1.7 x 60 = 102 µg of DFEs due to the higher bioavailability of folic acid. A folic acid supplement of 400 µg taken on an empty stomach would provide 800 µg of DFEs. It should be noted that DFEs were determined in studies with adults and whether folic acid in infant formula is more bioavailable than folates in mother's milk has not been studied. Use of DFEs to determine a folate requirement for the infant would not be desirable.

Table 1. Recommended Dietary Allowance for Folate in Dietary Folate Equivalents (DFEs)

Life Stage	Age	Males (µg/day)	Females (µg/day)
Infants	0-6 months	65 (AI)	65 (AI)
Infants	7-12 months	80 (AI)	80 (AI)
Children	1-3 years	150	150
Children	4-8 years	200	200
Children	9-13 years	300	300
Adolescents	14-18 years	400	400
Adults	19 years and older	400	400
Pregnancy	all ages	-	600
Breast-feeding	all ages	-	500

Genetic variation in folate requirements

A common polymorphism or variation in the sequence of the gene for the enzyme, 5, 10-methylenetetrahydrofolate reductase (MTHFR), known as the *MTHFR* c.677C>T polymorphism, results in a thermolabile enzyme (19). The substitution of a cytosine (C) by a thymine (T) at nucleotide 677 in the exon 4 of *MTHFR* gene leads to an alanine-to-valine transition in the catalytic domain of the enzyme. Depending on the population, 20% to 53% of individuals may have inherited one T copy (677C/T genotype), and 3% to 32% of individuals may have inherited two T copies (677T/T genotype) for the *MTHFR* gene (20). MTHFR catalyzes the reduction of 5,10-methylenetetrahydrofolate (5,10-methylene THF) into 5-methyl

tetrahydrofolate (5-MeTHF). The latter is the folate coenzyme required to form methionine from homocysteine (see **Figure 2** above). MTHFR activity is greatly diminished in heterozygous 677C/T (-30%) and homozygous 677T/T (-65%) individuals compared to those with the 677C/C genotype (21). Homozygosity for the mutation (677T/T) is linked to lower concentrations of folate in red blood cells and higher blood concentrations of homocysteine (22, 23). Improving folate nutritional status in elderly women with the T allele reduced plasma homocysteine concentration (24). An important unanswered question about folate is whether the present RDA is enough to compensate for the reduced MTHFR enzyme activity in individuals with at least one T allele, or whether those individuals have a higher folate requirement than the RDA (25).

Disease Prevention

Adverse pregnancy outcomes

Neural tube defects

Fetal growth and development are characterized by widespread cell division. Adequate folate is critical for DNA and RNA synthesis. Neural tube defects (NTDs) arise from failure of embryonic neural tube closure between the 21st and 27th days after conception, a time when many women may not even realize they are pregnant (26). NTDs include various malformations, such as lesions of the brain (e.g., anencephaly, encephalocele) or lesions of the spine (spina bifida), which are devastating and life-threatening (27). The prevalence of NTDs in the United States prior to fortification of food with folic acid was estimated to be 1 per 1,000 pregnancies (1). Results of randomized trials have demonstrated 60% to 100% reductions in NTD cases when women consumed folic acid supplements in addition to a varied diet during the periconceptional period (about one month before and at least one month after conception) (28, 29). The results of these and other studies prompted the US Public Health Service to recommend that all women capable of becoming pregnant consume 400 µg of folic acid daily to prevent NTDs. Women with a previously affected pregnancy were also advised to receive 4,000 µg (4 mg) of folic acid daily in order to reduce NTD recurrence (30). These recommendation were made to all women of childbearing age because adequate folate must be available very early in pregnancy, and because many pregnancies in the US are unplanned (31).

Despite the effectiveness of folic acid supplementation in improving folate status, it appears that globally only 30% of women who become pregnant correctly follow the recommendation, and there is some concern that young women from minority ethnic groups and lower socio-economic backgrounds are the least likely to follow the recommendation (32-34). To decrease the incidence of NTDs, the US FDA implemented legislation in 1998 requiring the fortification of all enriched grain products with 1.4 mg of folic acid per kg of grain (see Sources). The required level of folic acid fortification in the US was initially estimated to provide 100 µg of additional folic acid in the average person's diet, though it probably provides even more due to overuse of folic acid by food manufacturers (25, 35). The National Birth Defects Prevention Network reported about a 30% decrease in the prevalence of NTDs in the US compared to the pre-fortification period, and the post-fortification prevalence of NTDs is 0.69 cases per 1,000 live births and fetal deaths (36).

Also, a genetic component in NTD etiology is evidenced by the increased risk in women with a family history of an NTD and also by variations in risk among ethnicities (37). Moreover, NTD occurrence can be attributed to specific folate-gene interactions. The *MTHFR* c.677C>T polymorphism and other genetic variations can increase the folate requirement and susceptibility for an NTD-affected pregnancy. Prior to the fortification era, a case-control study showed that both red blood cell and serum folate concentrations were significantly lower in pregnant women with the T/T and C/T variants compared to the wild-type C/C genotype (22), suggesting inadequate folate metabolism with specific maternal genotypes. A meta-analysis of 25 case-control studies, including 2,429 case mothers and 3,570 control mothers, showed a positive association between the maternal *MTHFR* c.677C>T polymorphism and NTDs (38). Another *MTHFR* variant, an A-to-C change at position 1298, has also been associated with reduced MTHFR activity and increased NTD risk (39). Individuals heterozygous for both of these *MTHFR* variants (677C/T + 1298A/C) exhibit lower plasma folate and higher

homocysteine concentrations than individuals with 677C/T + 1298A/A (40). Combined genotypes with homozygosity G/G for the reduced folate carrier transporter (*RFC-1*) polymorphism (c.80A>G) could further contribute to NTD occurrence (41). The degree of NTD risk was also assessed with additional *MTHFR* polymorphisms (c.116C>T, c.1793G>A) (42), as well as with mutations affecting other enzymes of the one-carbon metabolism, including methionine synthase (*MTR* c.2756A>G) (43), methionine synthase reductase (*MTRR* c.66A>G) (44), and methylenetetrahydrofolate dehydrogenase (*MTHFD1* c.1958G>A) (45). While maternal genotype can impact pregnancy outcome, it appears that gene-gene interactions between mother and fetus influence it further. The risk of NTD was increased by certain genetic combinations, including maternal (*MTHFR* c.677C>T)-fetal (*MTHFR* c.677C>T) and maternal (*MTRR* c.66A>G)-fetal (*MTHFR* c.677C>T) interactions (43, 44, 46). Finally, vitamin B_{12} status has been associated with NTD risk modification in the presence of specific polymorphisms in one-carbon metabolism (47).

Cardiovascular malformations

Congenital anomalies of the heart are a major cause of infant mortality but also cause deaths in adulthood (48). Using data from the European Registration of Congenital Anomalies and Twins (EUROCAT) database, a case-control study, involving 596 cases and 2,359 controls, found that consumption of at least 400 µg/day of folic acid during the periconceptual period (one month before conception through eight weeks' post-conception, covering the period of embryonic heart development) was associated with an 18% reduced risk of congenital heart defects (49). Recent meta-analyses of 20 to 25 case-control and family-based studies observed positive associations between maternal, fetal, or paternal *MTHFR* c.677C>T variant and incidence of congenital heart defects (50, 51). Additional studies are needed to elucidate the effects of gene-nutrient interactions on the risk of congenital heart defects; however, the currently available research indicates that adequate folate intake may play an important role.

Orofacial clefts

Maternal folate status during pregnancy may influence the risk of congenital anomalies called orofacial clefts, namely cleft lip with or without cleft palate (CL/P) (52). A population-based case-control study in Norway investigated the impact of folic acid supplements in mothers of 377 newborns with CL/P, 196 with cleft palate only (CPO) and 763 controls (53). Although dietary intakes or supplements (during the first three months of pregnancy) on their own did not significantly modify the risk of CL/P, the study reported a 64% lower risk among women taking multivitamin and folic acid (≥400 µg daily) supplements in addition to dietary folates. In the same population, polymorphisms in the cystathionine β-synthase (*CBS*) gene (c.699C>T) or *MTHFR* gene (c.677C>T; when folate intake was below 400 µg/day) appeared protective, while other gene variants in the folate/one-carbon metabolism could not be linked to CL/P (54, 55). However, a recent meta-analysis of 18 studies showed an elevation of CL/P risk with the maternal 677T/T homozygosity (56). Additional studies are needed to evaluate the risk of CL/P while integrating both genetic polymorphism and folate intake parameters. Epidemiological evidence supporting a role for folate in the risk of CPO is lacking.

Other adverse pregnancy outcomes

Low birth weight has been associated with increased risk of mortality during the first year of life and may also influence health outcomes during adulthood (57). A recent systematic review and meta-analysis of eight randomized controlled trials found a positive association between folic acid supplementation and birth weight; no association with length of gestation was observed (58). Additionally, a prospective cohort study of 306 pregnant adolescents associated low folate intakes and maternal folate status during the third trimester of pregnancy with higher incidence of small for gestational age births (birth weight <10th percentile) (59). Moreover, the maternal c.677C>T *MTHFR* genotype and increased homocysteine concentrations, considered an indicator of functional folate deficiency, have been linked to lower birth weights (60).

Elevated blood homocysteine concentrations have also been associated with increased incidence of miscarriage and other pregnancy complications, including preeclampsia and placental abruption (61). A large

retrospective study showed that plasma homocysteine in Norwegian women was strongly related to adverse outcomes and complications, including preeclampsia, premature delivery, and very low birth weight, in previous pregnancies (62). A recent meta-analysis of 51 prospective cohort studies linked the c.677C>T *MTHFR* variant with increased risk of preeclampsia in Caucasian and East Asian populations, reinforcing the notion that folate metabolism may play a role in the condition (63). A large multicenter, randomized, controlled trial, the Folic Acid Clinical Trial (FACT), has been initiated to evaluate whether the daily supplementation of up to 5.1 mg of folic acid throughout pregnancy could prevent preeclampsia and other adverse outcomes (e.g., maternal death, placental abruption, preterm delivery) in high-risk women (64). Adequate folate intake during pregnancy protects against megaloblastic anemia (65). A recent case-control study found a reduction in risk of autism spectrum disorders with daily folic acid consumption of 600 µg or more before and during pregnancy when mother and child carried the c.677C>T *MTHFR* genotype (66).

Thus, it is reasonable to maintain folic acid supplementation throughout pregnancy, even after closure of the neural tube, in order to decrease the risk of other problems during pregnancy. Moreover, recent systematic reviews of observational studies found no evidence of an association between folate exposure during pregnancy and adverse health outcomes in offspring, in particular childhood asthma and allergies (67, 68).

Cardiovascular disease

Homocysteine and cardiovascular disease

The results of more than 80 studies indicate that even moderately elevated concentrations of homocysteine in the blood increase the risk of cardiovascular disease (CVD) (4). Possible predispositions to vascular accidents have also been linked to genetic deficiencies in homocysteine metabolism in certain populations (69). The mechanism by which homocysteine may increase the risk of vascular disease has been the subject of a great deal of research, but it may involve adverse effects of homocysteine on blood clotting, arterial vasodilation, and thickening of arterial walls (70). Although increased homocysteine concentrations in the blood have been consistently associated with increased risk of CVD, it is unclear whether lowering circulating homocysteine will reduce CVD risk (see Folate and homocysteine). Research had initially predicted that a prolonged decrease in serum homocysteine level of 3 micromoles/liter would lower the risk of CVD by up to 25% and be a reasonable treatment goal for individuals at high risk (71, 72). However, the analysis of recent clinical trials of B-vitamin supplementation has shown that lowering homocysteine concentrations did not prevent the occurrence of a second cardiovascular event in patients with existing CVD (73, 74). Consequently, the American Heart Association recommends screening for elevated total homocysteine concentrations only in "high risk" individuals, for example, in those with personal or family history of premature cardiovascular disease, malnutrition or malabsorption syndromes, hypothyroidism, kidney failure, lupus, or individuals taking certain medications (nicotinic acid, theophylline, bile acid-binding resins, methotrexate, and L-dopa.

Folate and homocysteine

Folate-rich diets have been associated with decreased risk of CVD, including coronary artery disease, myocardial infarction (heart attack), and stroke. A study that followed 1,980 Finnish men for 10 years found that those who consumed the most dietary folate had a 55% lower risk of an acute coronary event when compared to those who consumed the least dietary folate (75). Of the three B-vitamins that regulate homocysteine concentrations, folic acid has been shown to have the greatest effect in lowering basal concentrations of homocysteine in the blood when there is no coexisting deficiency of vitamin B_{12} or vitamin B_6 (see Nutrient interactions) (76). Increasing folate intake through folate-rich food or supplements has been found to reduce homocysteine concentrations (77). Besides, blood homocysteine concentrations have declined since the FDA mandated folic acid fortification of the grain supply in the US (25). A meta-analysis of 25 randomized controlled trials, including almost 3,000 subjects, found that folic acid supplementation with 800 µg/day or more could achieve a maximal 25% reduction in plasma homocysteine concentrations. In this meta-analysis, daily doses of 200 µg and 400 µg of folic acid were associated with a 13% and 20% reduction in plasma homocysteine, respectively (78). A supplement regimen of 400 µg of folic acid, 2 mg of vitamin B_6,

and 6 µg of vitamin B_{12} has been advocated by the American Heart Association if an initial trial of a folate-rich diet (see Sources) is not successful in adequately lowering homocysteine concentrations (79).

Several polymorphisms in folate/one-carbon metabolism modify homocysteine concentrations in blood (80). In particular, the effect of the c.677C>T *MTHFR* variant has been examined in relation to folic acid fortification policies worldwide. The analysis of randomized trials, including 59,995 subjects without a history of CVD, revealed that the difference in homocysteine concentrations between T/T and C/C genotypes was greater in low-folate regions compared to regions with food fortification policy (3.12 vs. 0.13 micromoles/liter) (81). Although folic acid supplementation effectively decreases homocysteine concentrations, it is not yet clear whether it also decreases risk for CVD. A recent meta-analysis of 19 randomized clinical trials, including 47,921 subjects with preexisting cardiovascular or renal disease, found that homocysteine lowering through folic acid and other B-vitamin supplementation failed to reduce the incidence of CVD despite significant reductions in plasma homocysteine concentrations (74). Other meta-analyses have confirmed the lack of causality between the lowering of homocysteine and the risk of CVD (80-82), including the risk of stroke (83, 84). Consequently, the American Heart Association removed its recommendation for using folic acid to prevent cardiovascular disease in high-risk women (85). It should be noted that the majority of prevention trials to date have been performed in CVD patients with advanced disease. The evidence supporting a beneficial role for folate and related B-vitamins appears to be strongest for the primary prevention of stroke (86). The introduction of mandatory folic acid fortification has been associated with a decline in stroke-related mortality in North America, adding further support to the potential benefit of enhancing folate status and/or lowering homocysteine in the prevention of stroke (87).

Despite the controversy regarding the role of homocysteine lowering in CVD prevention, some studies have investigated the effect of folic acid supplementation on the development of atherosclerosis, a known risk factor for vascular accidents. The measurement of the carotid intima-media thickness (CIMT) is a surrogate endpoint for early atherosclerosis and a predictor for cardiovascular events (88). The meta-analysis of 10 randomized trials testing the effect of folic acid supplementation showed a significant reduction in CIMT in subjects with chronic kidney diseases and in those at risk for CVD, but not in healthy participants (89). Endothelial dysfunction is a common feature in atherosclerosis and vascular disease. High doses of folic acid (400-10,000 µg/day) have been associated with improvements in vascular health in both healthy and CVD subjects (90). Although recent trials failed to demonstrate any cardiovascular protection from folic acid supplementation, low folate intake is a known risk factor for vascular disease, and more research is needed to explore the role of folate in maintaining vascular health (91).

Cancer

Cancer is thought to arise from DNA damage in excess of ongoing DNA repair and/or the inappropriate expression of critical genes. Because of the important roles played by folate in DNA and RNA synthesis and methylation, it is possible that inadequate folate intake contributes to genome instability and chromosome breakage that often characterize cancer development. In particular, DNA replication and repair are critical for genome maintenance, and the shortage in nucleotides caused by folate deficiency might lead to genome instability and DNA mutations. A decrease in 5,10-methylene THF can compromise the conversion of deoxyuridine monophosphate (dUMP) to deoxythymidine monophosphate (dTMP) by the enzyme thymidylate synthase (TS), causing uracil accumulation and thymine depletion. This could then lead to uracil misincorporation into DNA during replication or repair, and cause DNA damage, including point mutations and strand breaks (92). Since 5,10-methylene THF is also the MTHFR enzyme substrate, it is plausible that a reduction of MTHFR activity with the c.677C>T polymorphism may increase the use of 5,10-methylene THF for thymidylate synthesis and prevent DNA damage. However, this hypothesis might only be valid in a situation of folate deficiency (93). Conversely, it was argued that folic acid supplementation could fuel DNA synthesis, therefore promoting tumor growth. This is supported by the observation that TS can function like a tumor promoter (oncogene), while a reduction in TS activity is linked to a lower risk of cancer (94, 95). Additionally, antifolate molecules that block the thymidylate synthesis pathway are successfully used in cancer therapy (96). Folate also controls the homocysteine/methionine cycle and the pool of S-adenosylmethionine (SAM), the methyl donor for methylation reactions. Thus, folate deficiency may impair

DNA and protein methylation and alter the expression of genes involved in DNA repair, proliferation and cell death. Global DNA hypomethylation, a typical hallmark of cancer, causes genome instability and chromosome breaks (reviewed in 97).

The consumption of at least five servings of fruit and vegetables daily has been consistently associated with a decreased incidence of cancer (98). Fruit and vegetables are excellent sources of folate, which may play a role in their anti-carcinogenic effect. Observational studies have found diminished folate status to be associated with site-specific cancers. While food fortification is mandatory in the US (since 1998; see Sources), concerns about the impact of high folic acid intakes on health have delayed the practice in several other countries (99). However, the most recent meta-analyses of folic acid intervention trials (supplemental doses ranging from 500 to 5,000 µg/day for at least one year) did not show any specific benefit or harm regarding total and site-specific cancer incidence (100, 101).

Colorectal cancer

A pooled analysis of 13 prospective cohort studies, which followed a total of 725,134 individuals for a 7 to 20-year period, revealed a modest, inverse association between dietary and total (from food and supplements) folate intake and colon cancer risk. Specifically, a 2% decrease in colon cancer risk was estimated for every 100 µg/day increase in total folate intake (102). A large US prospective study, which followed 525,488 subjects, ages 50 to 71 years between 1995 and 2006, correlated dietary folate, supplemental folic acid, and total folate intakes with a decreased colorectal cancer (CRC) risk (103). However, when stratified by gender, there was no association between dietary folate intake and CRC risk in women (103, 104). A lack of association between CRC risk and dietary, supplemental, and total folate intakes was also reported in another prospective study that followed more than 90,000 US postmenopausal women during an 11-year period encompassing pre- and post-fortification periods (105). These data suggest the possible influence of gender over CRC risk modification by folate. In the latter study, a significant but transient risk elevation was also observed during the post-fortification era; however, some have asserted that this is unlikely to be caused by increased folate intake due to mandatory fortification (106). Finally, a meta-analysis of 18 case-control studies found a slight reduction in CRC risk with folate from food (107). However, it is important to note that the case-control studies were highly heterogeneous, and that the authors stated that dietary fiber, vitamins, and alcohol intake could have confounded their results. Moreover, the lower limit of the highest quantile of folate intake was highly variable, ranging from 270 to 1,367 µg/day (107).

While most epidemiological research shows a protective effect of folate against colorectal cancer development, it has been suggested that high doses of supplemental folic acid may actually accelerate tumor growth in cancer patients (108). Whereas higher folate status within the normal dietary range is widely considered to be protective against cancer, some investigators remain concerned that exposure to excessively high folic acid intakes may increase the growth of pre-existing neoplasms (108). Several clinical trials addressed the effect of folic acid supplementation in patients with a history of colorectal adenoma, with trials finding a risk reduction or no effect of supplemental folic acid (109-112). A recent meta-analysis of three large randomized controlled trials in high-risk subjects did not demonstrate any increase in colorectal adenoma recurrence in subjects supplemented with 500 or 1,000 µg/day of folic acid for 24 to 42 months when compared with placebo treatment (113).

As suggested earlier, the *MTHFR* 677T/T genotype might prevent uracil misincorporation and protect DNA integrity and stability under low-folate conditions. A meta-analysis of 62 case-control and two cohort studies revealed that while the T/T variant reduces CRC risk by 12% compared to both C/T and C/C genotypes, the risk was decreased by 30% with high (348-1,583 µg/day) versus low total folate intakes (264-450 µg/day), irrespective of the genotype (114). A common polymorphism (c.2756A>G) in the *MTR* gene, which codes for methionine synthase, was also examined in relation with the risk of colorectal adenoma and cancer. Methionine synthase catalyzes the simultaneous conversion of homocysteine and 5-methylene THF into methionine and TFH, respectively. The recent meta-analysis of 27 case-control studies showed no association between *MTR* variant and cancer risk (115).

Although alcohol consumption interferes with the absorption and metabolism of folate (16), one case-control and five prospective cohort studies have reported either reduction in CRC risk among nondrinkers compared to drinkers or a lack of association (107). However, in a large prospective study that followed more than 28,000 male health professionals for 22 years, intake of more than two alcoholic drinks (>30 grams of alcohol) per day augmented CRC risk by 42% during the pre-fortification period. CRC risk was not increased during the post-fortification period, suggesting that it is the combination of high alcohol and low folate intake that might increase CRC risk. Yet, another prospective study that followed more than 69,000 female nurses for 28 years did not report a significant increase in CRC risk with alcohol intake before and after the mandatory folic acid fortification (116). In some studies, individuals who are homozygous for the c.677C>T *MTHFR* polymorphism (T/T) have been found to be at decreased risk for colon cancer when folate intake is adequate. However, when folate intake is low and/or alcohol intake is high, individuals with the (T/T) genotype have been found to be at increased risk of colorectal cancer (117, 118).

Breast cancer

Several prospective cohort and case-control studies investigating whether folate intake affects breast cancer risk have reported mixed results (119). A meta-analysis of 15 prospective studies and one nested case-control study found no relationship with dietary folate intake (120). Moderate alcohol intake has been associated with increased risk of breast cancer in women (121). The results of three prospective studies suggested that increased folate intake may reduce the risk of breast cancer in women who regularly consume alcohol (122-124). Thus, high folate intake might be associated with a risk reduction only in women whose breast cancer risk is raised by alcohol consumption. A very large prospective study in more than 88,000 nurses reported that folic acid intake was not associated with breast cancer in women who consumed less than one alcoholic drink per day. However, in women consuming at least one alcoholic drink per day, folic acid intake of at least 600 µg daily resulted in about half the risk of breast cancer compared with women who consumed less than 300 µg of folic acid daily (124). Nevertheless, whether and how alcohol consumption increases breast cancer risk is still subject to discussion (125, 126). Finally, recent meta-analyses evaluating the influence of polymorphisms in one-carbon metabolism on cancer risk found that specific variants in the gene encoding thymidylate synthase increased the risk of breast cancer in certain ethnic populations (127, 128).

Childhood cancers

The incidence of Wilms' tumors (kidney cancer) and certain types of brain cancers (neuroblastoma, ganglioneuroblastoma, and ependymoma) in children has decreased since the mandatory fortification of the US grain supply in 1998 (129). However, incidence rates were unchanged between the pre- and post-fortification periods for leukemia—a predominant childhood malignancy. Despite earlier studies linking maternal folic acid supplementation during pregnancy with the reduced risk of childhood leukemia, more recent investigations have found little evidence to support a preventive effect of folic acid (130). Several meta-analyses have also found little to no protective effect with *MTHFR* polymorphisms; however, the most recent meta-analysis of 22 case-control studies found a reduction in the risk of acute lymphoblastic leukemia (ALL) with the c.677C>T variant in Caucasians and Asians (131).

Alzheimer's disease and cognitive impairment

Alzheimer's disease (AD) is the most common form of dementia, affecting more than 5 million individuals over 65 years old in the US (132). β-amyloid plaque deposition, Tau protein-forming tangles, and increased cell death in the brain of AD patients have been associated with cognitive decline and memory loss. One study associated increased consumption of fruit and vegetables, which are abundant sources of folate, with a reduced risk of developing dementia and AD in women (133). Through its role in nucleic acid synthesis and methyl donor provision for methylation reactions, folate is critical for normal brain development and function, not only during pregnancy and after birth, but also later in life (134). In one cross-sectional study of elderly women, AD patients had significantly higher homocysteine and lower red blood cell folate concentrations compared to healthy individuals. However, there was no difference in the level of serum folate between

groups, suggesting that long-term folate status, rather than recent folate intake, may be associated with the risk of AD (135).

Several investigators have described associations between increased homocysteine concentrations and cognitive impairment in the elderly (136), but prospective cohort studies have not found higher folate intakes to be associated with improved cognition (137, 138). Higher homocysteine concentrations were found in individuals suffering from dementia, including AD and vascular dementia, compared to healthy subjects (139, 140). Although deficiencies in folate, vitamin B_{12}, and vitamin B_6 could increase homocysteine concentrations, a reduction in vitamin concentrations in the serum of AD patients compared to healthy individuals could not be attributed to decreased vitamin intakes (141). It is not presently clear whether serum homocysteine is a risk factor for developing dementia or simply associated with the cognitive decline. In the last decade, a number of clinical trials have tested the use of B-vitamins to lower homocysteine and prevent or delay cognitive decline. A meta-analysis of nine randomized, placebo-controlled trials of folic acid supplementation (0.2 to 15 mg/day for a median duration of six months) in healthy individuals over 45 years of age failed to find a short-term effect on cognitive functions, including memory, speed, language, and executive functions (142). More recently, a meta-analysis of 19 randomized, placebo-controlled trials of B-vitamin supplementation found no difference in cognitive parameters between the treatment and placebo groups, despite the treatment effectively lowering homocysteine concentrations (143). Inconsistent findings across trials may be due to differences in design and methodology (reviewed in 144).

Nevertheless, a two-year randomized, placebo-controlled trial in 168 elderly subjects with mild cognitive impairment recently described the benefits of a daily regimen of 800 µg of folic acid, 500 µg of vitamin B_{12}, and 20 mg of vitamin B_6 (145, 146). Atrophy of specific brain regions affected by AD was observed in individuals of both groups, and this atrophy correlated with cognitive decline; however, the B-vitamin treatment group experienced a smaller loss of gray matter compared to the placebo group (0.5% vs. 3.7%). A greater benefit was seen in subjects with higher baseline homocysteine concentrations, suggesting the importance of lowering circulating homocysteine in prevention of cognitive decline and dementia. Although encouraging, the effect of B-vitamin supplementation needs to be further studied in larger trials that evaluate long-term outcomes, such as the incidence of AD.

Disease Treatment

Metabolic diseases

Folinic acid (see **Figure 1** above), a tetrahydrofolic acid derivative, is used in the clinical management of rare inborn errors that affect folate transport or metabolism (reviewed in 147). Such conditions are of autosomal recessive inheritance, meaning only individuals receiving two copies of the mutated gene (one from each parent) develop the disease.

Hereditary folate malabsorption

Hereditary folate malabsorption is caused by mutations in the *SLC46A1* gene coding for the folate transporter PCFT and typically affects gastrointestinal folate absorption and folate transport into the brain (148). Patients present with low to undetectable concentrations of folate in serum and cerebrospinal fluid, pancytopenia (low number of all blood cells), impaired immune responses that increase susceptibility to infections, and a general failure to thrive (149). Neurologic symptoms, including seizures, have also been observed (150). Clinical improvements have been recorded following parenteral provision of folinic acid (151).

Cerebral Folate Deficiency (CFD) syndrome

CFD is characterized by low levels of folate coenzymes in cerebrospinal fluid despite normal concentrations of folate in blood. Folate transport across the blood-brain barrier is compromised in CFD and has been linked

either to the presence of antibodies blocking the folate receptor FRα or to mutations in the *FOLR1* gene encoding FRα (152, 153). Neurologic abnormalities, along with visual and hearing impairments, have been described in children with CFD; autism spectrum disorder (ASD) is present in some cases. Folinic acid (also known as leucovorin) can enter the brain and normalize the level of folate coenzymes and has been shown to normalize folate concentrations and improve various social interactions in CFD, including mood, behavior, and verbal communication in children with ASD (152, 154, 155).

Dihydrofolate reductase (DHFR) deficiency

DHFR is the NADPH-dependent enzyme that catalyzes the reduction of dihydrofolic acid (DHF) to tetrahydrofolic acid (THF). DHFR is also required to convert folic acid to DHF. DHFR deficiency is characterized by megaloblastic anemia and cerebral folate deficiency causing intractable seizures and mental deficits. Although folinic acid treatment can alleviate the symptoms of DHFR deficiency, early diagnosis is essential to prevent irreversible brain damage and improve clinical outcomes (156, 157).

Sources

Food sources

Green leafy vegetables (foliage) are rich sources of folate and provide the basis for its name. Citrus fruit juices, legumes, and fortified foods are also excellent sources of folate (1); the folate content of fortified cereal varies greatly. A number of folate-rich foods are listed in **Table 2**, along with their folate content in micrograms (µg). For more information on the nutrient content of specific foods, search the USDA food composition database.

Table 2. Some Food Sources of Folate and Folic Acid

Food	Serving	Folate (µg DFEs)
Lentils (mature seeds, cooked, boiled)	½ cup	179
Garbanzo beans (chickpeas, cooked, boiled)	½ cup	141
Asparagus (cooked, boiled)	½ cup (~6 spears)	134
Spinach (cooked, boiled)	½ cup	131
Lima beans (large, mature seeds, cooked, boiled)	½ cup	78
Orange juice (raw)	6 fl. oz.	56

*To help prevent neural tube defects, the US FDA required the addition of 1.4 milligrams (mg) of folic acid per kilogram (kg) of grain to be added to refined grain products, which are already enriched with niacin, thiamin, riboflavin, and iron, as of January 1, 1998. The addition of nutrients to food in order to prevent a nutritional deficiency or restore nutrients lost in processing is known as fortification. The FDA initially estimated that this level of fortification would increase dietary intake by an average of 100 µg folic acid/day (26). However, further evaluations based on observational studies suggested increases twice that predicted by the FDA (35). The prevalence of low folate concentrations in both serum and red blood cells is currently below 1% in the US population, compared to 24% and 3.5%, respectively, before the fortification period (158).

Spaghetti (enriched, cooked)	1 cup	167*
White rice (enriched, cooked)	1 cup	153*
Bread (enriched)	1 slice	84*

*To help prevent neural tube defects, the US FDA required the addition of 1.4 milligrams (mg) of folic acid per kilogram (kg) of grain to be added to refined grain products, which are already enriched with niacin, thiamin, riboflavin, and iron, as of January 1, 1998. The addition of nutrients to food in order to prevent a nutritional deficiency or restore nutrients lost in processing is known as fortification. The FDA initially estimated that this level of fortification would increase dietary intake by an average of 100 µg folic acid/day (26). However, further evaluations based on observational studies suggested increases twice that predicted by the FDA (35). The prevalence of low folate concentrations in both serum and red blood cells is currently below 1% in the US population, compared to 24% and 3.5%, respectively, before the fortification period (158).

Supplements

The principal form of supplementary folate is folic acid. It is available in single-ingredient and combination products, such as B-complex vitamins and multivitamins. Doses of 1 mg or greater require a prescription (159). Additionally, folinic acid, a tetrahydrofolic acid derivative, is used to manage certain metabolic diseases (see Disease Treatment). Further, the US FDA has approved the supplementation of folate in oral contraceptives. The addition of levomefolate calcium (the calcium salt of MeTHF; 451 µg/tablet) to oral contraceptives is intended to raise folate status in women of childbearing age (160). According to a US national survey, only 24% of non-pregnant women aged 15-44 years are meeting the current recommendation of 400 µg/day of folic acid (161).

Safety

Toxicity

No adverse effects have been associated with the consumption of excess folate from food. Concerns regarding safety are limited to synthetic folic acid intake. Deficiency of vitamin B_{12}, though often undiagnosed, may affect a significant number of people, especially older adults (see the article on Vitamin B_{12}). One symptom of vitamin B_{12} deficiency is megaloblastic anemia, which is indistinguishable from that associated with folate deficiency (see Deficiency). Large doses of folic acid given to an individual with an undiagnosed vitamin B_{12} deficiency could correct megaloblastic anemia without correcting the underlying vitamin B_{12} deficiency, leaving the individual at risk of developing irreversible neurologic damage. Such cases of neurologic progression in vitamin B_{12} deficiency have been mostly seen at folic acid doses of 5,000 µg (5 mg) and above. In order to be very sure of preventing irreversible neurological damage in vitamin B_{12}-deficient individuals, the Food and Nutrition Board of the US Institute of Medicine advises that all adults limit their intake of folic acid (supplements and fortification) to 1,000 µg (1 mg) daily (**Table 3**). The Board also noted that vitamin B_{12} deficiency is very rare in women in their childbearing years, making the consumption of folic acid at or above 1,000 µg/day unlikely to cause problems (1); however, there are limited data on the effects of large doses.

Table 3. Tolerable Upper Intake Level (UL) for Folic Acid

Age Group	UL (µg/day)

*Source of intake should be from food and formula only.

Infants 0-12 months	Not possible to establish*
Children 1-3 years	300
Children 4-8 years	400
Children 9-13 years	600
Adolescents 14-18 years	800
Adults 19 years and older	1,000
*Source of intake should be from food and formula only.	

The saturation of DHFR metabolic capacity by oral doses of folic acid has been associated with the appearance of unmetabolized folic acid in blood (162). Hematologic abnormalities and poorer cognition have been associated with the presence of unmetabolized folic acid in vitamin B_{12}-deficient older adults (≥60 years) (163, 164). A small study conducted in postmenopausal women also raised concerns about the effect of exposure to unmetabolized folic acid on immune function (165). In a small, randomized, open-label trial in 38 women of reproductive age receiving 30 weeks of daily multivitamin supplements, daily supplementation with either 1.1 mg or 5 mg of folic acid resulted in the transient appearance of unmetabolized folic acid in blood over the first 12 weeks of supplementation (166). However, unmetabolized folic acid concentrations returned to baseline levels at the end of the study, suggesting that adaptive mechanisms eventually converted folic acid to reduced forms of folate. Nonetheless, the use of supplemental levomefolate (5-methyl THF) may provide an alternative to prevent the potential negative effects of unconverted folic acid in older adults.

Drug interactions

When nonsteroidal anti-inflammatory drugs (NSAIDs), such as aspirin or ibuprofen, are taken in very large therapeutic dosages (i.e., to treat severe arthritis), they may interfere with folate metabolism. In contrast, routine use of NSAIDs has not been found to adversely affect folate status. The anticonvulsant, phenytoin, has been shown to inhibit the intestinal absorption of folate, and several studies have associated decreased folate status with long-term use of the anticonvulsants, phenytoin, phenobarbital, and primidone (167). However, few studies controlled for differences in dietary folate intake between anticonvulsant users and nonusers. Also, taking folic acid at the same time as the cholesterol-lowering agents, cholestyramine and colestipol, may decrease the absorption of folic acid (159). Methotrexate is a folate antagonist used to treat a number of diseases, including cancer, rheumatoid arthritis, and psoriasis. Some of the side effects of methotrexate are similar to those of severe folate deficiency, and supplementation with folic or folinic acid is used to reduce antifolate toxicity. Other antifolate molecules currently used in cancer therapy include aminopterin, pemetrexed, pralatrexate, and raltitrexed (96). Further, a number of other medications have been shown to have antifolate activity, including trimethoprim (an antibiotic), pyrimethamine (an antimalarial), triamterene (a blood pressure medication), and sulfasalazine (a treatment for ulcerative colitis). Early studies of oral contraceptives (birth control pills) containing high doses of estrogen indicated adverse effects on folate status; however, this finding has not been supported in more recent studies that used low-dose oral contraceptives and controlled for dietary folate (168).

Linus Pauling Institute Recommendation

The available scientific evidence shows that adequate folate intake prevents neural tube defects and other poor outcomes of pregnancy; is helpful in lowering the risk of some forms of cancer, especially in genetically susceptible individuals; and may lower the risk of cardiovascular disease. The Linus Pauling Institute recommends that adults take a daily multivitamin/mineral supplement, which typically contains 400 µg of folic acid, the Daily Value (DV). Even with a larger than average intake of folic acid from fortified food, it is

unlikely that an individual's daily folic acid intake would regularly exceed the tolerable upper intake level of 1,000 µg/day established by the Institute of Medicine (see Safety).

Older adults (>50 years)

The recommendation for 400 µg/day of supplemental folic acid as part of a daily multivitamin/mineral supplement, in addition to a folate-rich diet, is especially important for older adults because blood homocysteine concentrations tend to increase with age (see Disease Prevention).

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The 2014 update of this article was underwritten, in part, by a grant from Bayer Consumer Care AG, Basel, Switzerland.

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