Vitamin B-12

Vitamin B-12 (cobalamin) is an essential nutrient required for growth and neurological development. There are 2 currently known enzymes in humans that utilize vitamin B-12 as a cofactor—namely, cytoplasmic methionine synthase and mitochondrial methylmalonyl-CoA mutase. The cytoplasmic reaction with vitamin B-12 in methylcobalamin form converts homocysteine to methionine for the synthesis of the principal methyl donor, *S*-adenosylmethionine, and regenerates tetrahydrofolates from 5-methyltetrahydrofolate for thymidylate synthesis. In the mitochondria, adenosylcobalamin is a cofactor for conversion of methylmalonyl-CoA to succinyl-CoA and so permits oxidation of odd-chain fatty acids and ketogenic amino acids.

Deficiencies

In recent years, there have been many scientific advances in understanding of the mechanism of absorption and intracellular processing of vitamin B-12. Despite this, the diagnosis of vitamin deficiency, resulting in anemia and neurological symptoms, and insufficiency (or subclinical deficiency), where symptoms are not well expressed but methylmalonic acid and/or homocysteine are often elevated, remains challenging. The 4 biochemical markers—total serum vitamin B-12, serum holotranscobalamin (HoloTC), plasma homocysteine, and methylmalonic acid—can be used for the assessment of status, but knowledge of how age, sex, ethnicity, pregnancy, polymorphisms, renal function, microbiome, and drug use affect these markers, independently of vitamin B-12 status, is essential for this assessment. It is now recognized that a combination of at least 2 of these markers is preferable when diagnosing patients. Serum vitamin B-12 and HoloTC are often used as first-line markers, followed by methylmalonic acid or homocysteine, if applicable (1). Serum vitamin B-12, with its typical cutoff for deficiency of $<$148 pmol/L (2), is still the most commonly used test but it is also the least sensitive. In vitamin B-12 insufficiency, serum vitamin B-12 concentration extends up to ~350 pmol/L, which is common, occurring in ~30% of the population (3). High concentrations of serum vitamin B-12 do not always exclude deficiency and should prompt further investigations and treatment if the patient is clinically responsive (4, 5). HoloTC, the biologically active form of vitamin B-12, is slightly more sensitive and offers a diagnostic advantage over serum vitamin B-12 in pregnancy and infancy, and is a useful test in the suspected diagnosis of transcobalamin and haptocorrin (vitamin B-12–binding proteins) deficiencies (1). Plasma homocysteine and methylmalonic acid are sensitive tests, but their use is less reliable, particularly in renal impairment and hypothyroidism. Additionally, deficiencies of riboflavin, vitamin B-6, and folate elevate homocysteine concentrations, whereas gut microbiota and alterations in fat metabolism may have an influence on methylmalonic acid (1). Overall, as there is no gold-standard test to define deficiency, we recommend that a positive clinical response to vitamin B-12 treatment should be an important guide to the diagnosis of deficiency.

Apart from macrocytosis, which includes ineffective erythropoiesis, moderate hemolysis, and inefficient leukopoiesis and thrombopoiesis, vitamin B-12 deficiency may be associated with 1 or more of a large number of signs, in particular relating to the nervous system, such as cognitive impairment, depression, stroke, brain atrophy, macular degeneration, impaired development in children, and neural tube defects (3).

Diet Recommendations

The RDA values for the United States were based on the assumption that 50% of vitamin B-12 is absorbed from the diet and the intakes will maintain normal hematological values and serum vitamin B-12 within reference ranges. Therefore, 0.4 μg/d was recommended for infants aged 0–6 mo old and 0.5 μg/d for those aged 7–12 mo old. Intakes of 0.9, 1.2, 1.8, and 2.4 μg/d are recommended for individuals aged 1–3 y, 4–8 y, 9–13 y, and $>$18 y old, respectively (6). Higher requirements were set for pregnancy (2.6 μg/d) and lactation (2.8 μg/d). Because of the high prevalence of food-bound malabsorption with increasing age, people $>$50 y old are recommended to consume food products fortified with crystalline vitamin B-12 as its absorption is not affected in this condition. However, it remains debatable if the above intakes are sufficient to achieve optimum vitamin B-12 status, considering that they do not take into account vitamin B-12 insufficiency as well as the many factors that affect serum vitamin B-12 concentration independently of vitamin B-12 intake. Following a summation of daily losses that need to be compensated for by dietary intake of vitamin B-12, Doets et al. (7) estimated that vitamin B-12 intakes between 3.8 μg/d and 20.7 μg/d would be sufficient to prevent deficiency.

Food Sources

Vitamin B-12 is only made in nature by bacteria. Herbivorous animals acquire vitamin B-12 from their gastrointestinal tract, where vitamin B-12 is synthesized by bacteria and then absorbed and incorporated into their tissues. However, omnivores and carnivores, including humans, do not have the ability to absorb bacterial vitamin B-12 from their large intestine and are dependent on animal-source foods, which reach the small intestine. Plants do not contain vitamin B-12 but some algae grow symbiotically with bacteria that produce vitamin B-12. As such, it is extremely important that vegans and vegetarians ensure that they take supplemental vitamin B-12, either in fortified foods or as tablets. Foods that are rich

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in vitamin B-12 include dairy products, meat (especially liver), eggs, fish, and shellfish. However, the bioavailability of vitamin B-12 from foods differs and vitamin B-12 in meat in particular is not as available as that from dairy foods and fish/shellfish (8).

The natural absorption of vitamin B-12 from food is a complex process and is saturated at ~10 μg/d (8), but intake from crystalline vitamin B-12 mostly occurs by diffusion, with ~1% of that consumed being absorbed.

Clinical Uses

Many malabsorption and intestinal disorders lead to vitamin B-12 deficiency and require treatment. Pernicious anemia, an autoimmune gastritis in which parietal cell antibodies lead to the destruction of intrinsic factor needed for absorption and produced by parietal cells in the stomach, requires vitamin B-12 treatment for life. The prevalence of pernicious anemia is 0.1% in the general population, increasing to 1.9% in people over the age of 60 y (1). Hypochlorhydria, which is associated with atrophic gastritis and common in the elderly, impedes the release of vitamin B-12 from food. In Crohn's and celiac disease, vitamin B-12 deficiency arises as a result of reduced absorption due to villous atrophy and mucosal impairment, respectively. In chronic pancreatitis, the enzymes required for the release of cobalamin from haptocorrin are impaired. All of these conditions often require vitamin B-12 therapy. Tropical sprue, intestinal lymphoma, amyloidosis, short bowel syndrome, bariatric surgery, the use of proton pump inhibitors, metformin, oral contraceptives, and nitrous oxide abuse have been associated with vitamin B-12 depletion and the monitoring of status is advisable. Cyanide poisoning is treated with large doses of hydroxocobalamin.

Toxicity

Vitamin B-12 administered in supra-physiological doses, either orally or by intramuscular injections, has so far not been found to be toxic. However, more research is required to demonstrate that no unwanted adverse effects arise following large doses of vitamin B-12 for a prolonged period of time. The formation of antibodies and immune complexes with vitamin B-12–binding proteins, leading to spuriously high serum vitamin B-12 or HoloTC concentrations but also to impaired vitamin B-12 absorption, have been reported (1, 4). One study has shown that the ability to deliver vitamin B-12 to cells in vitro was impaired and clearance of vitamin B-12 in vivo was abnormal in the presence of these antibody complexes (9).

Recent Research

Since no "gold standard" to define vitamin B-12 deficiency using biochemical markers currently exists, research has concentrated on improving the analytical utility and interpretation of concentrations of these markers in serum or plasma. It is now recognized that pregnancy- and infancy-specific reference ranges should be used for all markers of vitamin B-12 status (1). Ethnicity should be addressed when interpreting serum vitamin B-12 and possibly HoloTC values; however, more data are required to derive relevant deficiency and insufficiency cutoffs. Caution is required when methylmalonic acid in plasma is "normal" compounded by clinical symptoms related to vitamin B-12 deficiency. The use of a combined indicator of vitamin B-12 status (cB12), a mathematical model combining all 4 vitamin B-12 markers, has been evaluated in numerous studies and some diagnostic settings, showing an additional diagnostic benefit (1). The appreciation of the clinical significance of high serum vitamin B-12 in patients who are not on treatment led to the development of a screening strategy if unexplainable high concentrations of vitamin B-12 are incidentally encountered (10). A value of 1000 pmol/L has been suggested as a cutoff for high serum vitamin B-12 concentrations if a patient is not taking supplements (10). Lowering homocysteine (baseline value >11 μmol/L) with B vitamins, including vitamin B-12, has proven effective in slowing cognitive decline and brain atrophy (3). Although formal recommendations are required, the clinician should be especially alert to middle-aged and elderly patients with vitamin B-12 insufficiency in order to slow cognitive decline and perhaps avert or delay dementia by implementing appropriate treatment (1, 3). More evidence has emerged from work investigating high folate and low vitamin B-12 interactions, and although the mechanisms leading to potential adverse effects (cognitive impairment, gestational diabetes, increased cancer risk, insulin resistance, interference with epilepsy drugs) have not been elucidated, the findings from studies suggest that this interaction is genuine and needs to be urgently addressed. Vitamin B-12 status depletion by reduction in HoloTC concentration due to high folic acid intake was recently proposed as the mechanism for this interaction (11). This is especially relevant in countries where mandatory folic acid fortification without vitamin B-12 fortification has been implemented, and countries planning to introduce such a policy. The use of natural folate (i.e., 5-methyltetrahydrofolate), as opposed to synthetic folic acid, has so far not been associated with adverse effects on vitamin B-12 status. Despite many research advances in further understanding the complex absorption and metabolic processing of cobalamin, the pace of implementing vitamin B-12 knowledge into clinical practice has been slow; thus, many physicians still solely rely on a serum vitamin B-12 test when diagnosing and treating patients. Other pressing topics related to vitamin B-12 research that are currently being investigated include the understanding of the role of the microbiome and vitamin B-12 analogs on vitamin B-12 status, and their impact on markers of vitamin B-12 and elucidation of the mechanism of vitamin B-12 crossing the blood–brain barrier. A better understanding of the impact of long-term vitamin B-12 injections on vitamin B-12 status/markers is also warranted for future studies.

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