



## ASSOCIATION OF 25 (OH) VITAMIN D LEVELS WITH INCREASED PROCALCITONIN LEVELS IN SEPTIC PATIENTS AT H. ADAM MALIK GENERAL HOSPITAL MEDAN

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### ABSTRACT

**Introduction:** Sepsis is a complex syndrome caused by an uncontrolled systemic inflammatory response, caused infection. One factor that plays a role in the prognosis of sepsis is vitamin D. While Procalcitonin (PCT), a biomarker response to infection, and PCT levels can be increased in septic patients. The purpose of this study was to determine the association of 25 (OH) Vitamin D levels with increased PCT levels in septic patients.

**Methods:** This study was conducted with cross sectional design, from August to December 2016. The subjects of this study were septic patients who admitted to the intensive care unit at H. Adam Malik General Hospital Medan, aged >18 years. 25 (OH) vitamin D total and Procalcitonin (PCT) were measured by means of MINI VIDAS BRAHMS, correlation of 25 (OH) Vitamin D and PCT levels was tested using Spearman correlation test, statistical tests with p values <0.05 were considered significant.

**Results and Discussions:** A sample of 33 people consisted of 20 (60.6%) men and 13 (39.4%) women, with an average (SD) age of 53.6 (11.7) years. There were 27 (81.8%) patients reporting hypovitaminosis Vitamin D. That was found a significant negative correlation of total 25 (OH) vitamin D levels with procalcitonin levels ( $r = -0.434$ ,  $p = 0.012$ ).

**Conclusions:** In this study concluded that 81.8% of patients who treated in the ICU had hypovitaminosis Vitamin D. There was a negative correlation between 25 (OH) vitamin D levels and increased procalcitonin levels of septic patients who admitted to the ICU.

**KEYWORDS :** Sepsis, Vitamin D, Procalcitonin

### INTRODUCTION

Sepsis is a physiological syndrome, pathology and biochemical abnormalities induced by infection, is a major problem in public health. Although the actual incidence is not clearly known, sepsis is a major suspect to cause mortality and critical illness worldwide, especially in low-income countries where infection is still the most frequent disease causing death. In addition, there is an increasing concern in patients recovering from sepsis who experience long-term physical, psychological and cognitive paralysis. These patients need special care that can impact their social life.<sup>1</sup>

More than 200,000 deaths per year in the United States is caused by sepsis reactions. The incidence of severe sepsis and septic shock has increased in the last 20 years and the annual rate of sepsis cases is currently more than 700,000 (less than 3 per 1000 populations).<sup>2</sup> However, the global burden of sepsis is greatest in the middle and low-income countries (two-thirds of the world's population). Low living standards and poor hygiene along with malnutrition and bacterial infections, parasites and Human Immunodeficiency Virus (HIV) increase the burden of sepsis in these countries. According to World Bank data, Indonesia belongs to the low-middle income country.<sup>3</sup>

Study conducted in a hospital in Bandung, Indonesia, obtained patients suffering from sepsis were 192 patients.<sup>4</sup> There were 233 sepsis cases in 2015 at the Haji Adam Malik Central Hospital, Medan.<sup>5</sup>

However bacterial infection in the last decade is the main concern. Bacteremia infections are associated with high mortality rates. Fast and right treatment can reduce mortality. Several biomarkers of bacterial infections have been investigated, such as Procalcitonin (PCT), the parameters of procalcitonin in the recent years are superior (AUC 0.952) use as biomarkers of bacterial infections.<sup>6</sup>

Procalcitonin (PCT) is a hormone precursor of Calcitonin which is serological marker of acute bacterial infection. In bacterial infections there will be an increase in the expression of the Calcitonin-1 (Calc-1) gene which causes the release of PCT from all parenchymal cells and differentiated cells in the liver and mononuclear cells. The release of PCT inflammatory mediators can be induced through 2 processes, including: The release of toxins in bacteria (endotoxin) and cellular immunity response mediated by pro-inflammatory cytokines such as: IL-1 $\beta$ , IL-6 and TNF- $\alpha$ .<sup>7</sup>

One of the factors contributing in sepsis prognosis is vitamin D, which has recently been known to have other functions beyond its classic activity in bone and calcium homeostasis. The new functions are identified in regulation of hormone secretion, immune function, cellular proliferation and differentiation. The potential role of vitamin D and its active metabolite 1,25-dihydroxyvitamin D (1,25 (OH) 2D) in modulating the immune response was first identified by the discovery of vitamin D receptors in activated inflammatory cells of humans.<sup>8</sup>

Vitamin D dysfunction and insufficiency conditions are common throughout the world. About 1 billion people worldwide had vitamin D insufficiency and/or deficiency, which is defined as 25-hydroxyvitamin D (25 (OH) D) <30 ng/mL for insufficiency or <20 ng/mL for those with deficiency.<sup>10</sup>

Both of these conditions do not have a direct risk to healthy people. However, vitamin D deficiency is associated with mortality in critically ill patients, suggesting that deficiency or vitamin D insufficiency can affect the outcomes in critically ill patients.<sup>9,10</sup>

Septic patients treated in the emergency department (ED) and intensive care unit (ICU) have been shown to have lower levels of 25 (OH) Vitamin D compared to the healthy controls.<sup>11,12</sup>

Vitamin D status of patients are divided into 3 groups: deficiencies (0.0-19.9 ng / mL), insufficiency (20.0-29.9 ng/mL), sufficiency ( $\geq 30$ ng/mL).<sup>12</sup>

Based on these data, PCT and 25 (OH) Vitamin D have a close relationship in patients with critical conditions and those who had sepsis admitted to the intensive care unit (ICU). However, there are still few studies that examine the association between levels of 25 (OH) Vitamin D and increased PCT levels in septic patient, especially at H. Adam Malik Medan General Hospital.

**AIM**

To determine the association of levels 25 (OH) Vitamin D with increased PCT levels in septic patients at RSUP H. Adam Malik Medan.

**METHODS**

This is observational analytic study with a cross sectional study design, subject are 33 sepsis patients who were treated in the ICU ward H. Adam Malik Hospital Medan. sampling was collected consecutive on all accessible populations with inclusion criteria from November 2016 - January 2017. Inclusion criteria: age > 18 years, patients with sepsis who were diagnosed with the criteria of Bone et.al who were treated in the ICU ward of H.Adam Malik General Hospital Medan, willing to follow the research. exclusion criteria: patients with HIV disease, patients with pregnancy, patients with parathyroid disease, patients using drugs that affect 25 (OH) Vitamin D total levels, patients who have received Vitamin D supplements. Vitamin D status of patient was divided into 3 groups: deficiency (0.0-19.9 ng / mL), insufficiency (20.0-29.9 ng / mL), sufficiency ( $\geq 30$ ng / mL).<sup>12</sup>

Ethical clearance was obtained from the Health Research Committee of the Faculty of Medicine, University of North Sumatra. consent paper was obtained in writing from the patient's family. Data and physical examination are written in the status of the study. The sample for this examination are 3 cc patient's blood serum. The sample is taken once for both examinations, that are 25 (OH) Vitamin D total and PCT. Examination of 25 (OH) vitamin D total and PCT was using "MINI VIDAS BRAHMS" tool, with Sandwich principle using the ELFA (Enzyme-Linked Fluorescent Assay) method.

Data analysis was performed using SPSS software (Statistical Package for Social Sciences, Chicago, IL, USA) for Windows. Description of the characteristics research subjects are presented in tabulated form and described. The relationship of clinical characteristics with vitamin status was used Kruskal Wallis test. Correlation of levels 25 (OH) Vitamin D and PCT were tested using Spearman rank test. Statistical tests with p < 0.05 were considered significant.

**RESULTS**

This study was conducted to see the association of total 25 (OH) vitamin D levels and increased levels of procalcitonin in septic patients at H. Adam Malik General Hospital Medan, was conducted from August to December 2016. Samples were collected in accordance with the inclusions criteria in this study. A total of 33 septic patients were performed in ICU and then data analysis was carried out.

**Table 1: Patient Characteristic (n = 33)**

Variable	Value
Age	53.6 ± 11.7
Sex, n (%)	
Male	20 (60.6)
Female	13 (39.4)
Trombocyte, /uL	213363.6 ± 130550.7
Leukocyte, /uL	16627.5 ± 8093.2
PCT, ng/mL	26.2 (4.0, 252.0)
25 (OH) Total Vit. D, ng/mL	21.3 ± 11.6

Note: a: data are presented as mean ± SD, b: data are presented as median (min, max)

In table 1, the patient characteristics are presented, consists of 20 (60.6%) males and 13 (39.4%) females, with an average age (SD) at 53.6 years (11.7 years) at ICU. The mean (SD) value of platelet count is 213363.6/uL, 130550.7 u/L, the mean value (SD) of leukocytes is 16627.5 u/L (8093.2 u/L), and the mean value (SD) of level 25 (OH) Total vitamin D is 21.3 ng/mL (11.6 ng/mL), while procalcitonin levels with a median value of 26.2 ng/mL (4.0 ng/mL - 252.0 ng/mL).

In table 2, among of 33 patients were analyzed there were 18 (54.5%) patients reporting (OH) vitamin D deficiencies, 9 (27.3%) patients reporting insufficiencies, and 6 (18.2%) patients reporting sufficiencies.

**Table 2. Clinical Characteristics of Patients Based on Vitamin D Status**

Variable	Status 25 (OH) Vitamin D			P
	Sufficient ( $\geq 30.0$ ng/mL)	Insufficient (20.0 - 29.9 ng/mL)	Deficient (0.0 - 19.9 ng/mL)	
N (% total)	6 (18.2)	9 (27.3)	18 (54.5)	
Age	50.3 ± 8.1	59.0 ± 10.2	52.0 ± 13.0	.088
Sex, n (%)				.120
Male	5 (83.3)	7 (77.8)	8 (44.4)	
Female	1 (16.7)	2 (22.2)	10 (55.6)	
Trombocyte, /uL	312833.3 ± 165872.7	250444.4 ± 140734.7	161666.6 ± 87287.8	.041*
Leukocyte, /uL	17050.0 ± 10139.0	15282.2 ± 4039.1	17159.4 ± 9168.5	.964
PCT, ng/mL	11.3 (4.9, 29.7)	10.9 (4.0, 106.0)	47.2 (7.0, 252.0)	.004*
25 (OH) Vit.D, ng/mL	41.0 ± 12.0	22.7 ± 2.0	14.1 ± 4.12	<.001*

Note: a: data are presented as mean ± SD, b: data are presented as median (min, max), \* significant value p < 0.05 with the Kruskal Wallis Test.

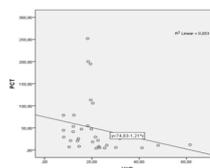
Table 2 analyzes the associations of vitamin D status and the clinical characteristics of patients. Patients with 25 (OH) vitamin D deficiency found a significantly higher procalcitonin level of 47.2 ng/mL (7.0 ng/mL, 252.0 ng/mL) (p = 0.004), and a significantly lower number of platelets 161666.6 uL ± 87287.8 uL (p = 0.041), 25 (OH) vitamin D levels which are 14.1 ng/mL ± 4.12 ng/mL. In contrast, in the 3 groups of vitamin D status there were no differences in age, sex and leukocyte count.

**Table 3. The Correlation Coefficient between Total 25 (OH) Vitamins D and Procalcitonin Values**

Variable	Procalcitonin	
	R	p
25 (OH) Total Vitamin D	- 0.434	0.012*

Note: \* significant correlation p < 0.05, with Spearman correlation test.

In Table 3 and Figure 1, the associations of the levels of 25 (OH) vitamin D and procalcitonin levels, which showed a total 25 (OH) vitamin D level, was significantly negatively correlated with procalcitonin levels (r = -0.434, p = 0.012).



**Figure 1. Correlation between of total 25 (OH) Vitamin D and Procalcitonin Values**

## DISCUSSIONS

In this study we present that 81.8% of sepsis patients treated in the ICU run into 25 (OH) vitamin D deficiency (<20 ng/mL) and 25 (OH) vitamin D insufficiency (20.0 - 30.0 ng/mL). The proportion of patients with hypovitamin D is high in accordance with previous studies in ICU-treated adult patients (prevalence range 17% - 79%). Chen et.al 2015 reported in his study there was 74,1% from 236 adult ICU patients in Guangxi hospitals in China had hypovitamin D.<sup>9,13</sup>

The results in this study show that hypovitamin D is generally still obtained in septic patients admitted to the Intensive Care Unit of H. Adam Malik Hospital in Medan. This can be explained as a result of lack of sun exposure which results in low amounts of vitamin D synthesis in the skin, malnutrition due to lack of dietary intake containing vitamin D and impaired absorption and/or hydroxylation of vitamin D in the liver. Sari DK et.al on Research on healthy women in Indonesia also reports that the factors that most influence vitamin D levels are sun exposure, work, and vitamin D intake.<sup>14,15</sup>

So that monitoring and improving levels of 25 (OH) vitamin D is very important in improving vitamin D status in some patients at risk for sepsis, as in elderly patients and patients who experience long-term care.

Another factor analyzed in this study was platelet counts in septic patients. The range of normal platelet values in adults is 150000 - 400000/uL. Thrombocytopenia is common in critical patients with an incidence of 20% to 50%, this situation is due to the production of various cytokines, endothelial damage, bone marrow suppression, excessive consumption due to the process of DIC (Disseminated Intravascular Coagulation) and organ dysfunction in septic patients.<sup>16,17</sup>

Thrombocytopenia is associated with mortality in patients admitted to the ICU. Claushuis et.al examined the relationship of thrombocytopenia with sepsis and outcome in 931 septic patients, patients with low platelet counts (<100 x 109/L) had more severe disease, this can be indicated by high APACHE and SOFA scores, more often shocked and organ dysfunction.<sup>16</sup>

In this study the mean value of platelet count was lower in the 25 (OH) vitamin D deficiency group than the other 2 groups (insufficiency and efficiency), but not yet experienced thrombocytopenia, as well as the total platelet count total of all patients still in the normal range, even though it was found 10 patients with thrombocytopenia. This can be explained by some patients who have not experienced shock or organ dysfunction.

The mean number of leukocytes in the study showed leukocytosis (>12000/uL) although it did not show a significant difference in the 3 groups of vitamin D status, but this corresponds to one of the clinical criteria of sepsis as follows: Leukocyte count >12,000 cells/ $\mu$ L or lower from 4000 cells/ $\mu$ L or >10% immature leukocytes.<sup>18</sup>

The importance of vitamin D has been recognized since this compound was discovered. In addition to maintaining optimal calcium levels and physical health, other effects of vitamin D have been widely evaluated at this time. The anti-inflammatory and regulatory effects of the hormonal system and the immunomodulatory effects on the kidneys, heart, and immune system of vitamin D have been of considerable concern.<sup>19</sup>

In this study, vitamin D deficiency was significantly negatively correlated with serum PCT levels in patients in our ICU (Figure 4.1) according to Chen et al's study that a significant negative correlation was found ( $r = -0.780, p = <0.001$ ).

Increased serum PCT levels are reported to be associated with an inflammatory response triggered by bacterial infections and have been used as acute phase markers in patients suspected of infection. Kibe et al & Lopez et al reported that PCT levels correlated with disease severity at onset (APACHE Score) and inflammatory process (CRP). Unlike PCT, which is regulated by inflammatory cytokines, vitamin D actually suppresses nuclear activation factor kappa B (Li 2010), thereby reducing proinflammatory cytokines such as IL-1, IL-2, IL-6, IL-8, IL-12, interferon- $\gamma$ , and TNF- $\alpha$ .<sup>19,20</sup>

Preliminary results of epidemiological and clinical studies also support the potential role of vitamin D in maintaining the balance of the immune system. In one study, supplementation of vitamin D in congestive heart failure patients reduced the proinflammatory situation, suggesting that vitamin D supplements could function as a new anti-inflammatory agent for the treatment of future congestive heart failure. In addition, 1,25 (OH) 2D can induce monocyte maturation into mature T cells, which immediately decreases the infectious process which causes an increase in PCT.<sup>21,22</sup>

Therefore, 25 (OH) vitamin D deficiency or insufficiency decreases the ability of the host immune system to suppress bacterial infection and inflammatory response, which causes increased circulating PCT levels.

## CONCLUSIONS AND SUGGESTIONS

In the study it can be concluded that 81.8% of septic patients treated in the ICU had 25 (OH) vitamin D deficiency. There was a negative correlation between 25 (OH) vitamin D levels and an increase in procalcitonin levels of septic patients admitted to the ICU. So that further research needs to be done to see the role of vitamin D supplementation in outcomes of septic patients.

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