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Clinical profile Of Vitamin B12 Deficiency In Rural Populations Of Western Up And Its Management

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Abstract

Background: Although cobalamin (vitamin B12) deficiency was described over a century ago, it is still difficult to establish the correct diagnosis and prescribe the right treatment. Symptoms related to vitamin B12 deficiency may be diverse and vary from neurologic to psychiatric. A number of individuals with vitamin B12 deficiency may present with the classic megaloblastic anemia. In clinical practice, many cases of vitamin B12 deficiency are overlooked or sometimes even misdiagnosed. In this study we describe the heterogeneous disease spectrum of patients with vitamin B12 deficiency in whom the diagnosis was based on low serum B_{12} levels. We discuss the possible clinical signs and symptoms of patients with B_{12} deficiency and the various pitfalls of diagnosis and treatment.

Aims and objective: To find out the various manifestations of Vitamin B12 deficiency and observe the therapeutic response to different modality of therapy in the rural population of western up. This observational study was performed in Department of General Medicine, in a tertiary care hospital of UP for duration of 1.5 year (January 2020-June 2021) on 100 patients of vitamine B12 deficiency admitted in the emergency ward, ICU and medical ward of UPUMS with confirmed consent and fit to the inclusion criteria were recruited for this study

Result: The literature search yielded a total of 100 applicable case studies, of which 57 involved females and 43 involved males. Fifteen cases included under 20 years of age forty young adults in the age range of 20-39 years, 26 cases included those in middle adulthood aged 40-59 years, and 19 cases included elderly individuals aged above 60. Among them 85 patients were strict vegetarian and 15 patients were non vegetarian. Eleven patients were on long term medication, among them 4 patients were diabetic and on Metformin and 7 patients were on Proton pump inhibitors. A total of 43 cases involved patients with B12 deficiency related neurological impairment. These patients ranged from 23-84 years of age and presented an array of symptoms. A common clinical manifestation experienced by patient's studied with neurological impairment is paresthesia or tingling, numbness, and/or burning in the skin (16 cases). Another prevalent clinical manifestation observed is ataxia (4 cases) or the lack of coordination of muscle movements, often accompanied by gait abnormalities. Other symptoms commonly reported included weakness (21 cases) and impaired cognitive abilities (5 cases). One case indicated that Lhermitte's sign, an electrical sensation which moves down spine, was a prevalent symptom especially during neck flexion. Three cases reported fatigue as a problematic occurrence. A total of 25 cases involved patients with B12 deficiency related psychiatric abnormalities. These patients ranged in age 31-79 years and exhibited a multitude of symptoms. The most prevalent features of patients experiencing psychiatric abnormalities as a result of B12 deficiency included irritability (5 cases), and decreased interest (5 cases) .Other manifestations included depression (25 cases) and sleep disturbances (4 cases). Thirteen cases reported oral manifestations of B12 deficiency, and included patients aged 19-84 years. These patients experienced varying symptoms. The most prevalent symptoms were glossitis (4 cases) and pain and burning sensations in the mouth. A

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total of 28 cases reported B12 deficiency-related dermatological manifestations which included hyper pigmentation of the knucles.
The majority of cases reported B12 replacement therapy via intramuscular (IM) hydroxycobalamine administration as the initiated treatment. A total of 86 cases utilized this method of B12 repletion. This typically consisted of daily injections for a week or longer, followed by weekly injections, then monthly injections thereafter. Doses for IM B12 therapy ranged from 100 µg to 1000 µg. 16 cases indicated that oral B12 was given at 1000 µg. Treatment with B12 replacement therapy resulted in full recovery in 64 cases. Improvement in symptoms, but not full remission was observed in 21 cases. 5 cases indicated little to no improvement in symptoms and 2 cases did not report a treatment outcome.
Conclusion: Vitamin B 12 deficiency was higher among middle age group adults aged between 20-39 years. Females had a higher B_{12} deficiency than male

Key words: Vitamine B12, Hydroxycobalamine, Megaloblastic, anemia, rural

Introduction: Vitamin B12 is classified as a water-soluble vitamin, and is distinctive among all vitamins due to its large size, complexity, and that it contains the metal ion cobalt.[1] It is necessary for appropriate nervous system function and for the metabolism of carbohydrate, protein, and fat. Deficiencies in B12 can lead to inefficient erythropoiesis and megaloblastic anemia.[2] Furthermore, neurological disorders such as neuropathy, myelopathy, memory impairment, dementia, depression, and brain atrophy may occur in those with low B12 status.[3] According to one study, a neuropathy known as combined degeneration of the spinal cord, is one of the most debilitating manifestations of vitamin B12 deficiency.[5] Neurological and psychiatric symptoms have been seen in patients without related anemia or macrocytosis, with B12 concentrations in the previously defined range of low-normal. This highlights the need for the prevention of B12 deficiency.[3] The main dietary sources of B12 are animal products including meat, fish, eggs, and dairy products.[3] Other sources include B12-containing, fortified plant products such as cereals, plant-based milks, soy products, and B12-fortified yeast extract.[5]

Vitamin B12 belongs to the family of cobalamins and serves as a cofactor for two important reactions in humans. As methylcobalamin, it is a cofactor for methionine synthetase in the conversion of homocysteine to methionine, and as adenosylcobalamin for the conversion of methylmalonylcoenzyme A (CoA) to succinyl-CoA. Vitamin B12 comes from the diet and is present in all foods of animal origin. The daily absorption of vitamin B12 is 5 mcg.[6]
The liver contains 2-5 mg of stored vitamin B12 . Since daily utilization is 3-5 mcg, the body usually has sufficient stores of vitamin B12 so that it takes more than 3 years for vitamin B12 deficiency to occur if all intake or absorption immediately ceases.[7]
Since vitamin B12 is present in foods of animal origin, dietary vitamin B12 deficiency is extremely rare but is seen in vegans—strict vegetarians who avoid all dairy products, meat, and fish. Pernicious anemia is an autoimmune illness whereby auto antibodies destroy gastric parietal cells (that produce intrinsic factor) and cause atrophic gastritis or bind to and neutralize intrinsic factor, or both. Abdominal surgery may lead to vitamin B12 deficiency in several ways. Gastrectomy will eliminate the site of intrinsic factor production; blind loop syndrome will cause competition for vitamin B12 by bacterial overgrowth in the lumen of the intestine; and surgical resection of the ileum will eliminate the site of vitamin B12 absorption. Rare causes of vitamin B12 deficiency include fish tapeworm (Diphyllobothrium latum) infection, in which the parasite uses luminal vitamin B12 ; pancreatic insufficiency (with failure to inactivate competing cobalamin-binding proteins [R-factors]); severe Crohn

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disease, causing sufficient destruction of the ileum to impair vitamin B12 absorption; and perhaps prolonged use of proton pump inhibitors.[8]

The megaloblastic anemias are a group of disorders characterized by the presence of distinctive morphologic appearances of the developing red cells in the bone marrow. The marrow is usually hypercellular and the anemia is based on ineffective erythropoiesis. The cause is usually a deficiency of either cobalamin (vitamin B12) or folate, but megaloblastic anemia may occur because of genetic or acquired abnormalities that affect the metabolism of these vitamins or because of defects in DNA synthesis not related to cobalamin or folate. Cobalamin and folate absorption and metabolism are described next, followed by the biochemical basis, clinical and laboratory features, causes, and treatment of megaloblastic anemia.[4]

Cobalamin (vitamin B12) exists in a number of different chemical forms. All have a cobalt atom at the center of a corrin ring. In nature, the vitamin is mainly in the 2-deoxyadenosyl (ado) form, which is located in mitochondria. It is the cofactor for the enzyme L-methylmalonyl coenzyme A (CoA) mutase. The other major natural cobalamin is methylcobalamin, the form in human plasma and in cell cytoplasm. It is the cofactor for methionine synthase. There are also minor amounts of hydroxocobalamin to which methyl- and adocobalamin are converted rapidly by exposure to light.

Cobalamin is synthesized solely by microorganisms. Ruminants obtain cobalamin from the foregut, but the only source for humans is food of animal origin, for example, meat, fish, and dairy products. Vegetables, fruits, and other foods of nonanimal origin are free from cobalamin unless they are contaminated by bacteria. A normal Western diet contains 5–30 μg of cobalamin daily. Adult daily losses (mainly in the urine and feces) are 1–3 μg (~0.1% of body stores), and because the body does not have the ability to degrade cobalamin, daily requirements are also about 1–3 μg. Body stores are of the order of 2–3 mg, sufficient for 3–4 years if supplies are completely cut of

Many symptomless patients are detected through the finding of a raised mean corpuscular volume (MCV) on a routine blood count. The main clinical features in more severe cases are those of anemia. Anorexia is usually marked, and there may be weight loss, diarrhea, or constipation. Glossitis, angular cheilosis, a mild fever in more severely anemic patients, jaundice (unconjugated), and reversible melanin skin hyperpigmentation also may occur with a deficiency of either folate or cobalamin. Thrombocytopenia sometimes leads to bruising, and this may be aggravated by vitamin C deficiency or alcohol in malnourished patients. The anemia and low leukocyte count may predispose to infections, particularly of the respiratory and urinary tracts. Cobalamin deficiency has also been associated in a few studies with impaired bactericidal function of phagocytes and with osteoporosis.

Vitamin B12 deficiency causes a moderate to severe anemia of slow onset; patients may have few symptoms relative to the degree of anemia. In advanced cases, the anemia may be severe, with hematocrits as low as 10-15%, and may be accompanied by leukopenia and thrombocytopenia. The deficiency also produces changes in mucosal cells, leading to glossitis, as well as other vague gastrointestinal disturbances such as anorexia and diarrhea. Vitamin B12 deficiency also leads to a complex neurologic syndrome. Peripheral nerves are usually affected first, and patients complain initially of paresthesias. As the posterior columns of the spinal cord become impaired, patients complain of difficulty with balance or proprioception, or both. In more advanced cases, cerebral function may be altered as well, and on occasion dementia and other neuropsychiatric abnormalities may be present. It is critical to recognize that the nonhematologic manifestations of vitamin B12 deficiency can be manifest despite a completely normal complete blood count. Patients are usually pale and

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may be mildly icteric or sallow. Typically, later in the disease course, neurologic examination may reveal decreased vibration and position sense or memory disturbance (or both).9
It is usually necessary to treat patients who have developed cobalamin deficiency with lifelong regular cobalamin injections. In the UK, the form used is hydroxocobalamin; in the United States, cyanocobalamin. In a few instances, the underlying cause of cobalamin deficiency can be permanently corrected, for example, fish tapeworm, tropical sprue, or an intestinal stagnant loop that is amenable to surgery. The indications for starting cobalamin therapy are a well-documented megaloblastic anemia or other hematologic abnormalities and neuropathy due to the deficiency. Patients with borderline serum cobalamin levels but no hematologic or other abnormality may be followed to make sure that the cobalamin deficiency does not progress. If malabsorption of cobalamin or rises in serum MMA levels have been demonstrated, however, these patients also should be given regular maintenance cobalamin therapy. Cobalamin should be given routinely to all patients who have had a total gastrectomy or ileal resection. Patients who have undergone gastric reduction for control of obesity or who are receiving long-term treatment with proton pump inhibitors should be screened and, if necessary, given cobalamin replacement.
Replenishment of body stores should be complete with six 1000-µg IM injections of hydroxocobalamin given at 3- to 7-day intervals. More frequent doses are usually used in patients with cobalamin neuropathy, but there is no evidence that they produce a better response. Allergic reactions are rare and may require desensitization or antihistamine or glucocorticoid cover. For maintenance therapy, 1000 µg hydroxocobalamin IM once every 3 months is satisfactory. Because of the poorer retention of cyanocobalamin, protocols generally use higher and more frequent doses, for example, 1000 µg IM, monthly, for maintenance treatment.[10]

Materials and methods: This observational study was performed in Department of General Medicine, in a tertiary care hospital of UP for duration of 1.5 year (January 2020-June 2021) on 100 patients of vitamine B 12 deficiency admitted in the emergency ward, ICU and medical ward of UPUMS with confirmed consent and fit to the inclusion criteria were recruited for this study
Inclusion criteria: Patients of more than 12 years of age having low vitamine B12 level
Exclusion criteria: Subjects not giving consent or subjects with a history of Vitamin B12 intake or pregnant or lactating women or subjects with history of recent blood transfusion
Study design: An Observational study was done over 1½ year time period on patients attending outpatient department of General Medicine and admitted in Medicine ward of UPUMS, Safai, Etawah, fulfilling the inclusion and exclusion criteria
Study duration: 1 ½ year from January 2020 to June 2021.
Study group : A 100 number of patients attending Department of Medicine UPUMS,SAIFAI was taken. An informed consent was taken from all the cases before their inclusion into the study. The study was undertaken as per approval of ethical committee UPUMS, Safai, Etawah
Methodology: All patients with anemia, non compressive myelopathy or neuropathy were screened for vitamine B12 deficiency.Patients with GBP of megaloblastic erythropoesis were also screened for vitamine B12 deficiency.Those who have low vitamine B12 level were included in the study. All the cases of vitamine B12 deficiency admitted in the emergency ward, ICU and medical ward of UPUMS during study period of Jan 2020 to July 2021 were included. A questionnaire developed and were pre-tested in 100 vitamine B12 deficient patients. Patients who meet the inclusion criteria for selection of study were identified. The

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purpose of the study was explained to the patients and those who were willing to give consent were included this study.

After informed consent each patient underwent detailed history, including demographics, leisure activities, past medical history, family medical history, medication history and nutritional habits were compiled and documented and all base line investigations including serum vitamine B12 level were conducted.
All the parameters under investigations were determined in the Venous blood samples collected from all known cases of vitamine B12 deficiency and to analyze Complete Blood Count,Erythrocyte Segmentation Rate,Reticulocyte count, General Blood Picture and Serum Vitamine B12 level by using standardized and automated assays.
Complete Blood Count, estimation is done using 2 ml anti-coagulated venous blood using EDTA vial, which is done by automated hematology(Sysmex1000) analyzer using electric impedence method.Genera Blood Picture is done by manual slide staining using Leishman stain. Reticulocyte count done by manually by supravital methyl crystal blue stain. Erythrocyte sedimentation rate done by westergren method.Vitamine B12 estimation is done by using 2 ml blood serum by Abott Architect i 1000 SR machine by chemiluminescence method.Serum AST (Aspartate Transaminase), ALT (Alanine Transaminase), ALP (Alkaline Phosphatase),Total Bilirubin, S. albumin, total protein, S. Creatinine and Blood Urea was measured at baseline using SELECTRA PRO-XL auto-analyzer machine in central lab of UPUMS.
Ethical clearance The research procedure followed was in accordance with the approved ethical standards of the ethical committee of UPUMS, Saifai, Etawah
Statistical analysis Analysis was done descriptively for the demographic details. Statistical software (GraphPad Prism5; version 5.01) was used for analysis. Statistical tests were considered significant if P-value was <0.05 at confidence interval of 95%. Data were analysed using IBM (Statistical Package for Social Sciences) SPSS version 24. Proportions and percentages used for representing qualitative data. Mean +/- SD is calculated for quantitative data. Chi-square test and fischer's exact test were applied to measure association between categorical data. P value less than 0.05 were considered statistically significant.

Results:

Table-1: Distribution of patients according to age

Age groups (years)	No. of patients	Percentage (%)
<20	15	15.0
20-39	40	40.0
40-59	26	26.0
60-79	16	16.0
≥80	03	03.0

Table-1 shows the distribution of patients according to age. More than one third of the patients were between 20-39 years (40%) followed by 40-59 years (26%) ,60-79 years (16%),less than 20 years (15%) and more than 80 years (03%).

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Table-2: Distribution of patients according to gender

Gender	No. of patients	Percentage (%)
Male	43	43.0
Female	57	57.0

Table-2 shows the distribution of patients according to gender. There were 43 % male and 57% female.

Table-3: distribution of patients according to diet

Diet	Percentage (%)
Vegeterian	85
Non vegeterian	15

Table-3 shows the distribution of patients according to diet. More than half of patients were vegetarian by diet (85%) and the rest (15%) were non-vegetarian by diet.

Table 4 : Distribution of CBC parameter

CBC parameter	Mean ±SD
WBC (10^3 /uL)	7.581±3.685
RBC (10^6/uL)	3.627±0.6907.581±3.685
HB (g/dL)	8.924±2.177
PLT (10^3 /uL)	157.979±88.317
MCV (fL)	109.039±16.924
MCH (pg)	33.891±4.288
MCHC (g/dL)	32.836±2.371
RDW (%)	14.58±1.155

Table-4 shows the distribution of CBC parameters. The mean of WBC (10^3 /uL), RBC (10^6 /uL), Hemoglobin (g/dl),Platelet count (10^3 /uL),MVC (Fl),MCH(pg),MCHC(g/dl) and RDW was 7.581±3.685, 7.581±3.685, 8.924±2.177, 157.979±88.317, 109.039±16.924, 33.891±4.288, 32.836±2.371 and 14.58±1.155 respectively.

Table 5 : Distribution of other biochemical parameter

Other biochemical parameter	Mean ±sd
Vitamin b 12(pg/ml)	108.99±23.701
Sr folate (ng/ml)	8.366±5.640
Sr ferritin (ng/ml)	294.96±120.271

Table-4 shows the distribution of other biochemical parameters. The mean of vitamin B 12 (pg/ml), Sr folate(ng/ml) and Sr ferritin (ng/ml) are 108.99±23.701, 8.366±5.640 and 294.96±120.271 respectively.

Tabnle 6 : Distribution of Other biochemical parameters

Other biochemical parameters	Mean±SD
Sr Billirubin (mg/dl)	1.554±1.001
Sr LDH (U/L)	585.81±325.064

Table-6 shows the distribution of other biochemical parameters. The mean of Sr Billirubin (mg/dl) and Sr LDH (U/L) are 1.554±1.001 and 585.81±325.064 respectively.

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Table 7 : Distribution of vitamine B 12 deficiency and intake of Metformin and PPI

Drug intake	Percentage of vitamine b12 deficiency
Metformin	4.0
PPI	7.0

Table-7 shows the distribution of vitamine B 12 deficiency and intake of Metformin and PPI. The mean of vitamine B 12 deficiency related to intake of Metformin and PPI are 4% and 7 % respectively.

Table 8: Distribution of Hematological Clinical Features

Hematological features	No of patients	percentage
Anemia	93	93.0
Leukopenia	28	28.0
Thrombocytopenia	55	55.0
Pancytopenia	24	24.0

Table-8 shows the distribution of hematological clinical features. In my study, majority of patients had anemia 93 (93%) followed by 55 (55%) reported thrombocytopenia. 28 (28%) cases reported Leukopenia, and 24(24%) patients reported Pancytopenia(fig 1).

Table 9: Distribution of Cutaneous manifestation

Cutaneous manifestation	No of patients	percentage
Hyperpigmentation	28	28.0
Jaundice	36	36.0

Table-9 shows the distribution of cutaneous manifestation. The mean of Hyperpigmentation and Jaundice among cutaneous manifestations are 28% and 36 % respectively.

Table 10: Gastrointestinal manifestation

Gastrointestinal manifestation	No of patients	percentage
glossitis	18	18.0

Table 10 shows that 18% of the patients had glossitis among 100vitamin B 12 deficient patients.

Table 11: Distribution of Neurological manifestation

Neurological manifestation	No of patients	percentage
paresthesia	16	16.0
Impaired vibration sense	9	9.0
Impared position sense	9	9.0
Muscle weakness	21	21.0
ataxia	4	4.0
anosmia	10	10.0
Taste abnormality	12	12.0
Peripheral neuropathy	6	6.0
Autonomic neuropathy	8	8.0

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Table-11 shows the distribution of neurological manifestation. In my study, majority of the patients had muscle weakness 21 (21%) followed by 16 (16%) reported parasthesia. 12 (12%) cases reported taste abnormality,10 (10%)cases reported anosmia,9 (9%) cases reported impaired vibration and position sense each,8(8%) cases reported autonomic neuropathy, 6(6%) cases reported peripheral neuropathy and 4(4%) patients reported ataxia.

Table 12: Distribution of Psychiatric and Cognitive manifestatuion

Psychiatric and Cognitive manifestatuion	No of patients	percentage
Dementia	16	16.0
Depression	25	25.0
Psychosis	8	8.0
Cognition impairment	5	5.0

Table-12 shows the distribution of psychiatric and cognitive manifestatuion. In this study, majority of patients had Depression 25 (25%) followed by 16 (16%) reported Dementia. 8 (8%) cases reported Psychosis and 24(24%) patients reported Cognition impairment.

Discussion: B12 deficiency can occur due to a number of causes and can affect people of various ages and backgrounds. As already mentioned, B12 is naturally present only in meats and foods of animal origin. Therefore, individuals that do not eat any animal products, such as vegans, are at high risk for deficiency if they do not consume adequate amounts of fortified products or supplements. A total of 85 cases reported deficiency due to vegetarianism. Of these, 52 were vegan 33 were vegeterian. Previous studies have shown that compared to omnivores, vegetarians have a lower average serum concentration of B12.[11] Some studies have also suggested that compared to omnivores, vegetarians have lower B12 serum concentrations. One study indicated that vegetarians develop deficiency regardless of the type of diet, geographical location where they reside, or any other factors.[12] It is imperative that those who follow a vegetarian or vegan diet consume adequate amounts of B12 fortified food products and/or take supplements containing B12 to prevent deficiency. Malabsorption of protein-bound vitamin B12 is often observed in the elderly population where gastric acid is not produced in sufficient amounts to proteolyze B12 from proteins. [13]
Competition for vitamin B12 leading to deficiency (4 cases) with use of Metformin for the treatment of diabetes. Although the responsible mechanism for metformin inducing B12 deficiency is controversial it is proposed that metformin affects the calcium-dependent ileal cell membrane receptors needed for B12-intrinsic factor uptake.[14] Metformin-induced vitamin B12 deficiency was found in five cases. [15-17] Vitamine b12 deficiency was also found in chronic proton pump inhibiters use for more than 12 months among 7 patients.
The clinical manifestations of B12 deficiency are varied and include neurological, psychiatric, oral, and dermatological signs and symptoms. A total of 37 cases involved neurological complications as a result of B12 deficiency. The main features of neurological impairment observed in the cases reviewed included weakness of the extremities s (21 cases), paresthesia (16 cases), ataxia (4 cases), and impaired proprioception (9 cases). Psychiatric abnormalities were observed in 25 cases. The main features of psychiatric manifestations observed were depression (25 cases), irritability (9 cases), decreased interest (6 cases), and sleep disturbances (4 cases). Oral manifestations were reported in 12 cases and symptoms were abnormal taste sensation and tingling sensation around mouth was found in 5 cases and glossitis among 18 cases.

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Conclusion: B12 deficiency can be prevented by adequate intake of B12 containing food products such as meat, fish, eggs, and dairy products or B12 containing fortified plant products such as cereals, plant-based milks, soy products, and B12- fortified yeast extract. However, meat and animal products are often recommended to be limited due to either prevention or management of chronic health conditions, while fortified foods usually contain small doses of this vitamin. If dietary adequacy is a probable cause, B12 containing supplements should be considered. Another method to prevent deficiency that has been proposed is the fortification of flour with B12.[18] In order to be effective among groups at a high risk for deficiency the flour would likely have to be fortified at a dose of 100 µg or greater per 100 g.[14] Keller et al. suggested a novel way of preventing B12 deficiency using B12-fortified tooth paste.[19] In their study with vegans and vegetarians with low B12 status all biomarkers measured including TCII, total homocysteine and serum B12 values were normalized after 5 weeks duration of the experiment.

References

1. Brody T. Classification of biological structure. Academic Press: San Diego, CA, USA. 1999:1-56.
2. Headstrom PD, Rulyak SJ, Lee SD. Prevalence of and risk factors for vitamin B12 deficiency in patients with Crohn's disease. Inflammatory bowel diseases. 2008 Feb 1;14(2):217-23.
3. Vogiatzoglou A, Smith AD, Nurk E, Berstad P, Drevon CA, Ueland PM, Vollset SE, Tell GS, Refsum H. Dietary sources of vitamin B-12 and their association with plasma vitamin B-12 concentrations in the general population: the Hordaland Homocysteine Study. The American journal of clinical nutrition. 2009 Apr 1;89(4):1078-87.
4. Selhub J, Morris MS, Jacques PF, Rosenberg IH. Folate–vitamin B-12 interaction in relation to cognitive impairment, anemia, and biochemical indicators of vitamin B-12 deficiency. The American journal of clinical nutrition. 2009 Feb 1;89(2):702S-6S.
5. Gilsing AM, Crowe FL, Lloyd-Wright Z, Sanders TA, Appleby PN, Allen NE, Key TJ. Serum concentrations of vitamin B12 and folate in British male omnivores, vegetarians and vegans: results from a cross-sectional analysis of the EPIC-Oxford cohort study. European journal of clinical nutrition. 2010 Sep;64(9):933-9.
6. Green R, Mitra AD. Megaloblastic anemias: nutritional and other causes. Medical Clinics. 2017 Mar 1;101(2):297-317.
7. Sobczyńska-Malefora A, Harrington DJ. Laboratory assessment of folate (vitamin B9) status. Journal of clinical pathology. 2018 Nov 1;71(11):949-56.
8. Loscalzo J. Personalized cardiovascular medicine and drug development: time for a new paradigm. Circulation. 2012 Jan 31;125(4):638-45.
9. Hesdorffer CS, Longo DL. Drug-induced megaloblastic anemia. New England Journal of Medicine. 2015 Oct 22;373(17):1649-58.
10. Stabler SP. Vitamin B12 deficiency. New England Journal of Medicine. 2013 Jan 10;368(2):149-60.
11. Selhub J, Morris MS, Jacques PF, Rosenberg IH. Folate–vitamin B-12 interaction in relation to cognitive impairment, anemia, and biochemical indicators of vitamin B-12 deficiency. The American journal of clinical nutrition. 2009 Feb 1;89(2):702S-6S.
12. Pawlak R, Parrott SJ, Raj S, Cullum-Dugan D, Lucus D. How prevalent is vitamin B12 deficiency among vegetarians?. Nutrition reviews. 2013 Feb 1;71(2):110-7.
13. Kannan R, Ng MJ. Cutaneous lesions and vitamin B12 deficiency: an often-forgotten link. Canadian Family Physician. 2008 Apr 1;54(4):529-32.

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14. Fujita H, Narita T, Yoshioka N, Hosoba M, Ito S. A case of megaloblastic anemia due to vitamin B12 deficiency precipitated in a totally gastrectomized type II diabetic patient following the introduction of metformin therapy. Endocrine Journal. 2003;50(4):483-4.
15. Kumthekar AA, Gidwani HV, Kumthekar AB. Metformin associated B12 deficiency. J Assoc Physicians India. 2012 Mar 1;60(3):58-9.
16. Liu KW, Dai LK, Jean W. Metformin-related vitamin B12 deficiency. Age and ageing. 2006 Mar 1;35(2):200-1.
17. Bell DS. Metformin-induced vitamin B12 deficiency presenting as a peripheral neuropathy. Southern medical journal. 2010 Mar 1;103(3):265-7.
18. Pawlak R, James PS, Raj S, Cullum-Dugan D, Lucus D. Understanding vitamin B12. American Journal of Lifestyle Medicine. 2013 Jan;7(1):60-5.
19. Keller M, Redemann B, Chavez-Zander U, Leitzmann C. Effect of cyanocobalamin-fortified toothpaste on vitamin B12 status of vegetarians and vegans. Abstract presented at the 6th International Congress on Vegetarian Nutrition. Loma Linda University. 2013