Proton pump inhibitor use and its effect on vitamin B12 and homocysteine levels among men and women: A large cross-sectional study

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ABSTRACT

Background: Previous studies have demonstrated an association between proton pump inhibitors (PPI) use and vitamin B12 deficiency. However, data regarding PPI use and elevated serum homocysteine level, an important marker of vitamin B12 deficiency, are scant.

Methods: Data were collected from medical records of subjects examined at a screening center in Israel. Cross sectional analysis was conducted on 25,953 subjects. Levels of vitamin B12 and homocysteine were compared between subjects who consumed PPI medications and those who did not.

Results: The mean age of the study population was 45 years and 33% were females. Subjects who received PPI medications had a minor higher vitamin B12 levels (320 pmol/L vs 300 pmol/L, p=0.024). Levels of vitamin B12 remained higher in females receiving PPI medications after performing a stratified analysis according to subjects' gender. Homocysteine levels were higher in subjects receiving PPI medications as compared to those who did not (12.0 μmol/L vs 11.6 0 μmol/L, p<0.001). Levels remained higher in female subjects after performing a stratified analysis according to subjects' sex. There was no statistically significant difference in the prevalence of vitamin B12 deficiency (according to two cutoffs: vitamin B12≤200 or ≤140 pmol/L) as well as the prevalence of hyperhomocysteinemia (defined as homocysteine >15.0 μmol/L) between the two groups.

Conclusions: According to our study, no association was found between PPI medication use and vitamin B12 deficiency or hyperhomocysteinemia. Patients receiving PPI medications had slightly higher levels of vitamin B12 and homocysteine, however these differences were too small to have any clinical relevance.

Key Indexing Terms: Vitamin B12; Homocysteine; Hyperhomocysteinemia; PPI, Proton pump inhibitor. **[Am J Med Sci 2022;364(6):746–751.]**

INTRODUCTION

Vitamin B12, also known as Cobalamin, is a water-soluble vitamin that plays a crucial role as a cofactor for enzymes involved in synthesis of fatty acids, myelin and DNA.[1] Vitamin B12 is derived exclusively from diet, mainly from meat, fish and dairy products, where it is found in a protein-bound form. After ingestion it undergoes a complex process through the gastrointestinal tract. In the stomach, the gastric acid and the enzyme pepsin convert it to the unbound form which later binds to salivary R proteins. In the duodenum, vitamin B12 is released from R proteins and binds to intrinsic factor (IF) and is eventually absorbed in the terminal ileum.[2]

It was previously suggested that the acid lowering effect of protein pump inhibitors (PPIs) may cause vitamin B12 deficiency by interfering with the above-mentioned processes in two manners. First, use of PPI medications elevates the pH of the stomach. This change in pH interferes with the cleavage of proteins from the protein-bound form of vitamin B12 and conversion to the unbound form of vitamin B12. Secondly, the high pH levels in the stomach also promote overgrowing of bacteria that affect the absorption process.[3–5]

Few studies have examined the association between acid suppressive therapy and vitamin B12 deficiency. However, the results are conflicting and therefore inconclusive.[5–9] The case definitions in these studies had a great

variability including coded diagnosis in population based database, decreased measured levels of vitamin B12, prescription of vitamin B12 supplement or elevated methylmalonic acid (MMA) or homocysteine levels.[5–9] Because the metabolic effect of vitamin B12 is intracellular, serum vitamin B12 levels have limited specificity and sensitivity for detecting tissue deficiency.[10] Homocysteine and MMA are precursors in two intracellular vitamin B12-dependent pathways and therefore their accumulation serves as a useful biochemical markers for vitamin B12 deficiency.[11] As opposed to MMA measurement that requires not readily available laboratory methods, homocysteine levels can be measured easily and previous studies reported that its sensitivity for identifying vitamin B12 deficiency is greater than 95%.[12–15] Despite the usefulness of this biomarker, the data regarding the effect of acid lowering agents on vitamin B12 deficiency using this method is very limited.[9] It is important to emphasize the limited evidence regarding the effect of acid suppressive agents on vitamin B12 metabolism, as reflected by poor case definition, in previously published studies.

The purpose of the present study was to examine the risk of vitamin B12 deficiency according to serum levels of both vitamin B12 and homocysteine among patients treated by PPI medications based on data from aa large-scale screening center database.

METHODS

The study was conducted in Rabin Medical Center (RMC), a large university-affiliated tertiary medical center in the central district in Israel. RMC provided annual medical evaluation for individuals above the age of 18, excluding pregnant females. In each visit, a thorough medical history was carried out including chronic medications usage. A complete physical exam was performed and a broad range of blood and urine tests as well as ergometric stress testing, spirometry and chest radiogram were done. We designed a retrospective cross-sectional study in which the medical records of individuals with documented vitamin B12 and homocysteine levels were retrieved. In cases of repeated visits, the first one was included. For the purpose of the study, we included individuals aged 18-65 years. Individuals taking vitamin B12, folate supplements or multivitamins were excluded. The extracted variables of interest were sex, age, body mass index (BMI) and the following laboratory parameters: serum vitamin B12, homocysteine, complete blood count, creatinine, fasting blood glucose, albumin, liver function tests, thyroid stimulating hormone (TSH) and lipids profile. In addition, data regarding chronic medications were collected (active medications according to the subject's report). Between May 2000 and May 2011, serum vitamin B12 levels were measured using Immulite® assay (Siemens). Between May 2011 and October 2014 (the end of the study), it was measured using Architect® assay (Abbott). For both machines, measuring methods were similar and reference level was 138−781 pmol/L. Serum homocysteine levels were measured using TDx® assay (Abbott) between May 2000 and January 2005 and using AxSYM® assay (Abbott) since then. Two cutoff values were used to define vitamin B12 deficiency: a liberal cutoff with serum concentrations lower than 200 pmol/L and a restrictive cutoff value with serum concentrations lower than 140 pmol/L.[16–18] Hyperhomocysteinemia was defined as serum homocysteine levels above 15.0 μmol/L.[19] The renal function was evaluated using the Chronic Kidney Disease Epidemiology Collaboration equation.

In this study we performed a cross-sectional analysis. Cases were defined as vitamin B12 deficient according to the three different definitions; levels of vitamin B12 ≤140 pmol/L, vitamin B12 ≤200 pmol/L, and hyperhomocystynemia defined as homocysteine >15.0 μmol/L. The association between PPI use and vitamin B12 status was examined, and analysis was performed for each one of the above-mentioned case definitions.

Statistical analyses were performed using SAS v. 9.4. Continuous variables were expressed as mean ± S.D, categorical variables were expressed as percentage. Continuous variables were analyzed using Student's t test or unpaired Wilcoxon test for variables with non-normal distribution. Chi-square test was used for categorical variables. A multivariate analysis was conducted implementing logistic regression model adjusted for universal confounders and possible interfering factors. Sensitivity analyses were thereafter carried out. As sex may interfere with both vitamin B12 and serum homocysteine levels, the analysis was stratified according to patient's sex.[20,21] P value of <0.05 was considered statistically significant. Data from each visit was entered into a spreadsheet created in Excel. Identifying details of all the participants were coded ahead of analysis. Since all data were anonymized, the Helsinki Ethics Committee of Rabin Medical Center (Petah Tikva, Israel) gave approval for the study, with no need for informed consent from the participants.

RESULTS

The cross-sectional analysis included 25,953 patients: 15,871 subjects with data regarding homocysteine levels and 13,356 with data regarding vitamin B12 levels. Among patients with homocysteine levels measurements, 520 were taking PPIs. Among patients with vitamin B12 levels measurements, 447 were taking PPIs. A flow chart of the study population is shown in Figure 1. Patients' baseline characteristics are presented in Table 1. The mean age of the study population was 45 ± 10 years and 33% were females. Men had a higher BMI, lower estimated glomerular filtration rate (eGFR), higher albumin, higher cholesterol, low-density lipoprotein (LDL) and triglycerides levels. Plasma homocysteine was higher was in males, and serum vitamin B12 and folate were higher in females (p<0.001).

The association between PPI use and vitamin B12 levels as well as homocysteine levels is presented in Table 2.

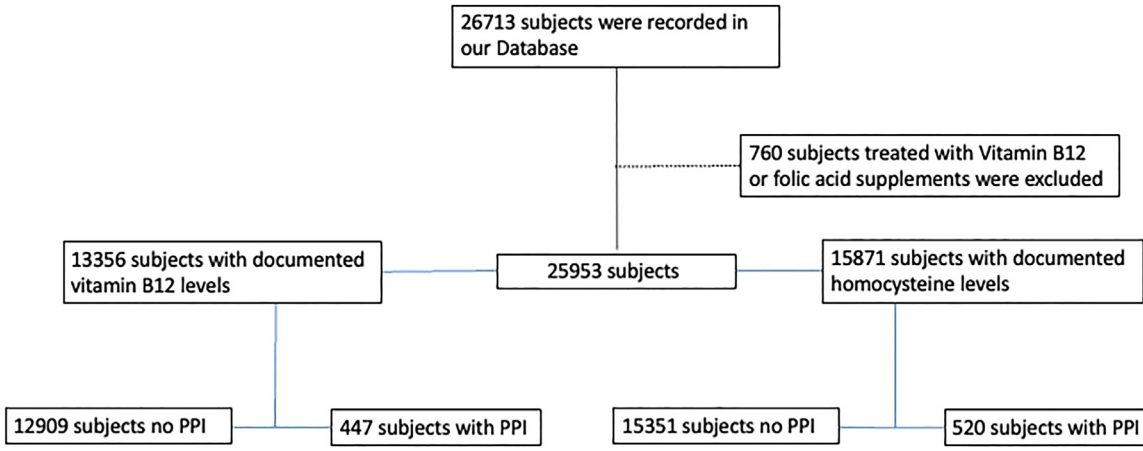

FIGURE 1. Study participants flow chart.

TABLE 1. Baseline characteristics of the study population (N = 25,953).

	Males (N = 17495)	Females (N = 8458)	P value
Age (years), mean (SD)	45 (10)	45 (11)	0.001
BMI (kg/m2), mean (SD)	27.08 (3.92)	25.31 (4.84)	<0.001
Smokers (%)	2824 (16.1)	1433 (17.0)	0.104
eGFR (CKD-EPI)	98.43 (14.01)	102.16 (14.39)	<0.001
Serum Albumin (mg/dl), mean (SD)	4.57 (0.27)	4.37 (0.28)	<0.001
Serum glucose concentration (mg/dl), mean (SD)	98 (19)	92 (14)	<0.001
Total cholesterol (mg/dl), mean (SD)	196 (37)	198 (38)	0.001
Triglycerides (mg/dl), mean (SD)	137 (92)	105 (65)	<0.001
LDL cholesterol (mg/dl)	122 (31)	116 (32)	<0.001
HDL cholesterol (mg/dl), mean (SD)	47 (10)	60 (14)	<0.001
Plasma homocysteine concentrations (mmol/L)	12.7 (6.5)	9.5 (3)	<0.001
Serum vitamin B12 concentrations (pmol/L) mean (SD)	291 (126)	319 (144)	<0.001
Serum folate concentrations (nmol/L) mean (SD)	19.6 (9.9)	22.9 (10.5)	<0.001

BMI=body mass index, HDL=high-density lipoprotein LDL=low-density lipoprotein, SD=standard deviation.

Subjects that were taking PPIs had higher vitamin B12 levels (p = 0.024). Levels remain higher in females taking PPIs after performing a stratified analysis according to subjects' sex. Additionally, homocysteine levels were higher in subjects taking PPIs as compared to those who do not (p < 0.001). Levels remain higher in female subjects after performing a stratified analysis according to subjects' sex. These was no statistically significant difference in the prevalence of vitamin B12 deficiency (according to the two cutoffs) as well as the prevalence of hyperhomocysteinemia between the two groups. There were no statistically significant differences between the two groups after conducting sex-stratified analysis. Multivariate analysis for PPI use as determining factor for B12 deficiency and hyperhomocysteinemia is presented in Table 3.

DISCUSSION

In the present study, we found that vitamin B12 deficiency is a common disorder among healthy individuals in Israel, reaching a prevalence as high as 21%. Our study did not find an association between PPI use and vitamin B12 deficiency, defined by two cutoffs as mentioned above. Nonetheless, in our study we found an association between PPI use and hyperhomocysteinemia which is an important marker for vitamin B12 deficiency.[10] Our results demonstrated slightly higher levels of vitamin B12 among individuals taking PPIs (320 pmol/L vs 300 pmol/L, p = 0.024), however this difference is too small to be considered clinically relevant. In addition, a slightly higher homocysteine levels (12.0 μmol/L vs 11.6 0 μmol/L, p < 0.001) were demonstrated in the PPI group, this finding may indicate a negative effect of PPIs on vitamin B12 metabolism to some extent, however this difference is very small and cannot be considered clinically relevant. One may consider these findings conflicting, however the higher levels of both vitamin B12 and homocysteine among the PPI group compared to the control group are negligible and probably do not reflect a real biochemical cascade of homocysteine accumulation. Interestingly the two-above mention differences remain statistically significant in females after performing

TABLE 2. Serum levels of B12 and homocysteine among patients with and without PPI treatment stratified by gender.

		PPI (N = 723)	No PPI (N = 25230)	P value
All population	Homocysteine (μmol/L) mean (SD)	12.0 ± 7.6	11.6 ± 5.7	<0.001
	Homocysteine > 15.0 (μmol/L) (%)	68 (13.1)	1748 (11.4)	0.234
	B12 (pmol/L) mean (SD)	320± 157	300 ± 132	0.024
	B12 ≤ 200 (pmol/L) (%)	92 (20.6)	2705 (21.0)	0.906
	B12 ≤ 140 (pmol/L) (%)	12 (2.7)	498 (3.9)	0.257
Males	Homocysteine (μmol/L) mean (SD)	12.6± 8.5	12.7 ± 6.4	0.519
	Homocysteine > 15.0 (μmol/L) (%)	63 (16.2)	1568 (15.3)	0.616
	B12 (pmol/L) mean (SD)	309 ± 145	290 ± 125	0.054
	B12 ≤ 200 (pmol/L) (%)	72 (21.2)	1953 (22.6)	0.596
	B12 ≤ 140 (pmol/L) (%)	8 (2.4)	382 (4.4)	0.076
Females	Homocysteine (μmol/L) mean (SD)	10.1 ± 3.4	9.5 ± 3.0	0.006
	Homocysteine > 15.0 (μmol/L) (%)	5 (3.8)	180 (3.5)	0.809
	B12 (pmol/L) mean (SD)	356 ± 188	318 ± 143	0.040
	B12 ≤ 200 (pmol/L) (%)	20 (18.5)	752 (17.7)	0.799
	B12 ≤ 140 (pmol/L) (%)	4 (3.7)	116 (2.7)	0.541

Data regarding homocysteine levels were available for 15871 subjects. Of those 520 were taking PPIs. Data regarding B12 levels were available for 13356 subjects. Of those 447 were taking PPIs.

a sex-sensitive analysis. Additionally, we conducted multivariable analysis models controlling for universal clinically relevant variables including age, smoking status, BMI, Diabetes mellitus, eGFR and sex; in these analyses there was also no association between PPI use and vitamin B12 deficiency as reflected by reduced vitamin B12 levels or elevated homocysteine when using acceptable clinically relevant cutoffs.

Previous studies that examined the association between PPIs and vitamin B12 deficiency have reached conflicting results. Marcuard et al. were among the first to demonstrate that there is a dose-dependent reduction of the absorption of vitamin B12 in healthy volunteers on short-term (two weeks) therapy with omeprazole.[22] Termanini et al. were the first to report an increased risk for vitamin B12 deficiency due to long-term use of PPIs among patients with Zollinger-Ellison syndrome.[23] Another large-scale case-control study based on the data from the Kaiser Permanente Northern California patient population was conducted by Lam et al.[8] This study included 210,155 patients and the case definition in this study was according to the problem list in medical records, diagnostic codes in the electronic medical records, low vitamin B12 value on blood test or administration of injectable vitamin B12. This study didn't use biochemical markers of vitamin B12 deficiency such as homocysteine or MMA. The authors found that the use of PPIs for 2 years or longer was associated with an increased risk of vitamin B12 deficiency.[8] A systematic review and meta-analysis published by Jung et al in 2015 found 4 case-control studies and one observational study regarding acid suppressive agents (not limited to PPIs). The pooled results showed that long-term acid lowering agent use was significantly associated with development of vitamin B12 deficiency (hazard ratio 1.83, 95% CI: 1.36–2.46, P-value 0.00).[5] It is important to mention that only one small study in this meta-analysis published by Valuck and Ruscin defined vitamin B12

TABLE 3. PPI use as determining factor for B12 deficiency and hyperhomocysteinemia stratified by gender.

		B12 ≤ 200 (pmol/L)	B12 ≤ 140 (pmol/L)	Homocysteine > 15.0 (μmol/L)
All population	Unadjusted	0.97 (0.77 - 1.23)	0.71 (0.40 - 1.27)	1.17 (0.90 - 1.52)
	Model 1[a]	0.99 (0.78 - 1.25)	0.69 (0.39 - 1.24)	1.03 (0.79 - 1.34)
	Model 2[b]	0.97 (0.76 - 1.23)	0.64 (0.35 - 1.18)	1.01 (0.77 - 1.32)
Males	Unadjusted	0.91 (0.70 - 1.20)	0.54 (0.27 - 1.10)	1.07 (0.81 - 1.41)
	Model 1[a]	0.93 (0.71 - 1.21)	0.54 (0.27 - 1.10)	1.05 (0.80 - 1.39)
	Model 2[b]	0.92 (0.70 - 1.21)	0.55 (0.27 - 1.12)	1.03 (0.78 - 1.36)
Females	Unadjusted	1.08 (0.66 - 1.77)	1.40 (0.51 - 3.86)	1.08 (0.44 - 2.68)
	Model 1[a]	1.25 (0.76 - 2.06)	1.54 (0.55 - 4.33)	0.81 (0.32 - 2.03)
	Model 2[b]	1.20 (0.71 - 1.98)	1.16 (0.36 - 3.77)	0.78 (0.31 - 1.98)

[a] Adjusted for age and gender (gender excluded in stratified analysis).
[b] Adjusted for age, smoking status, BMI, Diabetes mellitus, eGFR and gender (gender excluded in stratified analysis).

deficiency according to vitamin B12 levels as well as homocysteine and MMA levels.[9] In this study the authors concluded that there was association between chronic use of acid suppressive agents by older adults and the development of vitamin B12 deficiency.

Similar to our study results, Elzen et al. also reported that PPI use was not associated with reduced vitamin B12 levels in 125 patients aged 65 and above that had been treated with PPIs for more than 3 years.[24] In another study published by Bytqi et al, one of the few studies that used homocysteine measurements, long term PPI use was associated with reduced serum vitamin B12 levels as well as higher rates of hyperhomocystenemia, but no significant change was demonstrated in the mean homocysteine levels.[7] These results are not in line with our results, however the study population in this particular study was small and therefore the data regarding increased homocysteine levels due to PPI use are still scant.

As mentioned earlier, vitamin B12 levels have low sensitivity and specificity detecting vitamin B12 deficiency.[10] Vitamin B12 is involved in two intracellular biochemical pathways as an important cofactor for two enzymes; the first is mitochondrial methylmalonyl-coenzyme A (CoA) mutase which is involved in propionate metabolism by catalyzing isomerization of methylmalonyl-CoA to succinyl-CoA. The second enzyme is cytosolic methionine-synthase which is involved in the cytosolic transmethylation of homocysteine to methionine by 5-methyl-tetrahydrofolate.[25] Lack of vitamin B12 results in accumulation of the substrates in these two vitamin B12-dependent pathways, leading to increased measured levels of homocysteine and MMA in the serum.[11] High levels of homocysteine, beside serving as a marker of vitamin B12 deficiency, have been associated with several pathologic states such as cardiovascular diseases, chronic kidney disease, neurodegenerative conditions and peripheral neuropathy and therefore it is an important mediator to the damage caused by vitamin B12 deficiency.[26] This emphasizes the importance of measuring homocysteine levels when assessing vitamin B12 deficiency, an important strength of our study.

Interestingly, homocysteine levels were higher among patients taking PPIs and levels remained higher in females but not males after performing a stratified analysis according to sex. It has been previously suggested that women have greater flux of homocysteine through trans-sulfuration pathway.[21] In light of this, PPIs may affect differently the metabolism of homocysteine in the two sexes, however further study is needed to explore this concept.

Our study has certain limitations. First, PPI exposure was based on patients' self-reported medications list, which may include errors. In addition, a reporting bias might exist, that is, individuals could potentially avoid disclosing their real intake or alternatively report taking prescribed medication even though they do not take the drug. An important aspect of this limitation is that data regarding the duration of PPI use as well as the adherence of the participants were not available to us. Another limitation is that data on nutritional habits of the participants were unavailable, which may be an important confounder when studying vitamin deficiency; for example, a strict vegan diet may decrease vitamin B12 levels while consuming "fortified" foods may increase the levels. Another limitation is related to the fact that most of the examined population were white collar workers. This homogeneity may diminish the study's external validity. In addition, MMA levels were not available for us and therefore causes of isolated elevation of homocysteine without MMA elevation (e.g., folate and vitamin B6 deficiency) cannot be ruled out. Another limitation of our study is the lack of examination of other medications that may affect vitamin B12 levels, such as metformin among diabetes patients.

To the best of our knowledge, this study is one of the largest studies to date examining the association between PPI use and vitamin B12 deficiency as demonstrated by both measured vitamin B12 levels as well as homocysteine levels. According to our results there was no increased risk of vitamin B12 deficiency in subjects using PPIs, however due to conflicting study results and the wild use of this medication, further research is still needed.

AUTHORS CONTRIBUTIONS

T.L, E.C, E.G, I.G, I.K - conceptualization, methodology and writing, T.S- statistical analysis.

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DECLARATION OF COMPETING INTEREST

Nothing to declare.

REFERENCES

1. **Ankar A, Kumar A.** *Vitamin B12 Deficiency*. Treasure Island (FL): StatPearls; 2021. [Internet].
2. **Howden CW.** Vitamin B12 levels during prolonged treatment with proton pump inhibitors. *J Clin Gastroenterol*. 2000;30(1):29–33. https://doi.org/10.1097/00004836-200001000-00006.
3. **Kapadia C.** Cobalamin (Vitamin B12) deficiency: is it a problem for our aging population and is the problem compounded by drugs that inhibit gastric acid secretion? *J Clin Gastroenterol*. 2000;30(1):4–6. https://doi.org/10.1097/00004836-200001000-00003.
4. **Abraham NS.** Proton pump inhibitors: potential adverse effects. *Curr Opin Gastroenterol*. 2012;28(6):615–620. https://doi.org/10.1097/MOG.0b013e328358d5b9.
5. **Jung SB, Nagaraja V, Kapur A, et al.** Association between vitamin B12 deficiency and long-term use of acid-lowering agents: a systematic review and meta-analysis. *Intern Med J*. 2015;45(4):409–416. https://doi.org/10.1111/imj.12697.
6. **Cotter PE, O'Keeffe ST.** Use of proton pump inhibitors is not associated with Vitamin B12 deficiency and in older hospital patients: a case control study. *Eur Geriatr Med*. 2011;2(4):253–255. https://doi.org/10.1016/j.eurger.2011.02.007.
7. **Qorraj-Bytyqi H, Hoxha R, Sadiku S, et al.** Proton pump inhibitors intake and iron and vitamin B12 status: a prospective comparative study

with a follow up of 12 months. *Open Access Macedonian J Med Sci*. 2018;6(3):442–446. https://doi.org/10.3889/oamjms.2018.142.
8. **Lam JR, Schneider JL, Zhao W, et al.** Proton pump inhibitor and histamine 2 receptor antagonist use and vitamin B12 deficiency. *JAMA - J Am Med Assoc*. 2013;310(22):2435–2442. https://doi.org/10.1001/jama.2013.280490.
9. **Valuck RJ, Ruscin JM.** A case-control study on adverse effects: H2 blocker or proton pump inhibitor use and risk of vitamin B12 deficiency in older adults. *J Clin Epidemiol*. 2004;57(4):422–428. https://doi.org/10.1016/j.jclinepi.2003.08.015.
10. **Scarlett JD, Read H, O'Dea K.** Protein-bound cobalamin absorption declines in the elderly. *Am J Hematol*. 1992;39(2):79–83. https://doi.org/10.1002/ajh.2830390202.
11. **Green R, Allen LH, Bjørke-Monsen A-L, et al.** Vitamin B12 deficiency. *Nat Rev Dis Prim*. 2017;3(1):17040. https://doi.org/10.1038/nrdp.2017.40.
12. **Gilfix BM.** Vitamin B12 and homocysteine. *Can Med Assoc J*. 2005;173(11):1360. https://doi.org/10.1503/cmaj.1050170.
13. **Hølleland G, Schneede J, Ueland PM, et al.** Cobalamin deficiency in general practice. Assessment of the diagnostic utility and cost-benefit analysis of methylmalonic acid determination in relation to current diagnostic strategies. *Clin Chem*. 1999;45(2):189–198.
14. **Refsum H, Smith AD, Ueland PM, et al.** Facts and recommendations about total homocysteine determinations: an expert opinion. *Clin Chem*. 2004;50(1):3–32. https://doi.org/10.1373/clinchem.2003.021634.
15. **Savage DG, Lindenbaum J, Stabler SP, et al.** Sensitivity of serum methylmalonic acid and total homocysteine determinations for diagnosing cobalamin and folate deficiencies. *Am J Med*. 1994;96(3):239–246. https://doi.org/10.1016/0002-9343(94)90149-x.
16. **Stabler SP.** Clinical practice. Vitamin B12 deficiency. *N Engl J Med*. 2013;368(2):149–160. https://doi.org/10.1056/NEJMcp1113996.
17. **Chui CH, Lau FY, Wong R, et al.** Vitamin B12 deficiency—need for a new guideline. *Nutrition*. 2001;17(11–12):917–920. https://doi.org/10.1016/s0899-9007(01)00666-9.
18. **Morris MS, Jacques PF, Rosenberg IH, et al.** Folate and vitamin B-12 status in relation to anemia, macrocytosis, and cognitive impairment in older Americans in the age of folic acid fortification. *Am J Clin Nutr*. 2007;85(1):193–200. https://doi.org/10.1093/ajcn/85.1.193.
19. **Smith AD, Refsum H, Bottiglieri T, et al.** Homocysteine and dementia: an international consensus statement. *J Alzheimer's Dis : JAD*. 2018;62(2):561–570. https://doi.org/10.3233/JAD-171042.
20. **Margalit I.** Vitamin B12 deficiency and the role of gender: a cross-sectional study of a large cohort. *Ann Nutr Metab*. 2018;72(4):265––271. doi:10.1159/000488326.
21. **Cohen E, Margalit I, Goldberg E, et al.** Gender differences in homocysteine concentrations, a population-based cross-sectional study. *Nutr, Metab Cardiovasc Dis : NMCD*. 2019;29(1):9–14. https://doi.org/10.1016/j.numecd.2018.09.003.
22. **Marcuard SP.** Omeprazole therapy causes malabsorption of cyanocobalamin (Vitamin B_12). *Ann Intern Med*. 1994;120(3):211. https://doi.org/10.7326/0003-4819-120-3-199402010-00006.
23. **Termanini B, Gibril F, Sutliff VE, et al.** Effect of long-term gastric acid suppressive therapy on serum vitamin B12 levels in patients with Zollinger-Ellison syndrome. *Am J Med*. 1998;104(5):422–430. https://doi.org/10.1016/s0002-9343(98)00087-4.
24. **den Elzen WPJ, Groeneveld Y, de Ruijter W, et al.** Long-term use of proton pump inhibitors and vitamin B12 status in elderly individuals. *Aliment Pharmacol Ther*. 2008;27(6):491–497. https://doi.org/10.1111/j.1365-2036.2008.03601.x.
25. **Vincenti A, Bertuzzo L, Limitone A, et al.** Perspective: practical approach to preventing subclinical B12 deficiency in elderly population. *Nutrients*. 2021;13(6):1913. https://doi.org/10.3390/nu13061913.
26. **De Giuseppe R, Venturelli G, Guez S, et al.** Homocysteine metabolism in children and adolescents with epidermolysis bullosa. *BMC Pediatr*. 2016;16(1):173. https://doi.org/10.1186/s12887-016-0714-7.

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