

# Evaluation of vitamin B<sub>6</sub> supplementation in Wilson's disease patients treated with D-penicillamine

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## ABSTRACT

**Introduction** Wilson's disease (WD) is a copper metabolism disorder characterised by a progressive accumulation of this metal mainly in the liver and the brain. Treatment is based on the removal of copper operated by the chelators, among which, D-penicillamine (DP) is prescribed as a first-line treatment in most situations. There is some evidence in linking the use of DP with a risk of vitamin B<sub>6</sub>; therefore, vitamin supplementation is sometimes recommended, although non-consensually. The objective of our study was to evaluate the level of vitamin B<sub>6</sub> in WD patients treated with DP with and without associated supplementation.

**Methodology** All WD patients followed at the National Reference Centre for WD in Lyon between January 2019 and December 2020 treated with DP for more than 1 year were included and separated in two groups according to vitamin B<sub>6</sub> supplementation. The level of vitamin B<sub>6</sub> was measured by the determination of pyridoxal phosphate (PLP).

**Results** A total of 37 patients were included. Average age of 23.3±14.8 years, 15 patients with <18 years. Median duration of treatment was 51 (55.8) months. 15 patients were under vitamin B<sub>6</sub> supplementation and 22 had interrupted it for more than 1 year. The median PLP level was significantly higher in the group with supplementation, 137.2 (86.7) nmol/L vs 64.9 (30.8) nmol/L (p<0.01). No patient had a PLP level<35 nmol/L.

**Conclusion** Long-term stable WD patients under DP treatment probably do not need vitamin B<sub>6</sub> supplementation.

## INTRODUCTION

Wilson's disease (WD) is a rare, autosomal recessive disorder characterised by a progressive toxic accumulation of copper, mainly in the liver and central nervous system.<sup>1-3</sup> This accumulation is responsible for liver damage and secondarily for neurological and psychiatric symptoms due to copper mobilisation. The disease is exceptionally symptomatic before 3 years and after 50 years.<sup>1,4</sup> WD progresses to cirrhosis and irreversible central nervous system damage without early diagnosis and appropriate treatment.<sup>1-4</sup> Treatment of WD is based on lifetime

### WHAT IS ALREADY KNOWN ON THIS TOPIC

⇒ The antipyridoxine effect of D-penicillamine (DP) has been demonstrated, but there is no consensus regarding systematic vitamin B<sub>6</sub> supplementation.

### WHAT THIS STUDY ADDS

⇒ This is the first study addressing this subject after nearly four decades. No case of vitamin B<sub>6</sub> deficiency was observed in the not supplemented group. Most patients in the supplemented group had higher-than-normal vitamin B<sub>6</sub> levels.

### HOW THIS STUDY MIGHT AFFECT RESEARCH, PRACTICE OR POLICY

⇒ This study provides new evidence from a real-world experience unfavourable to recommending systematic vitamin B<sub>6</sub> supplementation when using DP.

administration of either copper chelators or copper absorption inhibitors associated with a low copper diet.<sup>1 2 5 6</sup> D-penicillamine (DP), a copper chelator and inducer of endogenous metallothioneins, is the first-line chelator for symptomatic WD. This drug is frequently responsible for adverse effects that require its discontinuation in up to 30% of cases.<sup>5-7</sup> These effects are multiple and comprise notably cutaneous (rashes), haematological (neutropenia, thrombocytopenia and lymphadenopathy), renal (proteinuria) and immunological effects.<sup>8,9</sup>

Vitamin B<sub>6</sub>, or pyridoxine, is a generic term that includes a group of six chemically related compounds with a common pyridine ring. The phosphorylated derivative, known as pyridoxal 5'-phosphate (PLP), is the biologically active form of vitamin B<sub>6</sub> that serves as a cofactor in more than 160 different catalytic reactions, particularly in the synthesis of haem, nucleic acids, as well as lipid, carbohydrate, amino acid metabolism.<sup>10 11</sup>

Studies carried out more than 40 years ago demonstrated an antipyridoxine effect of DP manifested by an increase in the urinary



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secretion of tryptophan metabolites without any clinical sign.<sup>12–14</sup> Pyridoxin deficiency has not been observed in animals or patients receiving DP,<sup>12–15</sup> however; few reports have found a potential link between cases of optic neuropathy and vitamin B<sub>6</sub> deficiency secondary to administration of DP.<sup>16–19</sup> Based on these studies, the European Association for the Study of the Liver (EASL) recommends routine vitamin B<sub>6</sub> supplementation in all patients receiving DP.<sup>5</sup> Some studies suggested that the supplementation may especially be needed in case of nutritional deficit and at times of growth spurts.<sup>12–14</sup> The paediatric recommendations for WD published by the European Society for Paediatric Gastroenterology, Hepatology and Nutrition (ESPGHAN) only indicate that there is a low level of evidence of pyridoxine deficiency associated with DP treatment and probably a high vitamin B<sub>6</sub> intake in a regular diet.<sup>6</sup>

Therefore, we aimed to assess the effect of DP on vitamin B<sub>6</sub> levels and evaluate the need for vitamin supplementation in the long-term treatment of WD patients with DP.

## PATIENTS AND METHODS

We carried out a retrospective review of medical records of WD patients treated with DP for >12 months year between January 2019 and December 2020 and followed at the national reference centre for WD in Lyon (France). Demographic, clinical and laboratory data were collected, including DP dose, vitamin B<sub>6</sub> supplementation and blood pyridoxine level.

Vitamin B<sub>6</sub> supplementation is routinely prescribed in our centre for patients at the start of DP treatment, 125 mg for children and 250 mg/week for adults. This supplementation is often later discontinued at the request of the patient or by the prescribing physician to improve adherence to the chelator treatment. Patients who discontinued vitamin supplementation for >12 months were compared with those who continued vitamin B<sub>6</sub> supplementation.

Vitamin B<sub>6</sub> level was determined by the quantification in whole blood of PLP, its biologically active form. This assay was carried out using high-performance liquid chromatography. Normal values ranged from 35.0 to 110.0 nmol/L. The quantification of PLP has been added to the routine follow-up of WD patients treated with DP since 2019.

Qualitative variables are presented as relative frequency (percentage), and as mean and SD for normally distributed quantitative variables and median (IQR) for non-normally distributed variables. Comparisons were made using the Mann-Whitney non-parametric test using SPSS software V.23.0 (IBM Corp. Armonk, NY).  $P < 0.05$  was considered significant.

This analysis was listed as a clinical practice audit, under the control of the research committee of our structure, and therefore did not require ethics approval. However, a note was sent to the institutional review board (CPP: Comités de protection des personnes) for information

and approval before the publication of the results of the study.

## RESULTS

### Study population

A total of 37 patients were included. The mean±SD age at inclusion was 23.3±14.8 years (range: 6–63); 15 patients (40.5%) were <18 years of age. The mean±SD age at diagnosis was 9.7±5.2 years. Thirty (81.1%) patients had hepatic manifestations at diagnosis, and seven (18.9%) had neurological manifestations. The median (IQR) duration of DP treatment was 59 (21.5–162.5) months. The mean±SD dose of DP was 17.4±5.3 mg/kg/day (table 1).

Two patients had moderate undernutrition, and there was no case of pregnancy.

### Vitamin B<sub>6</sub> supplementation

Of the 37 patients, 15 were supplemented with pyridoxine, and 22 has discontinued. There was no significant difference between these two groups in terms of mean age at inclusion, mean age at diagnosis, clinical form, mean dose of DP, median duration of treatment and mean level of transaminases and exchangeable copper (table 1).

### Pyridoxal phosphate (PLP)

The median (IQR) PLP level was significantly higher in the group with supplementation (137.2 (104.9–191.6) nmol/L) than in the group without supplementation (64.9 (2.6–83.4) nmol/L,  $p < 0.01$ ). No patient had a PLP level <35 nmol/L, defining a biological deficit in pyridoxal; 11/15 patients (73%) in the supplemented group had a PLP level higher than normal (> 110 nmol/L), while none in the group without supplementation did so ( $p < 0.01$ ; table 1). No patient had any clinical signs of vitamin B<sub>6</sub> deficiency. No patient presented with any symptom attributable to vitamin B<sub>6</sub> toxicity that led to discontinuing the supplementation.

## DISCUSSION

There is little published data on vitamin B<sub>6</sub> supplementation for patients treated with DP, and no consensus recommendation exists. Here, there was no case of vitamin B<sub>6</sub> deficiency defined by PLP level or clinical manifestation potentially attributable to this deficiency. Between 1963 and 1991, four cases of optic neuropathy were identified in patients treated with DP.<sup>16–19</sup> In three cases, this complication occurred between 13 and 30 months of treatment using dosages between 1 and 2 g per day. Only one case showed elevated urinary tryptophan metabolites indicating an antipridoxin effect. All these patients were supplemented with variable doses of vitamin B<sub>6</sub> in addition to a decrease in the DP dosage or a change in chelation therapy; one patient had also received corticosteroid therapy due to the concomitant appearance of antinuclear antibodies. All of them evolved

**Table 1** Demographic, clinical and laboratory characteristics

	Total n=37	Suppl. B <sub>6</sub> vit. n=15	No suppl. B <sub>6</sub> vit. n=22	P value
Sex M, n (%)	22 (59.5)	8 (53.3)	14 (63.6)	
Age at inclusion*	23.3±14.8	19.0±11.6	26.3±16.3	
Age at diagnosis*	9.7±5.2	10.0±4.4	9.5±5.8	
<18 years, n (%)	15 (40)	8 (53)	7 (32)	
Hepatic manifestations	30 (81)	11 (73)	19 (86)	
Neurological manifestations	7 (19%)	4 (27%)	3 (14%)	
Dose DP (mg)*	948.6±369.2	830±334.8	1029.5±376.9	
Dose (mg/kg)*	17.4±5.3	16.3±4.4	18.1±5.8	
Duration of treatment in months†	59 (21.5–162.5)	51 (22.3–78.1)	78.2 (21.1–224.7)	
PLP level (nmol/L)†	81.8 (61.9–117.4)	137.2 (104.9–191.6)	64.9 (52.6–83.4)	p<0.05
PLP<35 (nmol/L), n (%)	0	0	0	
PLP>110 (nmol/L), n (%)	11 (30)	11 (73)	0	p<0.05
Hb (g/L)*	14.1±1.5	14.0±1.5	14.1±1.6	
WBC (G/L)*	6.4±1.5	5.8±1.4	6.8±1.8	
Platelets (G/L)*	256.7±104.4	238.2±125.8	269.3±87.0	
ALT (U/L) *	46.9±33.8	41.5±32.8	50.6±34.7	
Elevated ALT, n (%)	11 (30)	5 (33)	6 (27)	
ASAT (U/L)*	34.5±10.9	32.6±11.5	50.6±34.7	
Elevated AST, n (%)	10 (27)	3 (20)	7 (32)	
GGT (U/L)*	36.9 (21.1)	33.1±21.7	39.5±20.7	
Elevated GGT, n (%)	16.0 (43)	6 (40)	10.0 (45)	
Alkaline phosphatase (U/L)*	158.0±119.4	175.2±120.8	147.4±120.2	
Exchangeable copper (umol/L)*	0.8±1.7	1.2±0.9	0.6±0.5	

\*Mean±SD.  
†Median (IQR).  
DP, D-penicillamine; PLP, pyridoxal phosphate.

into full vision recovery. Attributing these complications to the antipyridoxine effect of DP remains challenging to prove, considering that in one case, optic neuropathy appeared in the first weeks of treatment using DP in low doses; in another case, a vitamin B<sub>6</sub> supplementation was prescribed concomitantly.

In the supplemented group, more than three-quarters of patients had above-normal PLP levels, but we did not find any neurological manifestation that could be attributed to vitamin B<sub>6</sub> overdose; however, patients were assessed only by clinical neurological examination without a dedicated electrophysiology study. Multiple publications have described a link between the occurrence of peripheral neuropathy and vitamin B<sub>6</sub> overdose.<sup>20</sup> In most published cases, this complication appears with large doses of vitamin B<sub>6</sub> (2–6 g/d)<sup>21</sup>; but also with 50–600 mg/day dosages over several months to several years.<sup>20</sup>

All the patients included in this study received DP for a relatively long time, about 4 years on average, so these

results should not be extrapolated to patients in the initial phases of treatment.

Without an established toxicity threshold, it is recommended not to exceed 25–50 mg/day of vitamin B<sub>6</sub>.<sup>22 23</sup> Due to the ubiquity of vitamin B<sub>6</sub> in common animal and plant foods, the need for vitamin B<sub>6</sub> is generally satisfied. Supplementation may be necessary in exceptional cases (malnutrition and pregnancy). Our cohort only had two cases of moderate undernutrition and no pregnancy.

This study has many limitations: the retrospective and descriptive design, the small number of patients, the absence of a dietary evaluation and the predominance of chronic patients with long-term DP treatment.

Vitamin B<sub>6</sub> level assessment or monitoring of clinical symptoms of vitamin toxicity are not routinely recommended during WD follow-up. However, we recommend thoroughly assessing both until more firm evidence is available.



## CONCLUSION

No signs of vitamin B<sub>6</sub> deficiency were found in a group of long-term stable WD patients treated with DP who did not receive vitamin supplementation. Prospective studies are required to help determine if routine vitamin B<sub>6</sub> supplementation needs to be routinely recommended.

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## REFERENCES

- Czlonkowska A, Litwin T, Dusek P, *et al.* Wilson disease. *Nat Rev Dis Primers* 2018;4:21.
- Liu J, Luan J, Zhou X, *et al.* Epidemiology, diagnosis, and treatment of Wilson's disease. *Intractable Rare Dis Res* 2017;6:249–55.
- Schilsky ML. Wilson disease: diagnosis, treatment, and follow-up. *Clin Liver Dis* 2017;21:755–67.
- Ferenci P. Diagnosis of Wilson disease. *Handb Clin Neurol* 2017;142:171–80.
- European Association for Study of Liver. EASL clinical practice guidelines: Wilson's disease. *J Hepatol* 2012;56:671–85.
- Socha P, Janczyk W, Dhawan A, *et al.* Wilson's disease in children: a position paper by the Hepatology committee of the European Society for Paediatric Gastroenterology. *J Pediatr Gastroenterol Nutr* 2018;66:334–44.
- Aggarwal A, Bhatt M. Advances in treatment of Wilson disease. *Tremor Other Hyperkinet Mov (N Y)* 2018;8:525.
- Weiss KH, Thurik F, Gotthardt DN, *et al.* Efficacy and safety of oral chelators in treatment of patients with Wilson disease. *Clin Gastroenterol Hepatol* 2013;11:1028–35.
- Medici V, Trevisan CP, D'Inca R, *et al.* Diagnosis and management of Wilson's disease: results of a single center experience. *J Clin Gastroenterol* 2006;40:936–41.
- Ueland PM, Ulvik A, Rios-Avila L, *et al.* Direct and functional biomarkers of vitamin B6 status. *Annu Rev Nutr* 2015;35:33–70.
- Parra M, Stahl S, Hellmann H. Vitamin B6 and its role in cell metabolism and physiology. *Cells* 2018;7:84.
- Rumsby PC, Shepherd DM. The effect of penicillamine on vitamin B6 function in man. *Biochem Pharmacol* 1981;30:3051–3.
- JAFFE IA, ALTMAN K, MERRYMAN P. The antipyridoxine effect of penicillamine in man. *J Clin Invest* 1964;43:1869–73.
- Gibbs K, Walshe JM. Penicillamine and pyridoxine requirements in man. *Lancet* 1966;1:175–9.
- Porlas RV, de Castillo LLC, Dioquino CPC. Neurologic Wilson disease: case series on a diagnostic and therapeutic emergency. *Dialogues Clin Neurosci* 2018;20:341–5.
- TU J, BLACKWELL RQ, LEE PF. DL-penicillamine as a cause of optic axial neuritis. *JAMA* 1963;185:83–6.
- Lee AH, Lawton NF. Penicillamine treatment of Wilson's disease and optic neuropathy. *J Neurol Neurosurg Psychiatry* 1991;54:746.
- Goldstein NP, Hollenhorst RW, Randall RV. Possible relationship of optic neuritis, Wilson's disease, and DL-penicillamine therapy. *JAMA* 1966;196:734.
- Klinge TG, Burde RM. Optic neuropathy associated with penicillamine therapy in a patient with rheumatoid arthritis. *J Clin Neuroophthalmol* 1984;4:75–8.
- Malet L, Dayot L, Moussy M, *et al.* Peripheral neuropathy with hypervitaminosis B6 caused by self-medication. *Rev Med Interne* 2020;41:126–9.
- Schaumburg H, Kaplan J, Windebank A, *et al.* Sensory neuropathy from pyridoxine abuse. A new megavitamin syndrome. *N Engl J Med* 1983;309:445–8.
- Kulkantrakorn K. Pyridoxine-induced sensory ataxic neuropathy and neuropathy: revisited. *Neurol Sci* 2014;35:1827–30.
- Turck D, Bohn T, Castenmiller J, *et al.* Guidance for establishing and applying tolerable upper intake levels for vitamins and essential minerals: draft for internal testing. *EFSA J* 2022;20:e200102.