THE EFFECT OF BREAST-FEEDING OR FORMULA ON THE URINARY METHYL MALONIC ACID LEVELS

Arif Bahar, M.D. / Ferhan Karademir, M.D.
Gökhan Aydemir, M.D. / İsmail Göçmen, M.D.

Department of Pediatrics, Gülhane Military Medical Academy, Haydarpaşa Training Hospital, Istanbul, Turkey.

ABSTRACT

Objective: This study was performed to determine the effect of feeding with breast milk or formula on the serum vitamin B12 and folic acid levels and the urinary methyl malonic acid (UMMA) levels as a marker of cobalamine catabolism

Materials and Methods: The study group consisted of 42 healthy newborn infants. Subjects were divided into two groups. Group 1 and 2 included 22 pre-term and 20 full-term neonates, respectively. The levels of B12, folic acid and UMMA were measured on 20th postnatal day in all cases. In Group 1, twelve infants were fed with breast milk, and the others with formula. Half of the infants in group 2 were fed with breast milk.

Results: There was no statistically significant difference for the levels of vitamin B12 and folic acid between two groups (p=0.771, p=0.371). In group 1, the levels of UMMA were found 30.5±9.2 mmol MMA/mol creatinine in the breast-fed neonates, and 47.0±9.7 mmol MMA/mol creatinine in the formula-fed infants. There was a statistically significant difference between groups (p<0.001). In group 2, the levels of UMMA were found 11.0±4.5 mmol MMA/mol creatinine in the breast-fed neonates, and 19.0±9.5 mmol MMA/mol creatinine in the formula-fed neonates (p=0.001).

Conclusion: Our finding that the levels of UMMA in the formula-fed neonates were higher than the breast-fed infants suggested that breast-feeding has an effect of decreasing the UMMA levels.

Key Words: Breast-feeding, Pre-term, Full-term, Methyl malonic acid

INTRODUCTION

Recent developments in neonatology have caused a substantial decrease in neonatal mortality and morbidity. Preterm neonates or those with a low birthweight have often suffer from neurosensorial impairments, attention disorders and speech disorders, specific neuromotor disturbances, behavioral and socio-emotional abnormalities, and failure in school (1). Vitamin B12 deficiency is uncommon during childhood. An early diagnosis and treatment of this condition is important to prevent irreversible neurological damages. Most of the cases with vitamin B12 deficiency are caused by absorption disturbances during childhood (2).

With the exception of infants who are breast-fed by a vegetarian mother, the breast-fed infants receive about 0,3μg of vitamin B12 daily. Vitamin B12 is stored in the liver. Vitamin B12 is taken by

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Correspondence to: Arif Bahar, M.D. - Department of Pediatrics, Gülhane Military Medical Academy, Haydarpaşa Training Hospital, 81327 Istanbul, Turkey.
e.mail address: arbahar@hotmail.com

diet in the form of hydroxycobalamine, and this molecule is converted to deoxyadenosyl cobalamine and methyl cobalamine and become an active coenzyme. It is metabolized through lysosomes by converting to methyl cobalamine or adenosyl cobalamine. Patients having a cobalamine metabolism disorder may develop homocystinuria and methyl malonic acidemia. The earliest evidences of the deficiency include a serum level of vitamin B12 below 100 pg/ml and the occurrence of hypersegmented neutrophils. Megaloblastic anemia, which is similar to the deficiency of folic acid, leukopenia and thrombocytopenia, occur at the later stages of vitamin B12 deficiency. Neurological symptoms may vary from peripheral neuropathy to sensorial signs, dementia and coma (2-4).

Vitamin B12 which must be essentially taken with diet has been shown to be an important factor in maintaining the neuronal stability in adults, and its deficiency has been reported to involve in the pathogenesis of many neuropsychiatric diseases such as psychosis, depression and dementia (5,6).

The aim of this study was to assess the effect of feeding with breast milk or formula on the vitamin B12 and folic acid levels and the cobalamine metabolism in the full-term and pre-term neonates.

MATERIAL AND METHODS

This study was conducted in the GMMA Haydarpasa Training Hospital. Twenty-two preterm neonates whose gestational ages were between 30 and 37 weeks and birthweights between 1300 and 2400 g (Group 1) and 20 fullterm neonates whose birthweights between 3000 and 4500 g (Group 2) were included in the study. Their parents were informed about the study, and gave a written consent prior to the study.

On the 20th postnatal day, the blood levels of vitamin B 12 and folic acid were measured with an Axym immunoassay device using the method of enzyme immune particle inhibition assay. The urinary methyl malonic acid (UMMA) levels were measured and calculated as the methyl malonic acid (MMA)/ urinary creatinine ratio with a Philips UV/VIS Scanning device using a method of spectrophotometric assay on the spot urine sample (7), a modified form of the Giorgio and Plaut measurement methods (8).

For all cases, we assessed their prenatal data, form of labor, gestational ages, birthweights, status of intrauterine development, vitamins taken by mother, several risk factors such as cyanosis, apneic episodes, intracranial hemorrhage, hypoglycemia, hyperbilirubinemia, exchange transfusion, convulsion, sepsis and the need for ventilator, the treatment methods applied during the neonatal period, and the diseases observed throughout the study. The neonates who had a hereditary metabolic disease and congenital or chromosomal abnormalities were excluded from the study. Gestational ages were determined according to the date of last menstruation and using Ballard scoring method (9).

Students-t and Wilcoxon rank-sum tests were used for the statistical significance of the differences between two groups, with a p value <0.05 as an indicator of a statistically significant difference between the results.

RESULTS

Of the (preterm) neonates in Group 1 (n=22), 13 (59 %) were female, and 9 (41 %) were male, while of the (fullterm) neonates in Group 2 (n=20), 10 (50 %) were female, and 10 (50 %) were male. In Group 1, the mean birthweight and gestational age were found 1780±350 g and 34.5±1.8 weeks, respectively ($p<0.001$). Twelve infants were fed with breast milk, and the others with formula. In Group 2, the mean birthweight and gestational age were found 3000±450 g and 39.5±1.1 weeks, respectively ($p<0.001$). 10 (50%) of the infants in this group, were fed with breast milk, and 10 (50 %) with formula. The levels of vitamin B12 were 270±146 pg/ml and 284±156 pg/ml in Group 1 and 2, respectively. There was no statistically significant difference in the levels of vitamin B12 between two groups ($p=0.771$). The levels of folic acid were found 15±3 ng/ml and 16±4 ng/ml in Group 1 and 2, respectively. There was no statistically significant difference for the levels of folic acid between two groups ($p=0.371$).

The urinary methyl malonic acid (UMMA) levels were found 38.3±12.2 mmol MMA /mol creatinine and 15.0±8.3 mmol MMA/ mol creatinine in Group 1 and 2, respectively. There was a statistically significant difference in the UMMA levels in Group 1 and 2 (p<0.001) (Table I).

Table I: Characteristics of Grup-1 and Group-2

	Group 1 (Pre-term)	Group 2 (Full-term)	P
Number	22	20	
Birthweight (g)± SD	1750±350	3000±450	<0.001
Gestational age (weeks)± SD	34.5±1.8	39.5±1.1	<0.001
B12 level (pg/ml)	270±146	284±156	0.771
Folic acid level (ng/ml)	15.0±3.0	16.0±4.0	0.371
UMMA level (mmol/mol creatinine)	38.3±12.2	15.0±8.3	<0.001

In Group 1, the UMMA levels were found 30.5±9.2mmol MMA/mol creatinine and 47.0±9.7mmol MMA/mol creatinine in the breast-fed neonates and the formula-fed neonates, respectively (p<0.001). In Group 2, the UMMA levels were found 11.0±4.5 mmol MMA/mol creatinine and 19.0±9.5 mmol MMA/mol creatinine in the breast-fed neonates and the formula-fed neonates, respectively (p=0.001). In both groups, there was statistically significant difference in the UMMA levels between the breast-fed neonates and the formula-fed neonates (Table II).

Table II: The UMMA levels in the breast-fed neonates and the formula-fed neonates (mmol/mol creatinine

	UMMA levels		P
	The breast-fed	The formula-fed	
Pre-term	30.5±9.2	47.0±9.7	<0.001
Full-term	11.0±4.5	19.0±9.5	0.001
P	<0.001	<0.001	

DISCUSSION

For the assessment of cobalamine metabolism, the measurement of urine methyl malonic acid or homocystein, which provides data for the metabolic or tissue level, is preferred rather than the measurement of serum levels (10).

An increase in the methyl malonic acid levels has been reported to inhibit a mitochondrial enzyme succinate dehydrogenase, thus blocking the complex II at the respiratory chain (11,12).

In Group 1, the levels of vitamin B12 and folic acid were 270±146 pg/ml and 15±3 ng/ml, respectively; in Group 2 the levels of vitamin B12 and folic acid were 284±156 pg/ml and 16±4 ng/ml, respectively. These values were within the normal range, and there was no statistically significant difference between two groups (p>0.05). However, the finding that the UMMA levels were higher in the preterm infants may be explained by a disorder in the conversion of vitamin B12 into deoxy-adenosyl cobalamine or entering the cell, or an immature activity of these processes in the preterm neonates.

The finding that the UMMA levels were lower in the breast-fed infants than the formula-fed infants suggested that feeding with breast milk had an effect of decreasing the UMMA levels. Any significant increase in the UMMA levels indicates that methyl malonic acid leads to toxic effects on mitochondria, thus preparing a ground for possible neuropsychiatric conditions, which may develop in future (2,13).

The UMMA levels-lowering effect of feeding with breast milk on the UMMA levels may indicate that a factor existing in breast milk plays a role in the conversion of cobalamine into its active form, or taking it into the cell. Studies with large series of cases in which the neurological developments of the cases are followed on a long-term basis should be carried out in order to clarify these issues. However, the data from our study could highlight the economical, psychological and immunological advantages of the breast milk as well as other many metabolic and neurological advantages that would be established in the future.

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