

Research Article

Accuracy Tests of Serum Vitamin D and Calcium Levels in Chorioamnionitis Patients with Preterm Labor

Uji Kesesuaian Kadar Serum Vitamin D dan Kalsium pada Korioamnionitis Pasien Partus Prematur

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Abstract

Objective: To compare the sensitivity and specificity of the diagnosis of chorioamnionitis in preterm labor using serum vitamin D and calcium in Dr. Mohammad Hoesin Hospital Palembang.

Methods: This diagnostic test was carried out in the Obstetrics and Gynecology Department of Dr. Mohammad Hoesin Hospital Palembang from October 2018 to April 2019. Research subjects were women in preterm labor with chorioamnionitis. We collected 39 samples and 36 of them met the inclusion criteria.

Results: Eighteen patients with hypovitaminosis vitamin D had chorioamnionitis with a sensitivity value of 94.74%, specificity 5, 88%, positive predictive value (PPV) 52.9% and Negative Predictive Value (NPV) 50%. Two of the 4 patients with hypocalcemia had chorioamnionitis with sensitivity of 10.53%, specificity 88. 24%, Positive Predictive Value (PPV) 50% and Negative Predictive Value (NPV) 46. 9%.

Conclusions: Reliability of diagnosis accuracy of serum vitamin and calcium levels against chorioamnionitis is poor.

Keywords: accuracy tests, calcium, chorioamnionitis, vitamin D.

Abstrak

Tujuan: Membandingkan sensitivitas dan spesifisitas diagnosis korioamnionitis pada partus prematurus antara serum vitamin D dan kalsium di Rumah Sakit Dr. Mohammad Hoesin Palembang.

Metode: Uji diagnostik dilakukan di Departemen Obstetrik dan Ginekologi RSUP Dr. Mohammad Hoesin Palembang. Penelitian dilaksanakan sejak Oktober 2018 hingga April 2019. Subjek penelitian adalah perempuan hamil prematur yang didiagnosa korioamnionitis. Terdapat 39 sampel dimana 36 sampel termasuk kriteria inklusi.

Hasil: Terdapat 18 pasien dengan hipovitaminosis vitamin D memiliki luaran korioamnionitis memiliki nilai sensitivitas 94,74%, spesifisitas 5, 88%, positive predictive value (PPV) 52, 9% dan Negative Predictive Value (NPV) 50%. 2 dari 4 pasien dengan hipokalsemia memiliki luaran korioamnionitis memiliki sensitivitas 10, 53%, spesifisitas 88, 24%, Positive Predictive Value (PPV) 50% dan Negative Predictive Value (NPV) 46, 9%.

Kesimpulan: Akurasi diagnosis kadar serum vitamin dan kalsium terhadap luaran korioamnionitis memiliki derajat kesesuaian (reliabilitas) kurang baik.

Kata kunci: kalsium, korioamnionitis, uji kesesuaian, vitamin D

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INTRODUCTION

Preterm labour is one of the highest causes of newborn morbidity and mortality. A report from United Nations (UN) entitled *Born Too Soon, The Global Action Report on Preterm Birth* states that globally, 15 million babies are born prematurely every year. More than one million babies die from complications of preterm labour. According to the report, among countries with the highest premature babies, Indonesia is in the fifth position (675,700 babies), after India (3.5 million babies), China (1, 2 million babies), Nigeria (773,600 babies), and Pakistan (748,100) babies.¹

Many factors play a role in causing preterm labour. Intrauterine infection due to chorioamnionitis is suspected to be the most important and the most common cause of preterm.² The incidence of inflammatory chorionic plate in preterm labour was 35.5% versus 5.4% in term delivery.³

Inflammation in chorioamnionitis is a form of immune response. Some antioxidant nutrients play a role in producing and maintaining balance of immune cells.⁴ Vitamin D is known to have anti-inflammatory, anti-bacterial and immune-boosting activities. At 2016 found that women with preterm labor had lower serum vitamin D levels compared to women with sufficient term delivery.⁵ Risk of preterm labor in pregnant women with serum levels of 25 (OH)D ≥ 40 ng/ml is 57% lower compared to pregnant women with serum 25 (OH)D levels ≤ 20 ng/mL.⁶

Vitamin D deficiency is also associated with lack of calcium intake in pregnant women. Low levels of serum calcium in pregnant women are associated with a decrease in the auto production function of vitamin D in the body. Lack of calcium intake can cause secondary deficiency of vitamin D, which affects the amount of maternal serum 25 (OH) D concentrations.⁷

Based on the background above, this study aims to compare the value of conformity between examination of serum 25 (OH) D and calcium levels in woman with preterm labor due to chorioamnionitis in Dr. Mohammad Hoesin Hospital, Palembang.

METHODS

This study was a diagnostic test between serum vitamin D and calcium levels and preterm labour due to chorioamnionitis. This study was conducted at Department of Obstetrics and Gynecology Dr. Mohammad Hoesin Hospital Palembang between October 2018-April 2019. Subjects were women with preterm labour and suspected with chorioamnionitis who was treated at the study site during the study period. Inclusion criteria were preterm delivery with gestational age >24 weeks to <37 weeks suspected with chorioamnionitis (diagnostic criteria: fever (38°C) or 2 of following signs: maternal tachycardia, fetal tachycardia, vaginal discharge, CRP (+), leukocytosis >15.000), with single live fetus, and willing to participate in this study. Patients with obstetric complications, such as antepartum bleeding, preeclampsia, eclampsia, multiple pregnancies, medical complications, such as heart disease and diabetes mellitus and dead fetuses were excluded from this study. Based on sample size calculation using 95% confidence level, we need 35 samples. Samples were collected by purposive sampling. Blood samples were taken from antecubital veins and then analyzed to assess routine blood tests, blood chemistry, qualitative and quantitative CRP, serum vitamin D and calcium levels.

Data was then analyzed using SPSS software version 21.0. Descriptive analysis was performed to describe frequency distribution, sensitivity value, specificity, positive predictive value, and negative predictive value. Cut off point is determined by ROC curve. Conformity, sensitivity, specificity, positive predictive value and negative predictive value was analyzed using Med Calc statistics method.

RESULTS

During study period, 39 samples met our inclusion and exclusion criteria. Three samples were removed due to lysis on the results of laboratory tests.

Table 1 shows the characteristics of the research subjects based on vitamin D levels. Thirty four women (94.4%) suffered from hypovitaminosis vitamin D and only 2 people

(5.6%) had normal vitamin D levels. There were no characteristic differences between patients with hypovitaminosis and normal vitamin D levels ($p > 0.05$).

Table 1. Characteristics of Study Subjects Based on Vitamin D Levels

Variables	Vitamin D		P-Value
	Hypovitamin D (n = 34)	Normal (n = 2)	
Age			
Mean ± SD	29.26 ± 5,38	30.0 ± 2.83	0.972 ^a
Median (Min-Max)	30 (20-35)	30 (28-32)	
Gestational age			
Mean ± SD	31.76 ± 3.39	31.5 ± 2.12	0.529 ^a
Median (Min-Max)	33 (24-35)	31.5 (30-33)	
Parity			
Mean ± SD	2.26 ± 1.26	2.5 ± 0.71	0.641 ^a
Median (Min-Max)	2 (1-5)	2 (2-3)	
Education, n(%)			
Elementary school	1 (2.9)	0 (0)	0.979 ^b
Junior high school	1 (2.9)	0 (0)	
Senior high school	31 (91.2)	2 (100)	
University	1 (2.9)	0 (0)	
Occupation, n(%)			
Housewife	31 (91.2)	2 (100)	0.979 ^b
Teacher	1 (2.9)	0 (0)	
Entrepreneur	1 (2.9)	0 (0)	
Civil servant	1 (2.9)	0 (0)	

^aMann Whitney, $p = 0.05$, ^bPearson Chi Square, $p = 0.05$

Table 2 shows the characteristics of research subjects based on calcium levels. In this study, 4 people (11.1%) had hypocalcemia and 32 (88.9%) had normal calcium levels. There were no characteristic differences between patients with hypocalcemia and patients with normal calcium levels ($p > 0.05$).

Table 2. Characteristics of Study Subjects Based on calcium levels

Variables	Calcium		P-Value
	Hypocalcemia (n = 4)	Normal (n = 32)	
Age			
Mean ± SD	30.25 ± 7.09	29.19 ± 5.11	0.462 ^a
Median (Min-Max)	33 (20-35)	30 (20-35)	
Gestational age			
Mean ± SD	29.25 ± 6.08	32.06 ± 2.82	0.610 ^a
Median (Min-Max)	29 (24-35)	33 (24-36)	
Parity			
Mean ± SD	2.00 ± 1.41	2.31 ± 1.23	0.601 ^a
Median (Min-Max)	1.5 (1-4)	2 (1-5)	
Education, n(%)			
Elementary school	0 (0)	1 (3.1)	0.938 ^b
Junior high school	0 (0)	1 (3.1)	
Senior high school	4 (100)	29 (90.6)	
University	0 (0)	1 (3.1)	

Occupation, n(%)

Housewife	3 (75.0)	30 (93.8)	0.089 ^b
Teacher	0 (0)	1 (3.1)	
Entrepreneur	1 (25.0)	0 (0)	
Civil servant	0 (0)	1 (3.1)	

^aMann Whitney, $p = 0.05$, ^bPearson Chi Square, $p = 0.05$

Vitamin D level in clinical chorioamnionitis patients was 17.83 ± 6.97 with a range of 7.7 - 32.4, whereas in patients without chorioamnionitis it was 17.89 ± 6.76 with a range of 8.6 - 37.3 . There was no difference in vitamin D levels between patients with and without clinical chorioamnionitis ($p = 0.979$). Calcium level in clinical chorioamnionitis patients was 8.89 ± 0.32 with a range of 8-9, whereas in patients without chorioamnionitis calcium level was 8.88 ± 0.33 with a range of 8 - 9. There was no difference in calcium level between patients and without clinical chorioamnionitis ($p = 0.907$).

In this study 18 out of 34 patients (52.9%) with hypovitaminosis vitamin D had clinical chorioamnionitis and 1 in 2 patients (50%) with normal vitamin D levels did not experience chorioamnionitis. Vitamin D levels had a sensitivity value of 94.74%, specificity of 5.88%, positive predictive value (PPV) of 52.9% and negative predictive value (NPV) of 50%. With cut off point 17.5 (figure 1). Diagnosis accuracy of vitamin D levels in predicting clinical chorioamnionitis was 52.8%, it means that the degree of reliability was low.



Figure 1. Cut of point vitamin D

In this study, 2 of 4 patients (50%) with hypocalcemia had clinical chorioamnionitis and 15 of 32 patients (46.9%) with normal calcium levels did not experience chorioamnionitis. Calcium levels have a sensitivity value of 10.53%, specificity of 88.24%, positive predictive value (PPV) of 50% and negative predictive value (NPV) of 46.9%. With cut off point 8.5. Diagnosis accuracy of calcium levels in predicting clinical chorioamnionitis was 47.2%; it means that the degree of reliability was low.

DISCUSSION

The prevalence of preterm labour continues to increase. Major cause of preterm labor is premature rupture of membranes caused by urinary tract or vaginal infection (25.9%). Chorioamnionitis is an inflammation of fetal membrane, it is a manifestation of intrauterine infection (IUI). Chorioamnionitis is not an acute symptom, it is a chronic process and shows no symptoms until labor begins or premature rupture of the membranes occurs. Chorioamnionitis is asymptomatic and symptoms may vary between women. From various studies, it is known that clinical signs and symptoms of chorioamnionitis include: fever (intrapartum temperature >100.4°F or >38°C), significant maternal tachycardia (>120 x/minute), fetal tachycardia (>160x/minute), amniotic fluid or vaginal discharge odour or purulent, tense uterus, maternal leukocytosis (leukocytes >15000 cells/mm³).² Diagnosis of intrauterine infection is based on an increase in inflammatory markers. In this study 19 of 36 patients (52.8%) patients with preterm labor had clinical chorioamnitis.⁸

Vitamin D is a fat-soluble vitamin that is made in the skin when the skin is exposed to sunlight. Vitamin D metabolism during pregnancy and lactation has increased, serum Vitamin D Binding Protein (DBP) levels have increased from 46% to 103% during pregnancy. All pregnant women are encouraged to maintain circulating 25 (OH) D levels of at least 40 ng/mL during the early stages of pregnancy.⁹

Some evidence shows that vitamin D has a potential antimicrobial activity and has a damaging effect. Vitamin D can reduce the risk of infection through several mechanisms. One of them is the role of vitamin D in innate immunity by modulating the production of anti-microbial peptides (AMP) and cytokine responses. Pregnant women with vitamin D deficiency prone to disease and pregnancy problems including gestational diabetes, complications of pregnancy including preeclampsia, increased risk of infection and preterm labor.¹⁰

In this study 34 out of 36 patients (94.4%) patients with preterm labor had hypovitaminosis vitamin D. Statistical analysis found no difference in age, gestational age, parity, education or

employment among patients with and without hypovitaminosis vitamin D. This confirms that hypovitaminosis of vitamin D is not affected by age, gestational age, parity, education or patient work.

Vitamin D is very important for calcium homeostasis, bone mineralization, immune function, cell proliferation, and disease prevention. Vitamin D levels are strongly influenced by calcium, phosphorus, and fibroblast growth factors. The main role of calcium is for contraction and excitation of heart muscle and other muscles, synapses transmission of the nervous system, platelet aggregation, coagulation, and hormone secretion and other regulators that require exocytosis.¹¹

Pregnant women in developing countries generally have low calcium intake. Calcium supplements are quite effective in reducing the incidence of preterm labour in pregnant women with low calcium intake. Pregnant women who regularly consume at least 600 mg of calcium per day and additional calcium supplements 1500 mg/day are at lower risk for preterm labour.¹² A study in Cameroon showed that 94, 6% of pregnant women had inadequate calcium intake. Research in southern Thailand reported that 55% of pregnant women had inadequate calcium intake with an average calcium intake of 493.2 mg/day.^{13,14} However, it was different with the results of our study, we found that only 4 out of 36 (11.1%) pregnant women suffered from hypocalcemia.

Statistical analysis found no differences in age, gestational age, parity, education or employment among patients with and without hypocalcemia. This confirms that hypocalcemia is not influenced by age, gestational age, parity, education or employment.

Systemic inflammation is increasing in women with serum vitamin D deficiency. Vitamin D plays an important role in inflammation and infection of the placenta or chorioamnionitis through prevention of bacterial infections by stimulating cathelicidin in placental cells in both the maternal and fetal sides. In women with vitamin D deficiency, there is an increase in the production of TNF-alpha and inflammatory cytokines. Therefore, women with vitamin D deficiency are

more susceptible to chorioamnionitis because of a decrease in the inflammatory response to clinical and subclinical infections.¹⁵

In this study, 18 out of 34 patients (52.9%) with hypovitaminosis vitamin D had clinical chorioamnionitis and 50% of patients with normal vitamin D levels experienced clinical chorioamnionitis. Using a diagnostic test, we found that vitamin D levels had a sensitivity value of 94.74%, which meant the ability of vitamin D levels (hypovitaminosis vitamin D) to detect clinical chorioamnionitis was 94.74%. We also got a specificity value of 5.88% which means that the ability of vitamin D (normal vitamin D) levels to detect absence of clinical chorioamnionitis was only 5.88%. This means that vitamin D levels can be used to establish a clinical diagnosis of chorioamnionitis but cannot be used to exclude clinical diagnosis of chorioamnionitis.

In addition, in this study 2 out of 4 patients (50%) with hypocalcemia had clinical chorioamnionitis and 17 of 32 (53.1%) patients with normal calcium levels experienced clinical chorioamnionitis. With a diagnostic test, we found that calcium has a sensitivity value of 10.53%, which means the ability of calcium levels (hypocalcemia) to detect clinical chorioamnionitis is only 10.53%. We also get a specificity value of 88.24% which means the ability of calcium (normal calcium) levels to detect the absence of clinical chorioamnionitis is 88.24%. This means that calcium levels can be used to exclude clinical diagnosis of chorioamnionitis but not to establish clinical chorioamnionitis.

CONCLUSIONS

Vitamin D levels have positive predictive value (PPV) of 52.9% and negative predictive value (NPV) of 50%. This means that vitamin D levels can be used to establish a clinical diagnosis of chorioamnionitis but cannot be used to exclude clinical diagnosis of chorioamnionitis.

In this study, Calcium levels have positive predictive value (PPV) of 50% and negative predictive value (NPV) of 46.9%. This means that calcium levels can be used to exclude the clinical diagnosis of chorioamnionitis but not to establish clinical chorioamnionitis.

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