



ORIGINAL ARTICLE

Vitamin B₁₂ in type 2 diabetic patients treated with metformin[☆]

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Abstract

Objective: To test vitamin B₁₂ plasma levels in type 2 diabetic patients treated with metformin in our area.

Methods: A cross-sectional, observational study of consecutive type 2 diabetic patients on drug treatment attending an internal medicine outpatient clinic.

Results: One hundred and nine patients (81 treated with metformin) were enrolled into the study. Mean time on metformin treatment was 43.5 months and mean drug dose was 1779 mg/day. Patients treated with metformin had significantly lower vitamin B₁₂ plasma levels (393.5 vs 509 pg/mL, $p=0.0008$). Seven (8.6%) of 81 patients treated with metformin and none of the 28 patients not treated with the drug had vitamin B₁₂ plasma levels lower than 197 pg/mL. No correlation was found between vitamin B₁₂ plasma levels and metformin treatment time or dosage.

Conclusions: In type 2 diabetic patients, treatment with metformin is associated to lower vitamin B₁₂ plasma levels. Vitamin B₁₂ deficiency associated with metformin is relatively common in our area.

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Vitamina B₁₂ en pacientes diabéticos tipo 2 en tratamiento con metformina

Resumen

Objetivo: Estudiar los niveles plasmáticos de vitamina B₁₂ en pacientes diabéticos tipo 2 tratados con metformina en nuestro medio.

Métodos: Estudio observacional transversal de pacientes consecutivos con diabetes mellitus tipo 2 en tratamiento farmacológico atendidos en consulta de Medicina Interna.

Resultados: Se estudiaron 109 pacientes (81 en tratamiento con metformina). El tiempo medio en tratamiento con metformina fue 43,5 meses y la dosis media fue 1.779 mg/día. Los pacientes tratados con metformina tuvieron unas concentraciones plasmáticas de vitamina B₁₂ significativamente menores (393,5 frente a 509 pg/ml, $p=0,0008$). Siete (8,6%) de 81 pacientes tratados

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con metformina y ninguno de los 28 no tratados con metformina tuvieron unas concentraciones plasmáticas inferiores a 197 pg/mL. No hubo correlación entre las concentraciones de vitamina B₁₂ y el tiempo en tratamiento con metformina o la dosis de metformina.

Conclusiones: En pacientes diabéticos tipo 2, el tratamiento con metformina se asocia a concentraciones plasmáticas de vitamina B₁₂ más bajas. El déficit de vitamina B₁₂ asociado a metformina es relativamente frecuente en nuestro medio.

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Introduction

Vitamin B₁₂ deficiency is relatively common, particularly in the elderly.^{1,2} Its clinical signs are mainly hematological and neuropsychiatric in nature, and may sometimes be difficult to detect. Vitamin B₁₂ deficiency is usually due to vitamin malabsorption arising from various causes, including the classical pernicious anemia (PA).^{1,2}

As far back as 1971 it was reported that 4 (5.6%) out of 71 diabetic patients on long-term treatment with metformin had low plasma vitamin B₁₂ levels due to impaired absorption.³ Various reports confirming this association have subsequently been published. However, to our knowledge (and a search on MEDLINE), no observational studies have been conducted in Spain comparing patients with type 2 diabetes treated and not treated with metformin.

Methods

An observational, cross-sectional study was conducted on consecutive patients with type 2 diabetes mellitus on drug treatment seen at two internal medicine clinics of a first level hospital in the north of the province of Cáceres. Plasma levels of vitamin B₁₂ were measured in all patients. Vitamin B₁₂ deficiency was defined as levels less than 197 pg/mL (the lower limit of normal in our laboratory). Patients with vitamin B₁₂ deficiency were asked about their dietary habits and antiparietal cell and anti-intrinsic factor antibodies were measured. An upper GI tract endoscopy was also performed to rule out atrophic chronic gastritis when it was considered indicated and with the patient's consent. PA was diagnosed based on plasma vitamin B₁₂ levels less than 197 pg/mL, the presence of anti-intrinsic factor antibodies and/or atrophic chronic gastritis in gastric biopsies, and the response to treatment with vitamin B₁₂ in patients with hematological or neurological evidence of PA.^{1,2}

Statistical analysis was performed using a Chi-square test, and a Fisher exact test when any of the expected values was less than 5, to compare proportions, and a Student's *t*-test for means comparison. A value of $p < 0.05$ was considered statistically significant.

Results

The study sample consisted of 114 patients, of whom 5 (4.4%) were excluded due to PA having been diagnosed. The characteristics of patients treated and not treated with metformin are reported in Table 1. Among the patients treated

with metformin, mean treatment time was 43.5 months (range 6–200 months) and mean dose 1779 mg/day (range 425–2550 mg/day). Patients treated with metformin had significantly lower plasma vitamin B₁₂ levels (393.5 ± 184.2 versus 509 ± 176.4 pg/mL, $p = 0.0008$). Seven (8.6%) of the 81 patients treated with metformin and none of the 28 patients given metformin had plasma vitamin B₁₂ levels less than 197 pg/mL. No patient with vitamin B₁₂ deficiency (excluding those diagnosed with pernicious anemia) had macrocytic anemia, neuropathy, or cognitive impairment.

No correlation was found between metformin dose and plasma vitamin B₁₂ levels ($r = -0.02$, $p = 0.45$) or between treatment time with metformin and plasma vitamin B₁₂ levels ($r = 0.15$, $p = 0.78$). Among the patients treated with metformin, there was no significant difference in plasma vitamin B₁₂ levels between those taking and not taking a proton pump inhibitor (PPI) (409.4 ± 205.3 versus 385.5 ± 174.1 pg/mL, $p = 0.58$).

Discussion

Metformin causes vitamin B₁₂ malabsorption.^{2,3} The absorption of vitamin B₁₂ bound to intrinsic factor produced from ileum is calcium-dependent. Calcium in ileal lumen enhances uptake of the vitamin B₁₂-intrinsic factor complex by the ileal cell receptor, and metformin impairs calcium availability at ileal level.⁴ Various studies, including the current study, support the association of metformin therapy with decreased plasma levels of vitamin B₁₂. A study published in 1976 reported low plasma vitamin B₁₂ levels in 5 (16.7%) out of 30 diabetic patients treated with metformin.⁵ In another retrospective study, patients treated with metformin had significantly lower plasma vitamin B₁₂ levels (496 versus 637 pg/mL).⁶ In a large observational study, 5.8% of diabetic patients over 50 years of age treated with metformin for a mean of 5 years had vitamin B₁₂ deficiency, as compared to 2.4% of diabetic patients not given metformin.⁷ In a prospective study of patients with type 2 diabetes mellitus treated with insulin, 9.9% of patients given metformin at doses of 2,550 mg/day for longer than 4 years had vitamin B₁₂ levels less than 150 pmol/L (200 pg/mL), as compared to 2.7% of patients who received placebo.⁸ These results may be considered similar to those of the present study.

The effect of metformin in decreasing plasma vitamin B₁₂ levels is not transient and appears to increase with treatment duration.^{6,8,9} A progressive decrease over time (during more than 4 years of follow-up) in mean plasma vitamin B₁₂ levels has been reported in patients treated with metformin.^{6,8} A case-control study showed an

Table 1 Characteristics of patients with and without metformin treatment.

	With metformin (n = 81)	Without metformin (n = 28)	p
Age	71.6 ± 12.4	75.4 ± 8.3	NS
Sex (female)	56 (69.1%)	16 (57.1%)	NS
BMI (kg/m ²)	30.4 ± 5.4	29.3 ± 3.2	NS
Treatment with PPIs	27 (33.3%)	16 (57.1%)	0.02
Treatment with insulin	17 (21%)	11 (39.3%)	NS
HbA _{1c} (%)	7 ± 1.3	6.9 ± 0.9	NS

HbA_{1c}: glycosylated hemoglobin; PPIs: proton pump inhibitors; BMI: body mass index; NS: no statistically significant ($p \geq 0.05$).

association between vitamin B₁₂ deficiency and the dose and duration of metformin treatment.⁹ By contrast, our study found no correlation between metformin treatment dose and duration and plasma vitamin B₁₂ levels. It should be noted, however, that short-term treatment with metformin already decreases vitamin B₁₂ levels. In a 6-month study, treatment with metformin caused a mean 20 pg/mL reduction in vitamin B₁₂ levels.¹⁰ An additional 16-week study also showed a 14% reduction in plasma vitamin B₁₂ levels.¹¹ The use of PPIs has also been associated with vitamin B₁₂ deficiency.¹² Acid secretion inhibition impairs vitamin B₁₂ release from food.¹³ However, our study found no negative impact of treatment with PPIs on plasma vitamin B₁₂ levels. This observation agrees with the findings in another study.⁹

All the patients in this study with vitamin B₁₂ deficiency associated with metformin had so-called "asymptomatic" deficiency (defined as low plasma vitamin B₁₂ levels with no associated macrocytic anemia, neuropathy, or cognitive impairment).^{1,2} However, this deficiency is not always asymptomatic. In a series of 10 patients with vitamin B₁₂ deficiency associated with metformin, 9 patients had mild anemia and 3 had peripheral neuropathy.¹⁴ Isolated cases of patients with symptomatic vitamin B₁₂ deficiency associated with metformin have also been reported.¹⁵ There is no agreement as to whether patients with "asymptomatic" vitamin B₁₂ deficiency should be treated.^{1,2} There are signs of vitamin B₁₂ deficiency, particularly neurological signs, which are difficult to diagnose and may become irreversible.² In addition, vitamin B₁₂ deficiency is invariably associated with elevated homocysteine levels^{8,11} and their potentially harmful consequences.^{16,17} On the other hand, one study did not show an improvement in cognitive function after vitamin B₁₂ administration to elderly patients with mild vitamin B₁₂ deficiency.¹⁸

An additional concept, functional vitamin B₁₂ deficiency, is defined as the presence of normal vitamin B₁₂ levels together with increased plasma levels of methylmalonic acid and is based on the fact that vitamin B₁₂ deficiency invariably causes elevated methylmalonic acid levels.¹⁹⁻²¹ Such a functional deficiency has been associated with neuropathy and anemia.¹⁹⁻²¹ An increased frequency of neuropathy has recently been reported in patients with type 2 diabetes mellitus and this functional deficiency, as well as an improvement in neuropathy after the administration of vitamin B₁₂ and the normalization of plasma levels of methylmalonic acid.²¹

There is no agreement regarding the convenience of regular measurements of plasma vitamin B₁₂ levels in patients treated with metformin.^{8,9,14,15,22} Controversy also exists

concerning the management of patients with vitamin B₁₂ deficiency associated with metformin, and some authors even suggest that metformin should be discontinued. In agreement with other authors,^{3,8} we think it is appropriate to regularly measure plasma vitamin B₁₂ levels in patients treated with metformin, although other authors advise against such measurements.²² Moreover, we think it is appropriate to continue treatment with metformin in patients with vitamin B₁₂ deficiency and to treat such patients with oral or intramuscular vitamin B₁₂ even if the deficiency is "asymptomatic". In our experience (data not collected in the study) and that of other authors,¹⁴ plasma vitamin B₁₂ levels easily normalized after the administration of oral or intramuscular vitamin B₁₂. This approach is based on the benefits of metformin for patients with type 2 diabetes mellitus and on the fact that treatment with vitamin B₁₂ is simple, inexpensive, and safe and also has potential benefits.^{1,2} Oral calcium supplements may reverse vitamin B₁₂ malabsorption induced by metformin, and could be a treatment option.⁴

Conflicts of interest

The authors state that they have no conflicts of interest.

References

- Dali-Youcef N, Andrès E. An update on cobalamin deficiency in adults. *QJM*. 2009;102:17-28.
- Stabler SP, Lindenbaum J, Allen RH. Vitamin B-12 deficiency in the elderly: current dilemmas. *Am J Clin Nutr*. 1997;66:741-9.
- Tomkin GH, Hadden DR, Weaver JA, Montgomery DA. Vitamin-B12 status of patients on long-term metformin therapy. *BMJ*. 1971;2:685-7.
- Bauman WA, Shaw S, Jayatilleke E, Spungen AM, Herbert V. Increased intake of calcium reverses vitamin B12 malabsorption induced by metformin. *Diabetes Care*. 2000;23:1227-31.
- Carpentier JL, Bury J, Luyckx A, Lefebvre P. Vitamin B 12 and folic acid serum levels in diabetics under various therapeutic regimens. *Diabete Metab*. 1976;2:187-90.
- Kos E, Lizek MJ, Emanuele MA, Durazo-Arvizu R, Camacho P. The effect of metformin therapy on vitamin D and B12 levels in patients with diabetes mellitus type 2. *Endocr Pract*. 2012;18:179-84.
- Reinstatler L, Qi YP, Williamson RS, Garn JV, Oakley Jr GP. Association of biochemical B12 deficiency with metformin therapy and vitamin B12 supplements: the national health and nutrition examination survey, 1999-2006. *Diabetes Care*. 2012;35:327-33.

8. de Jager J, Kooy A, Lehert P, Wulffélé MG, van der Kolk J, Bets D, et al. Long term treatment with metformin in patients with type 2 diabetes and risk of vitamin B-12 deficiency: randomised placebo controlled trial. *BMJ*. 2010;340:c2181.
9. Ting RZ, Szeto CC, Chan MH, Ma KK, Chow KM. Risk factors of vitamin B(12) deficiency in patients receiving metformin. *Arch Internal Med*. 2006;166:1975-9.
10. Sahin M, Tutuncu NB, Ertugrul D, Tanaci N, Guvener ND. Effects of metformin or rosiglitazone on serum concentrations of homocysteine, folate, and vitamin B12 in patients with type 2 diabetes mellitus. *J Diabetes Complications*. 2007;21:118-23.
11. Wulffélé MG, Kooy A, Lehert P, Bets D, Ogterop JC, Borger van der Burg B, et al. Effects of short-term treatment with metformin on serum concentrations of homocysteine, folate and vitamin B12 in type 2 diabetes mellitus: a randomized, placebo-controlled trial. *J Internal Med*. 2003;254:455-63.
12. McColl KE. Effect of proton pump inhibitors on vitamins and iron. *Am J Gastroenterol*. 2009;104 Suppl. 2:5-9.
13. Varughese GI, Scarpello JH. Metformin and vitamin B12 deficiency: the role of H2 receptor antagonists and proton pump inhibitors. *Age Ageing*. 2007;36:110-1.
14. Andrès E, Noel E, Goichot B. Metformin-associated vitamin B12 deficiency. *Arch Internal Med*. 2002;162:2251-2.
15. Liu KW, Dai LK, Jean W. Metformin-related vitamin B12 deficiency. *Age Ageing*. 2006;35:200-1.
16. Smulders YM, Blom HJ. The homocysteine controversy. *J Inherit Metab Dis*. 2011;34:93-9.
17. Khanna S, Kapoor P, Pillai KK, Vohora D. Homocysteine in neurological disease: a marker or a cause? *CNS Neurol Disord Drug Targets*. 2011;10:361-9.
18. Eussen SJ, de Groot LC, Joosten LW, Bloo RJ, Clarke R, Ueland PM, et al. Effect of oral vitamin B-12 with or without folic acid on cognitive function in older people with mild vitamin B-12 deficiency: a randomized, placebo-controlled trial. *Am J Clin Nutr*. 2006;84:361-70.
19. Solomon LR. Disorders of cobalamin (vitamin B12) metabolism: emerging concepts in pathophysiology, diagnosis and treatment. *Blood Rev*. 2007;21:113-30.
20. Turner MR, Talbot K. Functional vitamin B12 deficiency. *Pract Neurol*. 2009;9:37-41.
21. Solomon LR. Diabetes as a cause of clinically significant functional cobalamin deficiency. *Diabetes Care*. 2011;34:1077-80.
22. Vidal-Alaball J, Butler CC. Reduced serum vitamin B-12 in patients taking metformin. *BMJ*. 2010;340:c2198.