

Betaine in human nutrition^{1,2}

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ABSTRACT

Betaine is distributed widely in animals, plants, and microorganisms, and rich dietary sources include seafood, especially marine invertebrates ($\approx 1\%$); wheat germ or bran ($\approx 1\%$); and spinach ($\approx 0.7\%$). The principal physiologic role of betaine is as an osmolyte and methyl donor (transmethylation). As an osmolyte, betaine protects cells, proteins, and enzymes from environmental stress (eg, low water, high salinity, or extreme temperature). As a methyl donor, betaine participates in the methionine cycle—primarily in the human liver and kidneys. Inadequate dietary intake of methyl groups leads to hypomethylation in many important pathways, including 1) disturbed hepatic protein (methionine) metabolism as determined by elevated plasma homocysteine concentrations and decreased S-adenosylmethionine concentrations, and 2) inadequate hepatic fat metabolism, which leads to steatosis (fatty accumulation) and subsequent plasma dyslipidemia. This alteration in liver metabolism may contribute to various diseases, including coronary, cerebral, hepatic, and vascular diseases. Betaine has been shown to protect internal organs, improve vascular risk factors, and enhance performance. Databases of betaine content in food are being developed for correlation with population health studies. The growing body of evidence shows that betaine is an important nutrient for the prevention of chronic disease. *Am J Clin Nutr* 2004;80:539–49.

KEY WORDS Betaine, osmolyte, methylation, homocysteine, lipotrope, liver, heart

INTRODUCTION

Betaine is found in microorganisms, plants, and animals and is a significant component of many foods (1–10), including wheat, shellfish, spinach, and sugar beets. Betaine is a zwitterionic quaternary ammonium compound that is also known as trimethylglycine, glycine betaine, lcyne, and oxynurine. It is a methyl derivative of the amino acid glycine with a formula of $(\text{CH}_3)_3\text{N}^+\text{CH}_2\text{COO}^-$ and a molecular weight of 117.2, and it has been characterized as a methylamine because of its 3 chemically reactive methyl groups (11). Betaine was first discovered in the juice of sugar beets (*Beta vulgaris*) in the 19th century and was subsequently found in several other organisms. The physiologic function of betaine is either as an organic osmolyte to protect cells under stress or as a catabolic source of methyl groups via transmethylation for use in many biochemical pathways (12).

The principle role for betaine in plants and microorganisms is to protect cells against osmotic inactivation (13). Exposure to

drought, high salinity, or temperature stress triggers betaine synthesis in mitochondria, which results in its accumulation in the cells. Betaine is a compatible osmolyte that increases the water retention of cells, replaces inorganic salts, and protects intracellular enzymes against osmotically induced or temperature-induced inactivation (11, 14–19). For example, spinach is grown in saline soil, and betaine can accumulate in amounts of up to 3% of fresh weight. This enables the chloroplasts to photosynthesize in the presence of high salinity (20).

Betaine has been used as a dietary feed supplement in animal nutrition for >50 y, and this use has provided useful insights into human nutrition. Betaine is added to farmed fish feed as an osmolyte to protect fish from the stress of moving from low to high salinity. Salmon liver mitochondria actively take up betaine when exposed to osmotic stress, and metabolic activity would be reduced to a much greater extent if betaine were not present (13). Betaine protects chick intestinal cells from coccidia infection, alleviates symptoms, and improves performance (21–23). Coccidiosis affects gut ionic balance and intestinal morphology, which leads to maldigestion, malabsorption, and dehydration. As a methyl donor, betaine provides the one-carbon units that can spare the amount of dietary methionine and choline required for optimal nutrition. For example, betaine improves growth and the efficiency of food utilization and reduces body fat in pigs and chicks (23–27).

Humans obtain betaine from foods that contain either betaine or choline-containing compounds. Betaine is present in foods in variable amounts that are generally related to growing and osmotic stress conditions. Some examples of food with high betaine content are shown in **Table 1**, and we estimate (SAS Craig, Danisco USA Inc, unpublished observations, 2004) that dietary intake of betaine ranges from an average of 1 g/d to a high of 2.5 g/d (for a diet high in whole wheat and seafood). The principle metabolic fate of choline is via irreversible oxidation to betaine in the liver and kidney (28–32) via a two-step process (**Figure 1**). First, choline is oxidized to betaine aldehyde by the enzyme choline dehydrogenase. This enzyme can also convert betaine aldehyde to betaine in the presence of NAD^+ (33). Choline dehydrogenase activity occurs in the mitochondria, on the matrix side of the inner membrane (34–36). Betaine aldehyde is then

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TABLE 1Food items with high betaine content¹

Food item	Betaine content
	mg/100 g
Wheat bran	1339
Wheat germ	1241
Spinach	600–645
Beets	114–297
Pretzels	237
Shrimp	219
Wheat bread	201
Crackers	49–199

¹ Data from reference 3.

oxidized to betaine by the NAD⁺-dependent enzyme betaine aldehyde dehydrogenase both in mitochondria and in the cytosol (37). The remainder of dietary choline is used to make acetylcholine and phospholipids such as phosphatidylcholine. A diet of normal foods is estimated to deliver 1 g choline/d (38).

A US Department of Agriculture database of the choline and betaine content of food has been developed (3). The betaine content of foods was found to vary when different sources of individual foods were analyzed and when different cooking methods were used; for example, boiling led to the greatest loss of betaine (39). Comprehensive databases will provide a more accurate determination of the dietary intake patterns of betaine and can be used in epidemiologic studies to establish any correlation with disease risk reduction. Earlier studies hypothesized that betaine in red wine (40) and whole grain (41) may protect against coronary artery disease. The US Department of Agriculture database is therefore seen as a high priority for further studies (42). Betaine can also be added to the diet as a supplement. It is

the primary nitrogenous component of sugar beets and is commercially extracted from molasses by water-based chromatographic separation and crystallization.

INTESTINAL ABSORPTION AND METABOLISM

Cellular absorption of betaine has been described in many organisms from bacteria to vertebrates (43–50). Accumulation is via active Na⁺ or Cl[−] coupled and passive Na⁺ independent transport systems. As an *N*-methylated amino acid, betaine is transported by amino acid transport systems—mainly by betaine γ -aminobutyric acid transport and amino acid transport system A (51–54). Animal studies showed rapid postprandial absorption of betaine in the small intestine via the duodenum (22, 50).

Human studies on betaine intake showed rapid absorption and distribution, with a peak increase in serum betaine at ≈ 1 –2 h (55–57). Resting concentrations of betaine in human serum range from ≈ 20 to 70 $\mu\text{mol/L}$ (55–60), with concentrations similar in diabetic subjects but lower in subjects with renal disease (60). High concentrations of betaine are found in human and animal neonates, probably because of choline metabolism (61). Betaine is mainly eliminated by metabolism, not excretion, even at relatively high doses of 100 mg/kg body wt (56). However, urinary excretion of betaine is elevated in subjects with renal disease or diabetes (60, 62–64). Subacute and subchronic rat studies determined that betaine is nontoxic at all doses studied (0–5% of the diet) (65). At high doses, red blood cell physiology was mildly perturbed, and variations in findings between studies were explained as a difference in chow formulation. The authors concluded that betaine is safe at a daily intake of 9–15 g (average of 12 g).

Betaine is catabolized via a series of enzyme reactions that occur mainly in the mitochondria of liver and kidney cells. These

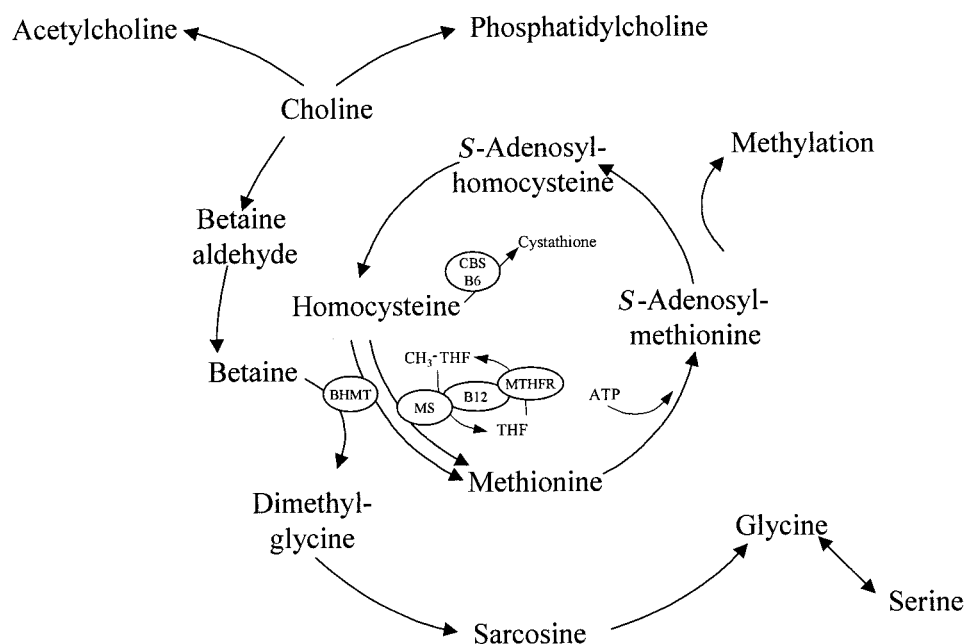


FIGURE 1. Betaine and transmethylation in the methionine cycle. B6, vitamin B-6; B12, vitamin B-12 (cobalamin); BHMT, betaine homocysteine methyltransferase; CBS, cystathionine β -synthase; MS, methionine synthase; MTHFR, methylenetetrahydrofolate reductase; THF, tetrahydrofolate; CH₃-THF, 5-methyltetrahydrofolate.

transmethylation reactions involve the transfer of methyl (one-carbon) groups via the methionine cycle (Figure 1) in vital biological processes (66–76). The conversion of homocysteine to methionine is important to conserve methionine, detoxify homocysteine, and produce *S*-adenosylmethionine (SAM) (77). Elevated total homocysteine concentrations and low SAM concentrations have been associated with chronic disease (discussed in detail later). The formation of methionine from homocysteine can occur either via betaine or via 5-methyltetrahydrofolate (CH₃-THF). Animal studies have shown that both pathways are equally important (78, 79) and that betaine is a vital methylating agent (80, 81). Betaine transfers a methyl group via the enzyme betaine homocysteine methyl transferase (BHMT) to become dimethylglycine. The CH₃-THF reaction utilizes the enzyme methionine synthase to transfer a methyl group to the cofactor cobalamin (vitamin B-12), forming methylcobalamin. Then methylcobalamin transfers the methyl to homocysteine to form methionine. Methylene-tetrahydrofolate reductase (MTHFR) is involved in reforming CH₃-THF from methylene-THF. In addition, homocysteine can be metabolized via transsulfuration to form cystathionine. This is catalyzed by cystathionine β -synthase (CBS), which utilizes vitamin B-6 (pyridoxine) as a cofactor. BHMT, formerly called betaine transmethylase, is a zinc metalloenzyme in a distorted barrel shape (82–84) with an optimum pH of 7.8 (85) that occurs in the human liver, kidney, and optic lens (85–89). Liver BHMT concentrations increase when rats are fed diets supplemented with betaine or choline (90), showing an adaptive change in the catabolism of betaine. BHMT is expressed in the kidney cortex and in liver hepatocytes in humans, pigs, and rats (91).

Betaine increases serum methionine, transmethylation rate, homocysteine remethylation, and methionine oxidation in healthy adults (92). Animals injected with betaine (93) show a dose-dependent increase in red blood cell SAM—a direct methyl donor in many important pathways, including synthesis of proteins, creatine, phospholipids, hormones, polyamines, carnitine, adrenaline, and DNA methylation. Thus, transmethylation metabolic pathways closely interconnect choline, betaine, methionine, CH₃-THF, and vitamins B-6 and B-12, intersecting at the formation of methionine from homocysteine. Perturbing the metabolism of one of these pathways results in compensatory changes in the others (94). For example, methylene-THF can be regenerated via serine or the betaine metabolites dimethylglycine, sarcosine, and glycine (55, 95).

OSMOLYTE

If betaine is not catabolized, it is used as an organic osmolyte. The regulation of cellular hydration state, and therefore cell volume, is important for maintenance of cell function. Sensitive metabolic pathways include protein turnover, amino acid and ammonia metabolism, carbohydrate and fatty acid metabolism, plasma membrane transport, bile excretion, pH control, and gene expression (96). Cells adapt to external osmotic stress by accumulating low-molecular-weight inorganic ions (eg, sodium, potassium, and chloride) and organic osmolytes (eg, methylated amines, certain amino acids, and sugar alcohols). However, the increase in intracellular concentrations of inorganic ions is limited because of their destabilizing effect on protein structure and enzyme function (43, 97–100). Betaine is nonperturbing to cellular metabolism, highly compatible with enzyme function, and stabilizes cellular metabolic function under different kinds of stress (7, 11, 37) in various organisms and animal tissues (101–103). Mechanistic studies showed that there is little or no binding

of betaine to protein surfaces, allowing cells to control the surface tension of water without affecting the ionic strength of the environment, eg, stabilization of lipase (104). Betaine is the most effective osmolyte studied for hydration of albumin (17), forming almost a complete monolayer of water around the protein, and betaine can maintain hemoglobin solvation (105).

There are many examples of the osmolyte function of betaine. It accumulates in the kidney (106–111) from exogenous origin rather than *de novo* synthesis (47, 112, 113) to protect cells from high concentrations of electrolytes (114) and urea (115). Betaine modulates immune function in osmotically stressed liver macrophages (Kupffer cells) via tumor necrosis factor α release (116), phagocytosis (117), and suppression of prostaglandin formation and cyclooxygenase 2 expression (118). It regulates water balance and movement across the intestinal epithelium (119). Betaine regulates erythrocyte (red blood cell) membrane ATPases via conformational changes, which results in cell volume control (120). It protects skeletal muscle myosin ATPases and prevents myosin structural changes due to urea (121). Betaine also protects early preimplantation embryos *in vitro* (122) and prevents apoptosis (programmed cell death) due to hypertonic stress in porcine pulmonary arterial endothelial cells (123). Therefore, this osmotic adaptation to stress helps a variety of cells and organs continue to function and protects against premature apoptosis.

LIVER HEALTH

Nonalcoholic fatty liver disease (NAFLD) is a relatively recent term that describes a progressive range of liver pathologies from simple steatosis (fat accumulation) to steatohepatitis (fatty inflammation), fibrosis (excessive fibrous tissue), and cirrhosis (serious liver damage) (124). NAFLD affects $\approx 20\%$ of the general global population, $\approx 50\%$ of diabetic subjects, $>50\%$ of obese persons, and 90% of morbidly obese persons (125). It is now recognized to be the hepatic manifestation of the metabolic syndrome (126). Alterations in hepatic transmethylation may contribute to various diseases, including coronary, cerebral, hepatic, and vascular diseases (127). One manifestation, hepatic steatosis (fatty liver), is a common result of obesity, consumption of a high-fat diet, insulin resistance, diabetes, alcohol consumption, and other liver damage (128).

Betaine is a lipotrope—something that prevents or reduces accumulation of fat in the liver. Studies on healthy and diabetic animals show that a high-fat diet leads to hepatic steatosis that is prevented by ingestion of betaine or choline (129–131). Hepatic steatosis also occurs as result of a sucrose-only diet (130) or a diet low in both lipotrope and fat (132). Betaine can prevent and cure cirrhosis in rats (133, 134), mobilize hepatic cholesterol and phospholipids in rats fed a high-cholesterol diet (135), treat hyperlipidemia (136), and be utilized in the synthesis of carnitine (137). Dietary betaine enhances the secretion of VLDL via methylation of phosphatidylethanolamine (138) to form phosphatidylcholine. Phosphatidylethanolamine *N*-methyltransferase knockout mice (139) develop hepatic steatosis and abnormal choline metabolite concentrations despite ingesting a recommended dietary intake of choline. This highlights the importance of the SAM route to maintain liver health. One hypothesis regarding the lipotropic properties of betaine is that it contains an electrophilic methyl group that ameliorates pathologic states induced by reductive and oxidative stress (140).

Humans with diabetes exhibit hepatic steatosis, and early studies showed that dietary betaine improves liver function (141–143), including a slight decrease in plasma cholesterol and lipids, a substantial decrease in plasma bilirubin, a reduction in liver size, and better diabetic control. Several reviews (125, 128, 144–149) recommend using betaine to treat NAFLD, including nonalcoholic steatohepatitis (NASH). A 1-y pilot study (150) showed that betaine (20 g/d) is safe and well tolerated and leads to significant biochemical and histologic improvement in patients with NASH. This includes improvements in serum concentrations of liver enzymes (aspartate aminotransferase and alanine aminotransferase), in steatosis, in necroinflammatory grade, and in stage of fibrosis. Betaine glucuronate supplementation (in combination with diethanolamine glucuronate and nicotinamide ascorbate) of patients with NASH lowers hepatic steatosis and significantly reduces liver transaminases (alanine aminotransferase, aspartate aminotransferase, and γ -glutamyltransferase) (151).

Betaine protects the rat liver from toxins such as chloroform (54, 152, 153), lipopolysaccharide (154), methotrexate (155, 156), and carbon tetrachloride (157–159). Specific effects include reduced hepatic lipidosis and necrosis; improved morphology of mitochondria, rough endoplasmic reticulum, Golgi complexes, and nuclear DNA; increased BHMT and SAM; and decreased alanine aminotransferase. Betaine also protects against bile-induced apoptosis via inhibition of the proapoptotic mitochondrial pathway (160). Coadministration of equimolar doses of betaine may alleviate the hepatotoxic risk associated with niacin therapy (161) because niacin catabolism leads to a high demand for methyl groups, leading to a reduction in hepatic concentrations of SAM. A methyl group deficiency may also contribute to niacin-induced insulin resistance by altering the membrane properties of skeletal muscle.

Ethanol feeding can affect several hepatic enzymes in animals, including decreasing methionine synthetase activity (162). This leads to increased BHMT activity to maintain hepatic SAM at normal concentrations (163–165). Betaine supplementation of alcohol-fed animals prevents and partially reverses alcoholic fatty liver (165–167); decreases homocysteine and *S*-adenosylhomocysteine (SAH) concentrations, endoplasmic reticulum stress, and liver injury (168–170); prevents the ethanol-induced accumulation of CH_3 -THF (methyl-folate trap) (171); protects erythrocyte membranes (172); and prevents vitamin A depletion and peroxidative damage (173). Ethanol-induced elevations in hepatic concentrations of SAH inhibit the activity of phosphatidylethanolamine methyltransferase, which is necessary to achieve adequate concentrations of phosphatidylcholine (139). This inhibition may reduce VLDL synthesis and transport of triacylglycerols in the liver (174) and thus at least partly explain the hepatic steatosis due to ethanol feeding. Elevations in the ratio of SAM to SAH by betaine may improve the altered signaling and genomic hypomethylation caused by ethanol and a high-fat diet (175). SAM is currently used successfully to treat liver disease and other maladies, and betaine may be an alternative therapy (77).

Elevated serum homocysteine is frequent in patients with chronic liver disease, and high dosages of folate, cobalamin, and vitamin B-6 frequently fail to normalize homocysteine concentrations (176). Also, plasma concentrations of betaine metabolites (dimethyl glycine and sarcosine) increase due to increased activity of BHMT (177). However, BHMT enzyme activity is reduced in cirrhotic rat livers, which may explain the elevated plasma homocysteine concentrations in cirrhosis (178). A recent

editorial (87) suggests that “Future studies should focus on the dietary content of choline and the possibility that supplementation with choline or betaine will restore the intracellular concentrations of betaine necessary for homocysteine methylation.”

HEART HEALTH

A series of papers (179–182) showed that a combination of betaine and guanidinoacetate (glycocyamine) improves the symptoms of subjects with chronic illness, including heart disease, without toxicity. Betaine can provide a methyl group to guanidinoacetate, via methionine, for the formation of creatine (179). Overall, treatment led to an improved sense of well-being, less fatigue, greater strength and endurance, and increased desire for (and performance of) physical and mental work. Subjects with cardiac decompensation (arteriosclerosis or rheumatic disease) (180) and congestive heart failure (181) had improved cardiac function. Subjects gained weight (improved nitrogen balance) and reported lessened symptoms of arthritis and asthma and increased libido, and those with hypertension experienced transient reduced blood pressure. Glucose tolerance increased in both diabetic subjects and subjects without diabetes. It was therefore concluded in 1951 that “. . . further evaluation of these compounds in patients with heart disease is both desirable and worthwhile” (182).

In the same year, Morrison (183) concluded that atherosclerosis is a preventable metabolic error and that ingestion of lipotropic agents is part of a preventative strategy. He showed that betaine treatment of atherosclerosis improves the ratio of serum phospholipids to serum cholesterol, the sense of well-being, exercise and activity, appetite, angina pain, dyspnea (shortness of breath), and libido (184). Morrison found “. . . betaine to be the most valuable and most effective lipotropic agent tested for treatment in the human subject” (184). Finally, a betaine-lipotrope combination (betaine, choline, liver extract, vitamin B-12, and a low-fat, low-cholesterol diet) led to an impressive decrease in the mortality of subjects with atherosclerosis (185). There was a 25% mortality rate after 12 mo in the control group but no deaths in the lipotrope group.

Betaine studies in animals with experimental atherosclerosis showed a reduction in elevated total and ester-bound cholesterol, β -lipoproteins, total lipids in serum, and total cholesterol and triacylglycerols in the liver (186). Betaine prevents a decrease in the content of nicotinamide coenzymes and adenine nucleotides in the liver and myocardium (187) and increases the transformation of cholesterol to biliary acids for subsequent excretion with the bile (188). This was suggested as one of the mechanisms for the antiatherosclerotic action of betaine. A study of athletes (57) exercising in the heat found that acute betaine ingestion rapidly lowered plasma LDL-cholesterol concentrations (measured after 145 min). However, betaine raised serum total and LDL-cholesterol concentrations slightly in a study of obese subjects consuming a hypoenergetic diet (189).

HOMOCYSTEINEMIA

Epidemiologic studies showed that persons with an elevated serum homocysteine concentration, termed hyperhomocyst(e)inemia or homocyst(e)inemia, have increased risks of cardiovascular disease, stroke, Alzheimer disease, dementia, neural tube defects, and other metabolic disorders (190–197). Elevated

fasting serum homocysteine and an elevated response of homocysteine after methionine loading (postmethionine) are independent risk factors for cardiovascular disease (198). Betaine can improve both risk factors, and in humans with cardiovascular disease, there is a significant inverse relation between plasma betaine and homocysteine concentrations in fasting (199) and postmethionine (200) states. A betaine-rich diet might lower cardiovascular disease risk in healthy humans (201).

Homocysteinemia is caused by an imbalance in the methionine cycle because of either genetic or nongenetic (eg, nutritional) defects that result in elevated serum homocysteine. Chronic elevation of homocysteine results in parallel increases in intracellular SAH, and this, in combination with the SAM:SAH ratio, is predictive of cellular methylation status (202). The severity of clinical symptoms is correlated with the degree of biochemical abnormality. Prevention or treatment, such as activation of defective (mutant) enzyme activity with the relevant cofactor or its precursor (eg, folic acid or vitamins B-12 or B-6), is based on the cause and severity of the defect. If homocysteinemia does not respond to this treatment, pharmacologic doses of betaine or folic acid have been used to enhance the alternative pathway of homocysteine turnover (203, 204).

Homocystinuria

Severe homocysteinemia, which is caused by hereditary homozygous genetic dysfunction (inborn errors of metabolism), is often termed *homocystinuria*. The incidence varies geographically and is estimated as 1/200 000 worldwide (205). Symptoms associated with homocystinuria include mental retardation, seizures, psychiatric disturbances, mental retardation, displacement of the lens of the eye (ectopia lentis), skeletal abnormalities (osteoporosis and scoliosis), and arteriosclerosis and thromboembolism that may lead to life-threatening complications (205). Homocystinuria can be caused by a deficiency of CBS, MTHFR, or methylcobalamin synthesis (Figure 1). Several investigators have determined that betaine can lower homocysteine concentrations and improve some clinical conditions with no adverse events (206). Some studies lasted for 13–16 y, and dosage was typically 6 g/d.

CBS condenses homocysteine and serine to form cystathionine. Vitamin B-6 is a cofactor for CBS, and $\approx 50\%$ of subjects with homocystinuria are responsive to vitamin B-6 therapy. Homocystinuria that is not responsive to vitamin B-6 is much more difficult to treat (207), but betaine, either alone or in conjunction with other therapies, improves clinical symptoms, reduces homocysteine, and increases plasma methionine, serine, and cysteine concentrations (208–221). Homocysteine also accumulates in the central nervous system in CBS deficiency and may play a role in neurologic complications. Betaine treatment in children significantly lowers homocysteine in cerebrospinal fluid and raises serine and SAM concentrations (222). Studies on MTHFR deficiency (223–230) showed that betaine improves homocysteine remethylation, lowers plasma homocysteine, normalizes very low plasma methionine, elevates SAM, and leads to clinical improvement. A defect in cobalamin-C (vitamin B-12) activation results in methylmalonic acidemia and homocystinuria, and treatment with cobalamin, folate, or vitamin B-6 does not completely correct the biochemical defect. Addition of betaine leads to normalized homocysteine and clinical improvement (231–234).

Betaine prevents seizures in animals induced by homocysteine (235), electroconvulsive shock and pentylenetetrazol (236, 237), and strychnine (238, 239) and delays the onset of neurologic impairment due to vitamin B-12 deficiency (240). Rett Syndrome is caused by mutations in the *MECP2* gene (241), and a current clinical study is evaluating whether betaine and folic acid can increase the degree of methylation of some CpG sites on subject DNA and thereby improve symptoms (242). An open-label pilot study of patients with Alzheimer disease did not find a benefit from betaine treatment (243), although efficacy could not be determined because there was no control group.

Mild homocysteinemia

Mild homocysteinemia is much more common than is homocystinuria and is characterized by mildly elevated fasting or postmethionine homocysteine concentrations. Pooled data from many studies showed that mild homocysteinemia occurs in 9–42% of subjects under 50 y of age who have peripheral or cerebral occlusive arterial disease, myocardial infarction, or thromboembolism (244). Mild postmethionine homocysteinemia is present in 21% of young patients with coronary artery disease, 24% of patients with cerebrovascular disease, and 32% of patients with peripheral vascular disease (245).

Mild homocysteinemia can be reduced immediately and in the long term by ingesting a combination of vitamin B-6, folic acid, and betaine (244, 245) or betaine (1.5–6 g/d) alone (57, 189, 198, 201, 246). Betaine, but not folic acid, is effective at preventing an increase in postmethionine homocysteine concentrations (198, 201, 244, 247, 248). Betaine-dependent remethylation occurs mainly in the liver and kidney, whereas folate-dependent remethylation occurs in most cells. Serum homocysteine concentrations may be represented by the extrahepatic supply to the liver via folate remethylation and by the hepatic output via betaine remethylation (198). Therefore, combined ingestion of folic acid and betaine may be the most effective method of lowering homocysteine.

Genetic variants of the enzymes of homocysteine metabolism influence the risk of homocysteinemia and therefore of cardiovascular disease and other common disorders (249). Mild MTHFR deficiency, due to a polymorphism caused by a substitution (677C→T) in the gene that changes an alanine residue to valine, is common (≈ 10 –15% homozygosity in North America) (250, 251). The resulting MTHFR is thermolabile, has decreased enzyme activity, and leads to moderate homocysteinemia if folate status is low. MTHFR-compromised mice with homocysteinemia are more sensitive to changes in betaine intake than are wild-type animals. In mice with mild or severe homocysteinemia due to heterozygosity or homozygosity for a disruption in the *MTHFR* gene, betaine supplementation decreases homocysteine, restores liver betaine and phosphocholine pools, and prevents severe steatosis (199). In mice with moderate homocysteinemia caused by a disruption in the *CBS* gene, betaine supplementation decreases homocysteine, increases liver betaine, and increases BHMT activity in both heterozygous and wild types (252). There are several mutations of the *BHMT* gene (253), and the *QQ* genotype, a substitution of glutamine for arginine (Q239R), is a common variant of *BHMT* in humans that occurs in ≈ 10 –17% of the adult population (253, 254). The *QQ* genotype is associated with decreased risks of neural tube defects (254) and coronary artery disease (249).



KIDNEY HEALTH

BHMT is abundant in primate and pig kidneys, occurs in very low concentrations in rat kidneys, and is essentially absent from the other major organs of monogastric animals (91). BHMT activity in human, pig, and rat kidneys is expressed only in the proximal tubules of the cortex and is absent from the medulla. Immunohistochemical staining patterns show cytosolic expression in both the kidney and the liver. The concentration of betaine in the liver and the kidney is controlled by tonicity *in vivo* via regulation of BHMT (255). Under hypertonic conditions, renal and hepatic BHMT decreases to conserve betaine to be utilized as an osmoprotectant. Conversely, under hypotonic conditions, BHMT increases to reduce betaine and help maintain optimal cell turgor. Betaine accumulation in kidney medullary cells exposed to hypertonic stress is dependent on cyclooxygenase 2 activity (110). Betaine protects against the nephrotoxic effects of carbon tetrachloride in rats (256), and in patients with renal disease taking folic acid, betaine can reduce postmethionine homocysteine concentrations but not fasting homocysteine (248, 257–259).

DNA METHYLATION

Incomplete DNA methylation may lead to genetic instability, senescence, and cancer (260–262), and optimal “methylation diets,” including betaine, have been suggested for the prevention and treatment of a variety of conditions (263). Synthesis of choline for packaging and secretion of fat from the liver consumes methyl groups and appears to compete with other methylation reactions. Thus, a high-fat diet exacerbates methyl deficiency, and a higher proportion of cofactors and methyl group sources relative to calories may explain the extended life span observed in caloric restriction experiments (260).


Epigenetics is the study of heritable traits that do not involve a change in gene sequence (ie, different phenotypes of identical genotypes). Yellow agouti mice have an epigenetic variation in coat color—more yellow is associated with altered metabolism, greater obesity, diabetes, cancer, and earlier mortality. In epigenetics, methylation is the addition of methyl groups to cytosine in DNA to form 5-methylcytosine by utilizing the enzyme DNA methyltransferase. In maternal yellow agouti mice, methyl supplementation, including supplementation with betaine, before and during pregnancy increases DNA methylation (5-methylcytosine) and changes the phenotype of their genetically identical offspring in the healthy, longer-lived direction, ie, less yellow coat (264–267). These studies help address why a cohort of animals with very little genetic or environmental variation will still yield a considerable spread in longevity and disease. Epigenetic variations that affect adult health and life span may be established during embryonic and fetal development involving incomplete methylation of genes and endogenous retroviruses.

PERFORMANCE

As discussed earlier, betaine provides a methyl group to guanidinoacetate via methionine to produce creatine *de novo* (69, 179). A human poliomyelitis study (268) found that ingestion of a betaine-guanidinoacetate combination leads to an improved sense of well-being, less fatigue, and greater general strength and endurance during recovery. Whenever motor unit activity or residual power remained in a muscle, betaine-guanidinoacetate led to a substantial increase. A patent was granted (269) for this

combination, or betaine alone, for the treatment of diseased and weak muscle tissues. A study of subjects with motor neuron disease (270) found that betaine-guanidinoacetate did not impart any objective benefit. However, some patients derived a slight subjective benefit, so the authors concluded that it might be useful for temporary psychological support. Betaine may improve athletic performance, because its addition to a carbohydrate-electrolyte fluid-replacement beverage results in improved mean sprint time to exhaustion and evidence of enhanced anaerobic and aerobic metabolism (57, 271). Rehydration with betaine also results in differential plasma volume changes and reduced LDL and homocysteine.

CONCLUSIONS

Betaine is an important human nutrient obtained from the diet from a variety of foods. It is rapidly absorbed and utilized as an osmolyte and source of methyl groups and thereby helps to maintain liver, heart, and kidney health. Betaine can reduce the elevated serum homocysteine concentrations associated with mild or severe hyperhomocystinuria via the methionine cycle and may play a role in epigenetics and athletic performance. 

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