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The Effectiveness of Parenteral Hydroxylcobalamin on Urine Mma in Children with Methylmalonic Acidemia

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Abstract

Background: Methylmalonic acidemia is a rare autosomal recessive disease in which there is a deficiency of intracellular cobalamin. This study aimed to assess the effectiveness of parenteral hydroxylcobalamin in the treatment of children with methylmalonic acidemia.

Method: This is a quasi-experimental study without a control group. The participants included under-18-year-old children with confirmed methylmalonic acidemia. There were only 17 patients with the inclusion criteria, all of whom were included in the study. They received 1mg hydroxylcobalamin injection for 3 or 7 days based on their clinical status. Data was gathered by a demographic questionnaire, along with laboratory tests of urine-MMA, and plasma homocysteine, measured before and after the intervention. Data analyses were performed using SPSS v. 26.

Results: The samples included 17 patients, most of whom were males (52.9%). They had various clinical manifestations consisting of hypotonia, seizure, verbal disorders, movement disorders, organomegaly, hematologic disorders, and ophthalmic disorders. The parenteral hydroxylcobalamin had a borderline significant effect on urine-MMA (p=0.05); this seems to be due to the sample shortage and can become strongly significant with sample increase.

Conclusion: The results revealed the effectiveness of parenteral hydroxylcobalamin in MMA patients. However, there is no standard guideline to suggest the perfect dose of it to acquire the optimum result; so it is suggested to conduct more clinical trials or cohort studies to be done.

Key Words: Children, Hydroxylcobalamin, Methyl malonic acidemia.

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1- INTRODUCTION

Methylmalonic acidemia (MMA) is a rare autosomal recessive disease in which there is a deficiency in conversion of methylmalonic CoA to succinyl CoA (1). This may occur alone or in combination with other biochemical abnormalities such as elevation of homocysteine and low methionine. Elevation of methylmalonic acid may be due to a defect in the metabolism of methylmalonyl-coenzyme A (CoA) or cobalamin (Cbl)/vitamin B12 (2). Clinical manifestations of MMA are widespread and can range from benign to a fetal disease. Usually, the age of presenting the symptoms is, generally, divided into early onset and late onset categories. In the early onset group, the first symptoms appear in infancy and include lethargy, vomiting, tachypnea, nutritional problems, hypotonia, and sepsis-like presentation which can lead to death if left untreated (3, 4). In the late onset group, which is rare and milder, the symptoms of hypotonia, developmental delay, and neurologic disorders appear in the next stages of childhood. Most of these children have normal IQ and mental development (5). Generally, the long-term treatment of MMA is restricted protein diet, along with L-carnitine supplement; and chronic use of bicarbonate might be essential for treating metabolic acidosis. Also, some patients may benefit from parenteral hydroxylcobalamin administration (6); though their responses are different based on the type of the MMA. Nearly all CblA patients respond to vitamin B12; but less than half of CblB patients respond and rarely patients with –mut type have an in vivo biochemical response (7, 8). Therefore, due to the challenging worldwide evidence regarding the use of hydroxylcobalamin in the treatment of MMA, and no evidence in Iran, we conducted this study to assess the effectiveness of parenteral administration of hydroxylcobalamin in the treatment of MMA.

2- MATERIAL AND METHODS

2-1. Design and participants

This is a quasi-experimental before-after study without control group which was done in Akbar hospital, a tertiary pediatric hospital, in Mashhad. It was approved by the ethics committee of Mashhad University of Medical Sciences (IR.MUMS.MEDICAL.REC.1400.828) in March 2022. Under-18-year-old patients with a diagnosis of isolated MMA or MMA with homocystinuria, confirmed by MS/MS and urine organic acid or genetic test, who were willing to participate, were included in the study and they were excluded if they hadn't enough cooperation with the treatment protocol or refused to continue the study. There were only 17 patients with confirmed MMA diagnosis, referring to Akbar Hospital, who had the inclusion criteria; so, all of them were recruited for the study. The study purpose was explained to them and informed consents were completed by their parents.

2-2. Data Collection

Data was gathered by a demographic questionnaire including age, gender, weight, height, parents' education, and history of MMA in other siblings, history of metabolic disorders in relatives, history of abortion or stillbirth, age of symptoms presentation, age of diagnosis confirmation, developmental delays, physical symptoms, drug history, and patient's diet. The IQ level of each patient was evaluated by a Goodenough draw-a-person test. In this test each person asked to make a picture of a person as best as he/she can do it, then each drawn part of the body scored at least 1 up to 7. Also, urine-MMA was checked before and after the experiment. The pre experimental test of urine-MMA was done after one week of

discontinuing the patient's previous dose of cobalamin.

2-3. Procedure

The intervention consisted of two protocols for clinically stable and unstable patients. In clinically stable patients 1mg hydroxylcobalamin was administered intramuscular for 3 days and in clinically unstable ones 1mg hydroxylcobalamin was administered intravenously for 7 days. The post experimental test of urine-MMA was done a day after the completion of hydroxylcobalamin injections. The urine-MMA (mmol/mol creatinine) test was assessed by Liquid chromatography–mass spectrometry (LC-MS/MS). Moreover, in 2 patients who had MMA with homocystinuria the plasma homocysteine (micromole/L) was checked by high pressure liquid chromatography (HPLC) before and after the experiment.

2-4. Data Analysis

The data was analyzed by SPSS, version 26, by the use of descriptive tests (mean, standard deviation, frequency, and percentage) and analytical tests (Mann-Whitney, Fisher's exact test, Wilcoxon, and spearman correlation). In all statistical tests, the significance level was considered less than 0.05.

3- RESULTS

The sample includes 17 patients, most of them were male (52.9%), and the parents' education was not academic (mothers: 70.6%, and fathers 64.7%) in most participants. There were histories of other siblings' metabolic disease or death in 5 cases (29.3%), and histories of metabolic disease in relatives in 4 cases (23.5%). Other demographic data were demonstrated in **Tables 1** and **2**.

Six patients had done genetic tests before the study showing 0 mut type (complete absence of enzyme function) in one patient, –mut type (the presence of enzyme with abnormal function) in 1 patient, 2 were CblB (isolated MMA), and 2 were CblC (MMA with homocystinuria) cases.

Kolmogrov-Smirnov test showed that urine-MMA didn't have a normal distribution; so for analyzing the effect of intervention on it, Wilcoxon test was implemented. The result showed a borderline significant effect of parenteral hydroxylcobalamin on urine-MMA ($p=0.05$) which can be attributed to the sample shortage ($n=17$) and most probably becomes more significant by the sample size increase. The patients with -mut, CblC type, and one with CblB type had good responses to hydroxylcobalamin and their urine-MMA decreased more than 50% after the intervention; but one patient with 0mut and the other patients with CblB type had no response to hydroxylcobalamin. Also, in CblC samples whose plasma homocysteine was measured, more than 50% decrease was seen after the hydroxylcobalamin injections.

Table-1: Quantitative demographic characteristics of patients with MMA

Variable	Mean	SD	Min	Max	Percentile		
					25	50	75
Age (year)	8.06	2.28	5.30	13.80	6.10	7.50	9.90
Age of presentation (months)	5.88	0.55	1.00	18.00	1.00	5.00	8.50
Age of confirmation (months)	13.11	10.90	2.00	48.00	7.50	11.00	16.50
Weight Z-score	-1.29	2.21	-7.00	1.90	-1.90	-1.40	0.45
Height Z-score	-1.55	1.84	-5.00	1.80	-2.85	-1.30	-0.55

Table-2: Qualitative demographic characteristics of patients with MMA

Variable		Frequency	Percent
History of abortion		4	23.5
History of stillbirth		5	29.4
History of developmental delay		8	47.1
Physical symptoms	Hypotonia	2	11.8
	Seizure	5	29.4
	Verbal disorders	7	41.2
	Movement disorders	4	23.5
	Organomegaly	2	11.8
	Hematologic disorders	11	64.7
	Ophthalmic disorders	2	11.8
IQ Level	Normal (90-110)	8	47.1
	High borderline (80-89)	3	17.6
	Low borderline (70-79)	2	11.8
	Mild intellectual disability (50-69)	2	11.8
	Moderate intellectual disability (30-49)	1	5.9
	Intensive intellectual disability (<30)	1	5.9
B12 supplement	Injection Hydroxylcobalamin	2	11.8
	Oral liposomal B12 (oral hydroxylcobalamin)	5	29.4
	Oral use of B12 shots (cyanocobalamin)	10	58.8
Special formula	Yes	8	47.1
	No	9	52.9

Other analyses revealed no significant difference between the responses to hydroxylcobalamin based on age (age of presenting symptoms, age of confirming the disease, and the patient's age), gender, weight, height, developmental delay, hypotonia, seizure, verbal disorders, movement disorders, IQ score, organomegaly, hematologic disorders, ophthalmic involvement, and special formula usage ($P > 0.05$).

4- DISCUSSION

In present study the effectiveness of parenteral hydroxyl-cobalamin on urine-MMA was assessed among 17 children with confirmed isolated MMA or with homocystinuria. 64.7% of the patients had positive response to hydroxylcobalamin and their urine-MMA was decreased more than 50%, which is considered as the criteria for response to the treatment (9). One of the effective factors on the patients' responses was the type of gene mutation. As mentioned before, the patient with 0mut and one of the patients with CblB type didn't respond to hydroxylcobalamin. Other studies have also had similar results; for example, Baumgartner (2014) showed that the type of gene mutation had an effect on the symptoms and the patients' response to treatment (10). Or, in another study, Fowler et al. (2008) found out that all patients with CblA and -mut, and one third of patients with CblB and CblD responded to treatment with hydroxyl-cobalamin (11). Although Yu et al. (2021) reported that completely and partially responsive patients to vitamin B12 had less clinical symptoms and optimum laboratory results (12), no difference was seen in our study between the symptoms and the responsiveness of patients to vitamin B12.

This might be due to the results of neonatal screening, early detection, and treatment of MMA before sequels development in children in China, while in our country we have neonatal metabolic screening test only for phenylketonuria, galactosaemia, and congenital hypothyroidism. This delay in detecting and treatment of MMA can affect the IQ level of children, too. Although only 47.1% of our patients had normal IQ levels, in other studies most of MMA patients had normal IQ levels. For example, Zhang et al. (2020) showed that 67.4% of patients with MMA had normal IQ levels and near 37% had just little developmental delay (5).

There was a difference between the type of MMA and its clinical manifestations. In patients with CblC more problems were observed, including developmental delay, low IQ level, seizure, verbal disorder, movement disorder, and megaloblastic anemia. This finding is consistent with those of other studies showing that neurologic problems and megaloblastic anemia are more common in CblC patients (4, 13). Although, CblC seems to be the most common defect in intracellular cobalamin metabolism associated with methylmalonic acidemia and homocystinuria, it has late clinical presentations and this may lead to a diagnostic delay and subsequently more sequels. In this regard, Brox-Torrecilla et al. (2021) reported on a 45-year-old case of CblC MMA with a 20-year history of kidney disease which had resulted in kidney transplantation (14). Other patients with 0mut, -mut, and CblB MMA had less clinical manifestation in comparison to those with CblC; this might be attributed to late onset and late diagnosis of CblC type which can cause delay in treatment and prevent the side effects of the disease.

Our results demonstrated that urine-MMA and plasma homocysteine decreased more than 50% after 1-mg-per-day parenteral administration of hydroxyl-cobalamin. This is notable that despite hydroxlcobalamin is considered as a common treatment in inborn errors of cobalamin metabolism, formal dosing guidelines do not exist for it. So, studies have used different doses of hydroxylcobalamin; for example, Obeid et al. (2015) demonstrated that 1-2 mg of parenteral hydroxyl-cobalamin has had obvious advantages in treating inborn errors of cobalamin metabolism (15). But, Carrillo-Carrasco et al. (2009) reported that by progressively increasing the dose of hydroxylcobalamin from 1 to 20 mg IM per day, a dose-dependent response was observed with 80% reduction of plasma MMA, 55% reduction of total homocysteine, and a greater than twofold increase in methionine (4). Although, they reported that higher doses of hydroxlcobalamin might have accompanied by better responses, it is unknown whether it may slow down or eliminate other complications. Therefore, it seem necessary to conduct more clinical trials to determine the effectiveness of different doses of hydroxylcobalamin therapy in patients with MMA.

5- CONCLUSION

The results manifested that there is a positive and direct correlation between urine-MMA before and after the administration of parenteral hydroxylcobalamin, confirming the effectiveness of it in MMA patients. However, there is no standard guideline to suggest the perfect dose of it to acquire the optimum result. Thus, more clinical trials or cohort studies are suggested to be conducted in this respect. Moreover, the importance of neonatal metabolic screening in early detection, treatment, and prevention of MMA complications in children was emphasized.

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7- REFERENCES

1. Eshraghi P. A methylmalonic acidemia case presenting with acrodermatitis enteropathica. International Journal of Pediatrics. 2014; 2(3.2):73-6.

2. Sarah E Sheppard, Can Ficicioglu. Methylmalonic acidemia. Uptodate; 2023. Contract No.: 22/01/2023.

3. Acquaviva C, Benoist JF, Pereira S, Callebaut I, Koskas T, Porquet D, Elion J. Molecular basis of methylmalonyl-CoA mutase apoenzyme defect in 40 European patients affected by mut° and mut–forms of methylmalonic acidemia: Identification of 29 novel mutations in the MUT gene. Human mutation. 2005; 25(2):167-76.

4. Carrillo-Carrasco N, Sloan J, Valle D, Hamosh A, Venditti C. Hydroxocobalamin dose escalation improves metabolic control in cblC. Journal of inherited metabolic disease. 2009; 32(6):728-31.

5. Zhang C, Wang X, Hao S, Zhang Q, Zheng L, Zhou B, Liu F, Feng X, Chen X, Ma P, Chen C, Cao Z, Ma X. Mutation analysis, treatment and prenatal diagnosis of Chinese cases of methylmalonic acidemia. Scientific reports. 2020; 10(1):1-7.

6. Rajan S, Wallace JI, Brodkin KI, Beresford SA, Allen RH, Stabler SP. Response of elevated methylmalonic acid to three dose levels of oral cobalamin in older adults. Journal of the American Geriatrics Society. 2002; 50(11):1789-95.

7. Tanpaiboon P. Methylmalonic acidemia (MMA). Molecular genetics and metabolism. 2005; 85(1):2-6.

8. Willard HF, Rosenberg LE. Inherited methylmalonyl CoA mutase apoenzyme deficiency in human fibroblasts: evidence for allelic heterogeneity, genetic compounds, and codominant expression. The Journal of Clinical Investigation. 1980; 65(3):690-8.

9. Zschocke J, Hoffman GF. Vademecum Metabolicum: Diagnosis and Treatment of Inborn Errors of Metabolism. 5th ed: Thieme; 2021 January 13, 2021.

10. Baumgartner MR, Hörster F, Dionisi-Vici C, Haliloglu G, Karall D, Chapman KA, Huemer M, Hochuli M, Assoun M, Ballhausen D, Burlina A, Fowler B, Grünert SC, Grünewald S, Honzik T, Merinero B, Pérez-Cerdá C, Scholl-Bürgi S, Skovby F, Wijburg F, MacDonald A, Martinelli D, Oliver Sass J, Valayannopoulos V, Chakrapani A. Proposed guidelines for the diagnosis and management of methylmalonic and propionic acidemia. Orphanet journal of rare diseases. 2014; 9(1):1-36.

11. Fowler B, Leonard J, Baumgartner M. Causes of and diagnostic approach to methylmalonic acidurias. Journal of Inherited Metabolic Disease: Official Journal of the Society for the Study of Inborn Errors of Metabolism. 2008; 31(3):350-60.

12. Yu Y, Shuai R, Liang L, Qiu W, Shen L, Wu S, Wei H, Chen Y, Yang C, Xu P, Chen X, Zou H, Feng J, Niu T, Hu H, Ye J, Zhang H, Lu D, Gong Z, Zhan X, Ji W, Gu X, Han L. Different mutations in the MMUT gene are associated with the effect of vitamin B12 in a cohort of 266 Chinese patients with mut-type methylmalonic acidemia: A retrospective study. Molecular Genetics & Genomic Medicine. 2021; 9(11):e1822.

13. Bartholomew DW, Batshaw ML, Allen RH, Roe CR, Rosenblatt D, Valle DL, Francomano CA. Therapeutic approaches to cobalamin-C methylmalonic acidemia and homocystinuria. The Journal of pediatrics. 1988; 112(1):32-9.

14. Brox-Torrecilla N, Arhip L, Miguélez-González M, Castellano-Gasch S, Contreras-Chicote A, Rodríguez-Ferrero ML, et al. Late-onset methylmalonic acidemia and homocysteinemia. Nutricion hospitalaria. 2021; 38(4):871-5.

15. Obeid R, Fedosov SN, Nexo E. Cobalamin coenzyme forms are not likely to be superior to cyano- and hydroxyl-cobalamin in prevention or treatment of cobalamin deficiency. Molecular nutrition & food research. 2015; 59(7):1364-72.