

ARTIKEL PENELITIAN

Gene Polymorphisms Vitamin D Receptor Against Dengue Infection In Children

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Abstrak

Tujuan: Penelitian ini bertujuan untuk mengetahui polimorfisme gen vitamin D terhadap infeksi dengue pada anak. **Metode:** Penelitian ini merupakan penelitian observasional dengan desain cross sectional, dimana variabel dependen dan independen diperiksa secara bersamaan pada pasien anak yang dirawat inap dengan gejala klinis infeksi dengue di RS Raden Mattaher Jambi dan RS Baiturrahim Jambi. yang memenuhi kriteria inklusi dengan IgM anti dengue positif. dan atau IgG anti dengue positif pada anak usia kurang dari dua tahun sampai delapan belas tahun. **Hasil:** Hasil penelitian menunjukkan bahwa hubungan antara pasien yang memiliki polimorfisme pada salah satu gen reseptor vitamin D akan mengalami infeksi dengue yang lebih berat daripada mereka yang tidak memiliki polimorfisme pada gen reseptor vitamin D dan perbedaan ini bermakna secara statistik ($p=0,005$). **Kesimpulan:** Penelitian ini menyimpulkan bahwa ada hubungan antara polimorfisme gen reseptor vitamin D dengan derajat infeksi virus dengue.

Kata kunci: Infeksi dengue pada anak; Polimorfisme gen reseptor vitamin D; tingkat vitamin D

Abstract

Objective: This study aims to determine gene polymorphisms of vitamin D against dengue infection in children. **Methods:** This study was an observational study with a cross-sectional design, where the dependent and independent variables were examined at the same time in pediatric patients who were hospitalized with clinical symptoms of dengue infection at Raden Mattaher Hospital Jambi and Baiturrahim Jambi Hospital who met the inclusion criteria with positive anti-dengue IgM. and or positive anti-dengue IgG in children aged less than two years to eighteen years. **Result:** The results showed that the relationship between patients who have polymorphisms in one of the vitamin D receptor genes will experience heavier dengue infection than those who do not have polymorphisms in the vitamin D receptor gene and this difference is statistically significant ($p=0.005$). **Conclusion:** This study concluded that there was a relationship between vitamin D receptor gene polymorphisms and the degree of dengue virus infection.

Keywords: Dengue infection in children; Vitamin D receptor gene polymorphism; vitamin D level

INTRODUCTION

Dengue virus infection in children is still common and is an infectious disease that causes children to be hospitalized. Dengue infection is a vector-borne disease associated with endemic areas in the tropics and subtropics. This disease has the potential to cause an outbreak with the incidence usually increasing, with clinical manifestations varying from mild to severe and can cause death. The incidence of dengue virus infection has increased by 30 times in the last 50 years, with an estimated 50-100 million cases of dengue fever and 250-500 thousand cases of dengue hemorrhagic fever (DHF) being infected every year with a death rate of 22,000 per year. The World Health Organization (WHO) reports that there has been an increase in cases from 2.2 million in 2010 to more than 3.34 million in 2016. WHO estimates 390 million dengue infections each year, of which 96 million degree of severity.

One of the natural immune responses of children who have dengue infection is through the activation of vitamin D into an active form in the blood which plays a role in reducing viral load competitively against the body's viral receptors and helping macrophages to phagolysosomes of the dengue virus by increasing levels of catelisin through the vitamin D receptors on macrophages and helping inhibit release of excessive pro-inflammatory mediators by the body.¹

Several studies of genetic polymorphisms in the form of non-Human

Leucocyte Antigen (non-HLA) and Human Leucocyte Antigen (HLA) have been identified and have an influence on the susceptibility and severity of disease due to dengue virus infection. Genetic polymorphisms are stable gene variants that have a minor effect on protein regulation or function but play an important role in susceptibility to a disease. One of the genetic polymorphisms that are thought to play a role in the non-HLA immune response is genetic polymorphism at the Vitamin D receptor (RVD) in the immune system.²

Some experts have found a link between low levels of vitamin D in the body and the body's ability to defend against dengue infection in children is low. The active metabolite of vitamin D is 1,25-dihydroxyvitamin D [1,25- (OH) 2D], an immunomodulatory hormone that plays an important role in the immune system. This hormone will increase the regulation of innate (natural) immunity through phagocytosis by monocytes or macrophages through catelisin and decrease the regulation of acquired immunity, by inhibiting lymphocyte proliferation and immunoglobulin production.³

METHODS

Types of research

This study is an observational study with a cross-sectional design, in which the dependent and independent variables are examined at the same time.

Samples

Blood samples were collected from hospitalized pediatric patients with clinical symptoms of dengue infection at Raden Mattaheer Jambi Hospital and Baiturrahim Jambi Hospital with routine laboratory examinations carried out at the Raden Mattaheer Jambi Hospital, Baiturrahim Jambi Hospital. Patients were selected by consecutive sampling, carried out socialization and informed consent about this study to parents. Determination of the sample, patients who experience acute fever (<7 days of acute fever phase) are subjected to peripheral blood tests, IgG and IgM tests. Dengue Ig G and Ig M examination and examination of vitamin D receptor gene polymorphisms, serum vitamin D levels and catelicidin levels were carried out in the Biomedical laboratory of the Faculty of Medicine, Andalas University, Padang.

ELISA examination

The EDTA tube was inserted with a 0.5 cc blood sample, labeled purple, and sent to the Clinical Pathology Laboratory of Raden Matther Jambi Hospital or Baiturrahim Jambi Hospital in accordance with the origin of the sample for routine blood tests (hemoglobin, leukocytes, count, hematocrit, and platelets). Entered a serum sample of 2-2.5 cc, given a yellow label, and sent to the Biomedical Laboratory of the Faculty of Medicine, Andalas University, Padang for

examination of Vitamin D leve (according to kit protocol).

RVD genetic polymorphism examination

DNA extraction using genomic DNA Mini Kit. DNA was extracted from the venous blood of each study subject. The test of success and effectiveness of DNA isolation was controlled using electrophoresis. Electrophoresis was running on 1.5% agarose gel with a voltage of 100 volts for 30 minutes, then the agarose gel was observed with GelDoc.

Ethical Clearence

This research was conducted after passing the ethical selection from the Research Ethics Commission of the Faculty of Medicine, Andalas University and permission from the Director of the Raden Mattaheer Jambi Regional Hospital and the Baiturrahim Jambi Hospital Pertamedika in accordance with the prevailing regulations. Informed consent was requested from the patient's parents in accordance with the research code of ethics established by the Faculty of Medicine, University of Andalas.

Data analysis

The data obtained were analyzed with the SPSS 15 computer system and presented in tables and graphs. The data analysis used was descriptive analysis. Descriptive analysis can describe for data such as vitamin D levels and catelicidin levels and the presence or absence of Vitamin D receptor polymorphisms.

RESULT AND DISCUSSION

Table 1. Vitamin D receptor gene polymorphisms with the degree of dengue virus infection in children

Degree of Dengue Virus infection	n=88	RVD gene polymorphisms	
		Polymorphism (%)	No polymorphism (%)
Mild	33	25 (32.1)	8 (80)
Severe	55	53 (67.9)	2 (20)

Table 1 shows that patients who have polymorphism in one of the vitamin D receptor genes will experience a heavier dengue infection than those who do not

have polymorphisms in the vitamin D receptor gene and this difference is statistically significant ($p > 0.05$).

Table 2. Distribution of Vitamin D receptor gene polymorphisms based on SNP motives and the degree of dengue virus infection in children

SNP	Genotype (Mutant)	Degree of Dengue Virus Infection		
		Dengue fever	DBD	Amount
		N=33 n (%)	N=55 n (%)	
Apa1 → GG	GT (mutant heterozygous)	14	18	32
	TT (mutant)	3	7	10
	GG (wild type)	16	30	46
Taq1 → CC	CT (mutant heterozygous)	4	5	9
	TT (mutant)	20	49	69
	GG (wild type)	9	1	10
Fok1 → TT	TC (mutant heterozygous)	19	19	38
	CC (mutant)	8	21	29
	TT (wild type)	6	15	21
Bsm1 → GG	GA (mutant heterozygous)	4	6	10
	AA (mutant)	0	1	1
	GG (wild type)	29	48	77

In this study, the results showed that the relationship between vitamin D levels and the degree of dengue infection in children was significantly related, where in the mild infection degree group vitamin D levels were still within normal limits or insufficiency. Whereas in the group of degrees of severe dengue infection, it was found that more people had vitamin D deficiency according to Nur Siyam research that adequate intake of vitamin D was recommended to prevent children aged 1-14 years from getting severe dengue infection.⁴ in the study Zaman *et al.*, (2017) concluded that the supplementation of vitamin D was 200,000 IU in adulthood (mean age 33.3 years) with cases receiving supplementation and control only placebo, the results showed that the group that was given supplementation was only 1 patient (1.6%.) who became severe dengue

infection compared to 17 patients (27%) control patients only received placebo.³ Trang *et al.*, (2016) which states that the incidence of DHF is more common in immunocompetent children and children with good nutritional status, but there is no significant difference between nutritional status and the incidence of dengue virus infection.⁵

In this study, the relationship between vitamin D receptor gene polymorphisms and the degree of dengue infection in children showed that if you experience polymorphism, there is a tendency for the child's dengue infection to become heavier. It was found that 78 patients had polymorphisms at the primary point carried out on the vitamin D receptor gene, while 10 patients with dengue infection were children without polymorphism, 8 patients with dengue infection in the mild children group and 2

patients with severe dengue infection in children.

Alagarasu (2012) study found that the vitamin D receptor gene had a relationship with the risk of dengue disease being treated in the dominant rs2228570 'T' allele polymorphism. If there is a mutation in the vitamin D receptor gene, it will affect the function of vitamin D for the body and the susceptibility to dengue infection tends to be severe and the risk of being hospitalized.⁶ In the study, it was found that the most influencing factors, among others, vitamin D levels and vitamin D receptor gene polymorphisms with the degree of childhood dengue infection were the increased levels of catelicidin. This study concluded that there was a relationship between vitamin D receptor gene polymorphisms and the degree of dengue virus infection.

In the blood, vitamin D in the form of 25-hydroxy vitamin D (25 (OH) D), is converted into the active form 1,25 (OH) 2D3 in the kidneys. 1,25 (OH) 2D3 will work in target organs / cells, one of which is immune cells, namely monocytes. The valid biomarker to determine vitamin D status is 25 (OH) D (2,3). Vitamin D acts as a downregulation of Toll Like Receptors

(TLRs) on the surface of monocytes, preventing the entry of viruses into cells, so that viral replication and viral load can be prevented. Therefore vitamin D acts as an antiviral.⁷ This is supported by the research of Guardo, Hernandez, Rosales, Ludert and Angel (2012), regarding the effect of vitamin D (1,25 (OH) 2D3 on human hepatic cells Huh-7, myelomonocyte (U9837) infected with dengue. 25 (OH) 2D3 reduces the number of infected cells and reduces the production of proinflammatory cytokines, thereby reducing disease severity.⁴

CONCLUSION

In this study it was concluded that there was a relationship between vitamin D receptor gene polymorphisms and the degree of dengue virus infection.

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CONFLICT OF INTEREST

Nothing.

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