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LEVELS OF INTERLEUKIN-6 AND TUMOR NECROSIS FACTOR ALPHA IN PREGNANT PATIENTS WITH PREECLAMPSIA AND PATIENTS WITH NORMAL PREGNANCY

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ABSTRACT

Preeclampsia is a pathological condition often found during pregnancy with a prevalence of about 5-7%. Abnormal implantation, abnormal trophoblast invasion, and endothelial cell dysfunction will result in increased proinflammatory cytokines. One of preeclampsia's pathogenesis is increasing levels of TNF-alpha and IL-6. Because of this, the researchers wanted to know the level of expression of TNF-alpha and Interleukin-6 in pregnant females with preeclampsia. This was an analytical study with a case-control design, done on 30 preeclampsia patients at the Adam Malik Hospital Medan in August - October 2016, who fulfilled the inclusion criteria of the study. IL-6 and TNF-alpha level were then examined, engage ethnic by ELISA with Human Interleukin-6 QY-E04262 reagent and Human Tumor Necrosis Factor-Alfa QY-E00182 reagent. The results of IL-6 in patients with preeclampsia increased (48.60 ± 10.85) as compared to the control (31.99 ± 2.65). TNF-alpha levels in patients with preeclampsia also increased (79.24 ± 10.43) compared to the control (79.24 ± 10.43). Statistical results showed significant differences in the expression of TNF-alpha and IL-6 levels in patients with preeclampsia compared with controls ($p=0.0001$). In this research, there were significant differences in the expression of TNF-alpha and IL-6 levels in preeclampsia patients.

Key word: Preeclampsia, TNF-alfa, IL-6

INTRODUCTION

Preeclampsia is a condition that most often occurs during pregnancy, characterized by hypertension and proteinuria.¹ In developed countries, 16% of maternal deaths occur due to hypertension.² The average prevalence of preeclampsia in Indonesia is between 3-10%.³

A new hypothesis regarding the etiology of preeclampsia has been targeted at the immune response. Cytokines are immunoregulatory substances that may be involved in the pathogenesis of preeclampsia. Type 1 cytokine receptors (interleukin-2), type 2 cytokine receptors (interferon), and type 2 cytokine receptors (tumor necrosis factor alpha-TNF α) are generally produced in an inflammatory process induced by preeclampsia. Several previous types of research even have already shown variations in the levels of cytokines in preeclampsia females although the roles of pathogenesis and expressions in the disease were still controversial.⁴

The sources of TNF- α production in preeclampsia, furthermore, are neutrophils,

monocytes, and possibly placenta. One of the possible mechanisms in preeclampsia was that factors derived from the placenta stimulated monocytes and neutrophils to produce TNF- α resulting in endothelial disruption. Therefore, it seemed that increased serum TNF- α might be part of the pathology of preeclampsia. Meanwhile, interleukin-6, usually identified as a B-cell differentiating factor, is a multifunctional cytokine in various tissues and cells.^{4,5} Interleukin-6 is a pleiotropic cytokine mainly involved in the regulation of inflammation, immune response, and hematopoiesis.^{5,6}

As a result, it can be said that preeclampsia involves a number of maternal, placental, and fetal factors, such as: Adaptation of the placenta with abnormal trophoblasts passing into uterine arteries; The occurrence of maladaptive immunology was in between placenta or paternal, maternal tissue, and fetal tissue; No adaptation between maternal tissue and vascular or inflammatory tissue was found in normal pregnancy; Gene predisposing factors was found in the form of inherited genetic factors along with the presence of epigenetic influences.²

The diagnosis of preeclampsia using the classic diagnostic triad included recent hypertension (increased systolic blood pressure greater than 140 mmHg or diastolic blood pressure greater than 90 mmHg), proteinuria, and edema.² Proteinuria was characterized with the presence of 24-hour proteinuria more than 300 mg, the protein to creatinine ratio of ≥ 0.3 , or the urine protein dipstick of 1+ (30mg/dL persistent) in random urine samples.²

METHODS

This research used a research analysis design in the form of a case-control study. This research was conducted at the Clinical Pathology Department in collaboration with the Department of Obstetrics and Gynecology at the General Hospital of Adam Malik Hospital Medan and its network (Dr. Pirngadi Medan Hospital, Medan Haji Hospital, Putri Hijau Rumkit TK II). Interleukin-6 and TNF- α tests then were carried out at the Clinical Pathology Laboratory of the Adam Malik General Hospital in Medan from August 2016 to October 2016.

The total number of samples in this research was 30 pregnant females with preeclampsia and 30 normal pregnant females as the control group. Those samples had already met inclusion criteria, such as pregnant females diagnosed with preeclampsia, an intrauterine pregnancy, and willing to participate in this research by signing on the research approval sheet. On the other hand, the exclusion criteria in this research were high-risk pregnant patients with DM, kidney disorders, liver disorders, infectious diseases (pneumonia), or acute rheumatic fever. All blood samples then were taken, and IL-6 and TNF- α were examined.

RESULTS AND DISCUSSION

Table 1. Characteristics of interleukin-6 and factor necrosis tumors- α in pregnant patients with preeclampsia and normal pregnant patients

Patients	Interleukin-6 (mean \pm SD)	TNF- α (mean \pm SD)
Preeclampsia	48.60 \pm 10.85	79.24 \pm 10.43
Control	31.99 \pm 2.6	61.36 \pm 5.04

Table 2. The relationship between interleukin-6 and TNF- α values in both preeclampsia and control groups

Variables	N	Preeclampsia (Mean \pm SD)	Control (Mean \pm SD)	P
Interleukin-6	30	48.60 \pm 10.85	48.60 \pm 10.85	0.0001
TNF- α	30	79.24 \pm 10.43	79.24 \pm 10.43	0.0001

Based on Table 1, the mean level of Interleukin-6 in the subjects with preeclampsia was 48.60 ± 10.85 , while the mean level of TNF- α was 79.24 ± 10.43 . On the other hand, it was also known that the mean level of interleukin-6 in the subjects with normal pregnant was 31.99 ± 2.65 , while the mean level of TNF- α was 61.36 ± 5.04 .

Next, the relationship between interleukin-6 and TNF- α in both of the preeclampsia and control groups was examined using an independent t-test. Results of the independent t-test indicated that there were significant differences in the levels of interleukin-6 and TNF- α in both groups with a p-value of 0.001.

Various theories have already analyzed the etiology and pathogenesis of preeclampsia. Endothelial damage, for instance, is considered as an essential factor found in the final stages of preeclampsia process. If endothelial damage does not occur during preeclampsia, an inflammatory response in the form of an increase in proinflammatory cytokines will accumulate macrophages to the perivascular system, known as atherosclerosis. Cytokines as intermediate proteins are secreted by immune cells to regulate the function of other immune cells. In addition to inflammatory cells and immune cells that secrete cytokines, there are also growth factors, chemokines, oncogenes, factors involved in growth differentiation and viability.⁴⁻⁶

Cytokines, moreover, are divided into six groups, namely interleukin, colony-stimulating factor, interferon, tumor necrosis factor, growth factor, and chemokine. Many types of cytokines can increase when preeclampsia occurs which might be used as a marker in preeclampsia. Interleukin-6 and TNF- α are also known as proinflammatory mediators that are released and then trigger damage to endothelial cells. This hypothesis, therefore, suggests that endothelial dysfunction plays a significant role in the pathogenesis of preeclampsia.^{2,7,8}

Preeclampsia is also known to be characterized by systemic endothelial cell dysfunction. Several previous types of research even have already indicated that interleukin-6 was considered as a marker holding the key in the circulation which played a role in endothelial cell dysfunction.

Interleukin-6 could also elevate in the proliferation of trophoblasts, invasion, and oxidative stress during preeclampsia. An excessive maternal inflammatory response then results in an abnormal invasion of the trophoblast.²

Besides, several previous types of research have revealed variations in the levels of cytokines, such as TNF- α and interleukin-6, in preeclamptic females although the role of pathogenesis and expression in disease is still controversial. Hence, this research aimed to examine the relationship between interleukin-6 and TNF- α in the preeclampsia and control groups using an independent t-test. The results of independent t-test found that there was a significant difference between interleukin-6 and TNF- α in both of the preeclampsia and control groups with a p-value of 0.001. Similarly, research conducted by Afshari *et al.* also argued that interleukin-6 levels were significantly higher in pregnant females with preeclampsia than in normal ones. However, this research did not find any significant changes in TNF- α concentrations in preeclamptic females compared to normal pregnant females.⁹

Another previous study conducted by Casart also noticed an increase in interleukin-6 levels in preeclampsia, but it could not be associated with hCG or IL-1 β .¹⁰ Like those previous researches, a research conducted by Roudsari *et al.* study revealed an increase in mean TNF- α level in maternal plasma of pregnant patients with severe preeclampsia compared to those with mild preeclampsia and normal condition.⁴ Nevertheless, the increase in TNF- α concentration did not reach statistical significance. Similarly, study conducted by Raghupathy explained that there was an increase in plasma levels of interleukin-6 and IL-8 in amniotic fluid from pregnant patients with preeclampsia. Thus, increased levels of interleukin-6 could be associated with the onset of preeclampsia.¹¹

CONCLUSION AND SUGGESTION

In conclusion, there was a significant increase in levels of Interleukin-6 and TNF- α in pregnant females with preeclampsia compared to normal ones with a p-value of 0.001.

Further research should be done by combining a

view mediator inflammation parameter, hopefully, interleukin-6 and TNF α can be additional examinations for early diagnosis of preeclampsia.

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