Trifolamin™

Highly bioavailable forms of B12 and folate

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Science first.

By Cristiana Paul, MS & Suzanne Copp, MS

Trifolamin™ lozenges provide a synergistic combination of the three bioavailable forms of vitamin B12, as methylcobalamin, hydroxycobalamin, and adenosylcobalamin, with the natural active form of folate as [6s]-5-methyltetrahydrofolate. These three forms of B12 are naturally present in foods, and can be readily converted to the active intracellular forms of B12, such as methylcobalamin, adenosylcobalamin, glutathionylcobalamin, and possibly other physiologically active B12 forms.

Trifolamin™ lozenges are formulated to support healthy levels of vitamin B12. Folate is included because vitamin B12 and folate work synergistically in supporting methylation and cell replication, and in helping to promote balanced concentrations of homocysteine.

Supplement Facts

Serving Size 1 lozenge

Amount Per Serving		% Daily Value
Folate (as Quatrefolic® [6S]-5-methyltetrahydrofolate, glucosamine salt 800 mcg)	680 mcg DFE	170%
Vitamin B12 (as Methylcobalamin, Hydroxycobalamin and Adenosylcobalamin)	3,000 mcg	125,000%

Other Ingredients: Mannitol, modified cellulose, natural orange flavor and color, vegetable stearate, citric acid, and luo han guo.

MADE WITH GMO-FREE INGREDIENTS

The Lozenge Advantage

Trifolamin™ uses a lozenge delivery system, allowing for two routes of absorption for B12 and folate: 1) directly through the oral mucosa, and 2) bound by haptocorrin (binding protein secreted in saliva) and carried to the GI tract.

Conditions Associated with B12 Deficiency

There are many conditions where a B12 deficiency acts as an etiological or aggravating factor. Many of these have been shown to improve with adequate B12 supplementation (very high doses of 0.5-1 mg). It is often necessary to optimize the status of many other synergistic nutrients in order to support the reversal of these conditions. Folate is one of the most important cofactors that helps realize the benefits of optimizing B12 status. Other B vitamins, essential minerals, and fatty acids may also have crucial effects in allowing B12 benefits to be maximized, thus the need to supplement with multivitamin, multimineral and essential fatty acid formulas should always be evaluated.

Supplementation with B12 and Folate May Be Beneficial For:

- **Nervous system related impairments:** brain and nerve function, conditions where the need for myelin repair is increased, such as during chemotherapy (for cancer, arthritis, lupus) or any condition involving neurological autoimmunity. Many of these conditions may involve poor cognition, dementia, dizziness, postural hypotension, tinnitus, neuropathy, or hyporeflexia.
- **Impaired healing of oral or GI mucosa:** canker sores, various inflammatory conditions of the gut, or during treatment with NSAIDs or chemotherapy. Gastric mucosa damage causes B12 malabsorption, thus creating a vicious cycle of worsening B12 status and the health of all mucosal membranes.[23]
- **Low white blood cells:** which impairs immunity.
- **Low platelet count:** which impairs healing.
- **Anemia, and/or macrocytic red blood cells:** (high mean corpuscular volume), impaired circulation and oxygen supply.
- **Elevated homocysteine:** associated with increased risk of birth defects, cardiovascular disease and Alzheimer's. It is also an indicator of systemically impaired capacity for methylation, which is involved in neurotransmitter synthesis and clearance, hormonal methylation and genetic expression through DNA methylation. Some of these conditions may involve depression, anxiety, mania, poor appetite, loss of taste, and a higher risk of cancer. Conditions that involve impairment of energy production and low oxygen supply to cells, such as generalized fatigue, anemia, heart failure, respiratory disease, and shortness of breath.
- **Poor sleep quality:** B12 may be involved in normalization of circadian rhythm.[105]
- **Impotence, incontinence**
- **Infertility or high risk of miscarriage or birth defects (5X higher for spina bifida) and poor cognitive and motor development of the fetus or infant,** due to mother's B12 deficiency during pregnancy and breastfeeding.

Synergistic Mechanisms of Action of B12 and Folate

B12 and folate are in high demand by cells with high turnover rate, such as epithelial cells in skin, oral and GI mucosa. B12 and folate are instrumental to new DNA synthesis needed for optimal cell replication and differentiation. Pregnancy is an especially demanding condition where healthy cell division and differentiation is needed for adequate fetal development.

B12 supports energy production from branched chain amino acids, odd-chain fatty acids and cholesterol by providing the mitochondrial enzyme cofactor adenosylcobalamin. Buildup of odd-chain fatty acids may cause demyelination.

B12 and folate support the immune response to various infectious agents (oral or systemic). Adequate B12 and folate statuses are required for the proper proliferation of immune cells.

B12 has been shown to alleviate certain aspects of the autoimmune response, which has been implicated in canker sore outbreaks. B12 supplementation has been shown to alleviate the severity of canker sores[25,26,57,59,66,73,115] and various autoimmune diseases such as atopic dermatitis[92] and rheumatoid arthritis.[87] B12 was shown to have a balancing effect on the immune response by down-regulating inflammatory processes mediated by inducible nitric oxide (iNOS).

B12 was shown to reduce toxicity of heavy metals such as mercury. Heavy metal toxicity may contribute to various pathologies of the nervous system or autoimmune response by binding to body proteins, which dysregulates immune tolerance.[95]

Common Causes of Functional B12 Deficiency

a. Inadequate dietary intake of B12. Vitamin B12 is readily available only in animal products, primarily dairy, meat, and eggs,[64] making vegetarians and vegans at highest risk for deficiency. Many vegan sources of B12, such as fermented spirulina or brewer's yeast, may include a high proportion of B12 analogues with no B12 activity in human physiology.[186,187] Also, as much as 33% of vitamin B12 may be lost due to heating, which is commonly used with animal-derived foods.[153]

b. Malabsorption of B12 from food. This becomes more prevalent with aging, especially after the age of 50.[150]

B12 malabsorption may be caused by any of the following[117,132]:

b.1. Inadequate stomach acidity. Hydrochloric acid (HCl) releases B12 from food proteins, which is a prerequisite for its binding to haptocorrin and then to intrinsic factor (IF), which carries it during GI absorption.[63] However, the metabolism of B12 found in nutritional supplements does not require optimal HCl levels.

Insufficient stomach acidity may be due to:

- Suboptimal HCl production, which may be the result of conditions such as atrophic gastritis or H. Pylori infection. Atrophy of the gastric mucosa is more prevalent in the elderly[62] and those infected with H. Pylori (H. Pylori has a 30%-40% incidence in the US and 70% in developing countries.[165])
- Pharmaceutical drugs that neutralize HCl or reduce HCl production
- Nutritional supplements containing calcium carbonate

b.2. Inadequate pancreatic protease production, which cleaves B12 from haptocorrin and transfers it to IF

b.3. Impaired production of IF. This may be due to congenital IF deficiency, atrophic gastritis or autoimmune conditions that cause antibodies to gastric parietal cells or IF (also referred to as pernicious anemia). Other potential causes include gastric surgeries (gastric bypass), gastric ulcers or cancer.

b.4. GI disorders: ulcerative colitis, celiac disease and other gastro-pathologies due to GI inflammation/infections; treatment with antibiotics or chemotherapy.[66]

b.5. Pharmaceutical drug side-effects. Metformin interferes with B12 absorption.[65]

b.6. Poor B12 binding to transcobalamin (a B12 transport protein in the blood), due to genetic polymorphisms

c. Poor intracellular metabolism of B12. Various genetic polymorphisms may result in poor release of cobalamin from the lysosome or poor conversion of cobalamin to the active forms of B12. Glutahione-S transferase is involved in this step.

Absorption Routes and Rates of Supplemental B12 Absorption

It is commonly accepted that vitamin B12 is absorbed and transported across cellular plasma membranes in the terminal ileum by two alternate mechanisms:

(i) Endocytosis of B12 bound to gastric IF. This route has limited efficiency due to IF saturation at around 2 mcg per meal, and it is severely impaired in various conditions, as described above.

(ii) Passive diffusion directly into the intestinal cell by mass action of very high doses of B12.

One study evaluated absorption rates of B12 (as cyanocobalamin) when given at escalating doses.[116] The results revealed absorption rates of approx. 50% for doses up to 0.5 mcg, 20% for doses around 1 mcg, and only 1-1.2% for doses around 500 mcg. By extrapolation, approximately 10-12 mcg may be absorbed from a total of 1000 mcg B12 ingested at one time. Since the natural forms of B12 are more bioavailable, they may have higher absorption rates than cyanocobalamin, but as of yet this has not been studied.

Special Features of the B12 Forms in Trifolamin™

(A more detailed explanation is found in "Position Paper on Vitamin B12 Forms", available upon request from Designs for Health)

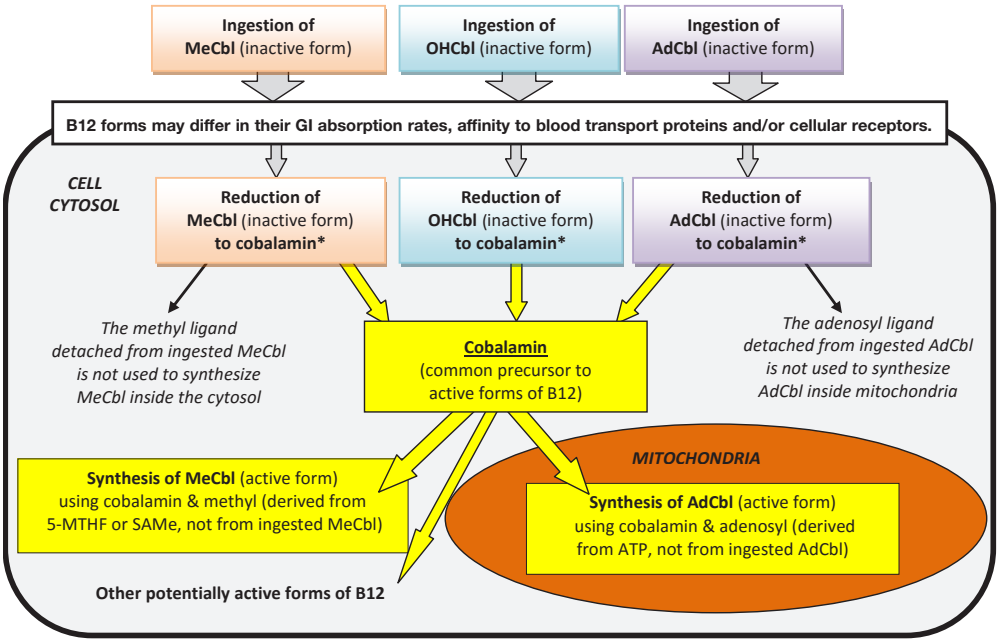
The three forms of B12 found in Trifolamin™ are naturally occurring, and are mostly found in animal foods as well as in some fermented vegetarian foods. These B12 forms are highly bioavailable, most likely because they are bioidentical to the forms occurring in human physiology.[116,173] Specifically, these B12 forms are more bioavailable than the cyanocobalamin form found in many supplements and used for food fortification.[143,186] Cyanocobalamin is not a naturally occurring molecule and some individuals have genetic difficulties breaking it down to enable activation in the body. One study that compared its bioavailability with methylcobalamin showed that it had 3X higher excretion in the urine and much less retention in the liver.[143] In addition, the hydrolysis of this compound produces small amounts of cyanide. Historically, cyanocobalamin was the most common form of supplemental B12 and presented stability advantages in solution for B12 injections and in fortified foods. Today, however, there is no reason to use this form since bioidentical forms of B12 are available and stable.

B12's metabolic conversions are illustrated in Fig.1. Each of the three B12 forms found in Trifolamin™ converts first to a cobalamin intermediate and then to the two main intracellular active forms of B12: methylcobalamin (MeCbl) and adenosylcobalamin (AdCbl). It is important to note that the methyl and adenosyl ligands are cleaved upon cell entry and are not utilized as such during further metabolic activations. It may be that the physiological rationale for this is to even the playing field by making the synthesis of B12's active forms independent of the form of B12 ingested.

Adenosylcobalamin was shown to be broken down to cobalamin at a rate 67X slower than MeCbl;[124] thus, it may provide a more sustained released source of B12 than the other forms. However, no studies have revealed any clinical significance for this metabolic aspect. AdCbl has an approximately 17% higher molecular weight than MeCbl or hydroxycobalamin (OHCbl). Since supplementation with high doses of B12 relies mostly on the passive diffusion route of absorption, it is possible that less AdCbl is absorbed than the other two B12 forms, although no studies have evaluated this to date.

The amounts of MeCbl and AdCbl synthesized from each of the three B12 forms is dictated by cellular conditions and sometimes influenced by genetic polymorphisms. Genetics can affect the activity of enzymes involved in B12 metabolism, absorption and/or binding to blood transport proteins. However, there is no genetic data available at this time that points to specific advantages of any particular form of B12. **Thus, it is impossible to predict whether any particular individual will respond better to any one of the three natural, commercially available forms of B12. Based on this rationale, Trifolamin™ includes all three forms in equal amounts for maximal clinical efficacy in spite of various genetic polymorphisms involving B12 metabolism and related pathways.**

Fig. 1 Genetic SNPs may affect various steps in B12 absorption, blood transport and/or conversions to intra-cellular active forms of B12. (adapted from references 133, 142, 168)



*B12 is converted to cobalamin at different rates among B12 forms and using enzymes specialized for their particular ligand

Clearing up Misconceptions about B12

1. There is a misconception that promotes the concept that the methyl group from supplemental MeCbl participates in intracellular methylation reactions, thus affecting the cellular levels of SAMe (universal methyl donor). In contrast, studies have shown that only 5-MTHF or SAMe can provide methyl groups that bind to cobalamin and form MeCbl inside the cytosol.[173,183] In these reactions, MeCbl donates its methyl group to homocysteine, while being reduced at the same time back to cobalamin. Then the cycle repeats itself where cobalamin proceeds to pick up another methyl group from 5-MTHF or SAMe, and so on. Studies show that these reactions are not influenced by the methyl groups included with ingested MeCbl. The only other methyl group providers for the homocysteine methylation pathways are betaine and choline. As a result of this misconception, another invalid concept has been promoted stating that supplementing with OHCbl versus MeCbl may result in lower intracellular levels of SAMe. This has been disproven by research reviewed in the "Position Paper on Vitamin B12 Forms."

2. Similarly, there is a misconception that the adenosyl group from supplemental AdCbl is used in the synthesis of intra-mitochondrial AdCbl or the synthesis of SAMe, thus making supplemental AdCbl more suited for those with genetic impairment of AdCbl or SAMe synthesis. This was disproven by research that looked at individuals with methylmalonic aciduria who have a severe genetic impairment of synthesizing AdCbl and could not be helped by AdCbl supplementation or any other form of B12. Research shows that the adenosyl ligand is synthesized from ATP inside mitochondria, while the adenosyl ligands brought in by supplemental AdCbl are not utilized as such.[131,133, 168] The only approach that may help individuals with such polymorphisms would be to supplement with higher doses of B12 (ideally all three forms) to hopefully "push" some of these pathways by mass action in hopes of producing more of the metabolites needed.

Advantages of Vitamin B12 from Supplements versus Food[162]

- **High doses (>1000 mcg) of B12** can be incorporated in supplements at levels approximately 100X higher than even those found in healthy/Paleo diets at 10-20 mcg B12/day. A US survey found an average intake of B12 of 5.3 mcg/day from food alone and 24.4 mcg/day from food + B12 fortified food + B12 supplements.
- **Circumvent the deficiency of IF** and enable B12 absorption through the GI border by diffusion at rates of 1-1.2%. Absorption by IF may be saturated at around 1-2 mcg per meal.
- **Supplemental B12 forms are not protein-bound,** thus they bypass the need for HCl and proteolytic enzymes, which are typically required to separate B12 from food proteins.
- **B12 deficiency may take a long time to be corrected without high dose supplementation.** Body stores of B12 (mostly in the liver) have been estimated at 2500 mcg. Thus, a patient who is very depleted in B12 may need approximately 70 servings of Trifolamin™ to support a significant repletion (assuming that 10-12 mcg are absorbed from 1 mg B12). The amount of B12 derived from most multivitamins and fortified foods is often inadequate, as they contain much less than 1000 mcg of B12 per dose and are often in the cyanocobalamin form, which may only be 70-80% bioavailable for some individuals, but not bioavailable at all for those with genetics-related difficulty cleaving the cyano ligand.

RDA of B12 May Not Support Optimal Health

The generally accepted guidelines set by the Institute of Medicine (IOM) through the **RDA tables recommend an intake of 2.4 mcg of vitamin B12 per day** for adults under 50 years old. The recommended B12 RDAs for pregnant and nursing women are 2.5 mcg and 2.6 mcg, respectively.[150] For individuals over 50 years of age, the RDA for B12 is150-200 mcg. This dose, which is much higher than the regular RDA for the younger population, is justified by the fact that 10-30% of older individuals may absorb B12 poorly, whether from natural foods, fortified foods or supplements.[150] However, malabsorption may obviously happen for younger individuals as well due to various conditions described in this paper. One study showed that 4-7 mcg of B12 are needed to optimize the B12 status markers homocysteine and methylmalonic acid.[184]

Researcher M. Fenech has published a number of studies proposing new criteria for optimal intake of certain vitamins, including folate and B12.[77, 144, 145,158] He found that a level of **B12 intake above 7 mcg/day is optimal,** as long as serum levels of B12 are also maintained above 406 ng/ml. His research shows that meeting these goals correlates with the lowest level of chromosomal damage found in lymphocytes and probably other cells.[145] In addition, adequate folate status is needed to support healthy DNA replication because the metabolic pathways of B12 and folate are interrelated in supporting this physiological function.

Prominent Paleo diet researcher Loren Cordain provided a novel frame of reference for optimization of macro- and micronutrient intakes. Cordain has published an analysis of a sample Paleolithic-type diet implemented with readily available modern foods.[146] He estimated that the average B12 content of a 2200 Kcal Paleolithic-type diet was 17.6 mcg/day, while that of folates was 891mcg/day. Consequently, we propose here that an optimal intake of vitamin B12 may be at least as high as the average evolutionary intake of vitamin B12 to which humans have adapted for millions of years of 17.6 mcg/day, which also meets and exceeds recommendations based on Fenech's criteria of 7mcg/day. In addition, for individuals who may absorb as little as 1% of the ingested B12 dose, the recommendation needs to be approximately 100X higher, which is approximately 1760 mcg B12.

Superiority of the Active Form of Folate (5-MTHF)

Quatrefolic® contains a novel form of folate, the glucosamine salt of 5-MTHF, which dissociates prior to GI absorption and provides pure 5-MTHF for systemic use. In contrast, naturally occurring folates contain 5-MTHF bound to polyglutamate chains, which need to be removed prior to GI absorption, a process that is not very efficient in certain individuals. Thus, Quatrefolic® provides a form of folate that is bioidentical to the naturally occurring folate compound 5-MTHF but with a more reliable absorption.[152, 157,165,167]

It is also important to note that the 5-MTHF form of folate is superior to folic acid, which is a synthetic, non-naturally occurring molecule utilized as a precursor to metabolically active folates in human physiology. The human body is obviously best adapted to metabolize natural folates (such as 5-MTHF and folinic acid), while folic acid is metabolized on pathways that are not properly regulated, likely because this is not a naturally occurring molecule.[70,159,190, 193]

Thus, it is not surprising that a significant percentage of the population has genetic polymorphisms that cause impaired folic acid utilization. Most importantly, numerous studies are revealing a link between folic acid (derived from fortified foods and/or supplements) and risk of various cancers, reduced NK cell activity, and other detrimental effects.[159,192]

How to Take:

- Take one lozenge per day, or as directed by a health care practitioner. Allow lozenge to dissolve slowly and completely in mouth and swallow.
- Best taken after a meal. The B12 portion carried to the GI tract needs to be released from the haptocorrin transport protein by proteases, which are typically secreted in the GI tract in response to food.
- For instances where folate supplementation is not desired, please consider our Tricobalamin™ lozenges, which contain the same formulation as Trifolamin™ without the additional folate.

For a list of references cited in this document, please visit:

http://catalog.designsforhealth.com/assets/itemresources/TrifolaminB12References.pdf