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DIFFERENCES IN AVERAGE SERUM CORTISOL LEVELS AND SERUM 25 HYDROXYVITAMIN D LEVELS IN PREECLAMPSIA AND NORMAL PREGNANCY

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Abstract

According to the World Health Organization, hypertension ranks second as the direct cause of maternal death in pregnancy; namely gestational hypertension (14%). Causes of maternal death in 2021 related to hypertension in pregnancy totaling 1,077 cases. Preeclampsia involves a number of biological processes that affect immune dysfunction, placental implantation, abnormal angiogenesis, excessive inflammation and high oxidative stress reactions and diseases related to maternal vascularization or blood vessels are possible contributing causes. Research with a case-control design was conducted at RSUP Dr. M. Djamil, RSIA Siti Hawa, and RSIA Cicik Padang from February 2021–August 2022. Preeclampsia and normal pregnancy research samples consisted of 27 case samples and 27 control samples. Serum cortisol levels and serum 25-hydroxyvitamin D levels were examined by the ELISA method. The Independent Sample T-Test was used to analyze the data. The mean difference in serum cortisol levels in preeclampsia was 48.70 ± 15.23 µg/dL, higher than that in normal pregnancy, 38.62 ± 8.27 µg/dL (p-value = 0.004, p <0.05). Differences The mean serum level of 25 Hydroxyvitamin D in preeclampsia was 13.63 ± 6.12 ng/ml, lower than that in normal pregnancy, 14.49 ± 3.94 ng/ml (p-value =0.539, p>0.05). The study concluded that there were differences in cortisol levels in preeclamptic pregnancies with normal pregnancies and there were no differences in serum levels of 25 Hydroxyvitamin D in preeclamptic pregnancies with normal pregnancies.

Keywords: Serum cortisol levels; Levels of 25 Hydroxyvitamin D Serum; Preeclampsia and Normal Pregnancy;

INTRODUCTION

Pregnancy is part of the physiological processes of human reproduction, which in the course of which can become pathological. Preeclampsia is one of the pathological conditions in pregnancy that is one of the leading causes of maternal death. Preeclampsia is hypertension in pregnancy characterized by systolic blood pressure of 140 mmHg or more or diastolic blood pressure of 90 mmHg or more on

two examinations with a minimum distance of 4 hours, Preeclampsia after 20 weeks gestation, accompanied by proteinuria ≥ 30 mg / mmol (Chappell et al., 2021).

Preeclampsia is associated with 2% to 8% of pregnancy-related complications worldwide. Preeclampsia causes 9% to 26% of maternal deaths in low-income countries and 16% in high-income countries. Preeclampsia causes more than 50,000 maternal deaths, and more than 500,000 fetal deaths worldwide (Karrar & Hong, 2022)

MMR in West Sumatra Province increased from 2017 to 2018, which was 16 cases to 17 cases. The annual report of the Padang City Health Office found that the causes of maternal death in 2017 were Preeclampsia consisting of 6 cases (37.5%), bleeding 5 cases (31.25%), bronchial asthma 2 cases (12.5%), sepsis 1 case (6.25%), carcinoma recti 1 case (6.25%), and hyperemesis gravidarum 1 case (6.25%). Preeclampsia is the most important cause of maternal and perinatal death in obstetrics (Usman, 2021).

Preeclampsia can cause interference for both the fetus and the mother. Preeclampsia can lead to various severe complications in the mother such as post partum hemorrhage, placental abruption, kidney disorders, edema and pulmonary embolism, Elevated Liver Enzymes Low Platelet Count (HELLP) hemolysis syndrome to death. Preeclampsia and eclampsia conditions will adversely affect fetal health due to decreased placental utero perfusion, hypovolemia, vasospasm, and placental vascular endothelial cell damage so that it can affect the growth and development of the baby (Prawirohardjo, 2014).

In research Salustiano et al., (2013) stated that the involvement of several hormonal systems in the pathogenesis of preeclampsia, one of which is the hormone cortisol. Cortisol is the primary glucocorticoid secreted by the adrenal cortex. In addition to having metabolic, anti-inflammatory and immunosuppressive effects, the hormone cortisol also has a permissive effect on the activity of another hormone, namely epinephrine. Cortisol can increase epinephrine activity so that it can cause vasoconstriction (narrowing of blood vessels) so that this can trigger an increase in blood pressure (Black & Hawks, 2014; Sherwood, 2014)

Cortisol is the primary glucocorticoid secreted by the adrenal cortex. Cortisol plays an important role in the regulation of blood pressure. In addition to having metabolic, anti-inflammatory and immunosuppressive effects, the hormone cortisol also has a permissive effect on the activity of other hormones. Cortisol hormone levels and gestational hypertension have a significant relationship with p value = 0.000 ($p < 0.05$) (Sherwood, 2014; Usman, 2021)

According to research Purswani et al., (2017) stated that preeclampsia is also caused by vitamin D deficiency. Vitamin D deficiency can indirectly result in an increase in blood pressure in the body of pregnant women. Renin Angiotensinogen System (RAS) plays an important role in blood pressure regulation (Purswani et al., 2017).

Various studies show the relationship of vitamin D with hypertension, Vitamin D deficiency conditions are proven to be associated with increased blood pressure through impaired mechanisms in renin transcription, parathyroid hormone imbalance, vascular vasoconstriction, and increased sympathetic nerve activity.

Various epidemiological studies have proven the effect of vitamin D in preventing and treating chronic diseases such as hypertension in pregnancy. Various clinical trial results show that vitamin D has an inconsistent effect on blood pressure in mothers during pregnancy (Farapti & Fadilla, 2021).

Research conducted by Dror et al., (2012) shows supporting evidence that vitamin D levels play a role in the early part of pregnancy in regulating risk factors for complications in pregnancy, supporting fetal growth, bone development and immune maturity. Vitamin D deficiency, as measured by serum levels of 25-hydroxyvitamin D [25(OH)D] is common in pregnant women. Many studies have shown that the risk of preeclampsia increases when serum vitamin D levels are low. A significant association between vitamin D deficiency and preeclampsia has been previously reported (odds ratio, 4.2; confidence interval 95%, 1.4-12.8; p value 0.04).10

Cortisol and vitamin D have been thought to play a role in the development of complications during pregnancy. High cortisol levels mediate the association between stressful conditions during pregnancy, hypertension, and increased risk of preeclampsia. An increase in cortisol during pregnancy is caused by a 2-3-fold increase in cortisol-binding globulin. Vitamin D-binding proteins also increase 2-fold during pregnancy. Both the increase in cortisol-binding globulin and vitamin D-binding proteins are believed to be driven by increased estrogen production during pregnancy. Variations in serum diurnal cortisol have also been observed for 25(OH)D.10

Dr. M. Djamil Padang Central General Hospital (RSUP) is a government general hospital in Padang City and as a referral center for West Sumatra Province and its surroundings. According to data obtained from the Medical Record of Dr. M. Djamil Padang Hospital, in 2018 preeclampsia patients treated in obstetric, gynecological and outpatient inpatient installations were 169 people, in 2019 there were 92 people and again increased in 2020 by 174 people. The high number of preeclampsia cases handled at Dr. M. Djamil Padang Hospital and cases that have been categorized as preeclampsia. For this reason, it is important during pregnancy to pay attention to blood pressure during pregnancy, as well as vitamin D and cortisol so as to prevent preeclampsia in pregnancy, based on the background above, researchers are interested in discussing the "Differences in Average Serum Cortisol Levels and Serum 25 Hydroxyvitamin D Levels in Preeclampsia Pregnancy and Normal Pregnancy at Dr. M. Djamil Hospital, Siti Hawa Hospital and RSIA Cicik Padang.

RESEARCH METHODS

This study used an observational analytical research design with the research design used was a retrospective case control, namely by identifying a group of cases (patients suffering from the effects or disease being studied) compared to a control group (those who did not suffer from the disease or effects).

This research was carried out at the Midwifery Department of Dr. M. Djamil Padang Hospital, Siti Hawa Padang Hospital and Cicik Padang Hospital with sample

collection starting from February 2021 to August 2022. Examination of serum cortisol levels and serum 25-hydroxyvitamin D levels was carried out at the Biomedical Laboratory of the Faculty of Medicine, Andalas University using the Enzyme Linked Immunosorbent Assay (ELISA) method.

The population in this study was all pregnant women who experienced Preeclampsia and normal pregnant women at RSUP M.Djamil, RSIA Siti Hawa and RSIA Cicik Padang. The sample is part of the population that meets the inclusion and exclusion criteria from research at RSUP Dr. M. Djamil, RSIA Siti Hawa and RSIA Cicik Padang. The inclusion criteria in this study were willing to participate in the study by signing informed consent, patients with a diagnosis of Preeclampsia based on clinical symptoms and laboratory examination results and mothers with normal pregnancies, and maternal age 20-35 years. The exclusion criteria in this study were mothers with a history of chronic diseases based on history / in treatment such as heart, diabetes, kidney and mothers with a history of chronic hypertension. Sampling in this study was carried out using consecutive sampling techniques until it met the required number of samples, namely 27 pregnant women with a diagnosis of Preeclampsia (cases) and 27 normal pregnant women (controls) at Dr. M. Djamil Hospital, Siti Hawa Hospital and Cicik Padang Hospital. The independent variables in this study were Serum Cortisol levels and 25 Serum Hydroxyvitamin (25(OH)D) Levels. The dependent variables in this study were Preeclampsia pregnant women and normal pregnant women. All preparations and examinations of serum cortisol levels and serum 25 Hydroxyvitamin D are carried out at the Biomedical Laboratory of Andalas University. The examination was assisted by officers of the Biomedical Laboratory of Andalas University.

The entire population of Preeclampsia mothers and normal pregnant women at RSUP Dr M. Djamil, RSIA Cicik and RSIA Siti Hawa were determined by inclusion criteria according to the number of samples determined both case samples and normal samples. Case samples were obtained through the Emergency Department (IGD) of RSUP M. Djamil, RSIA Cicik and RSIA Siti Hawa, while normal samples were obtained at the polyclinics of RSUP M. Djamil, RSIA Cicik and RSIA Siti Hawa.

The blood collection process is carried out 1 time for Preeclampsia pregnant women who come to the Emergency Department (IGD) of RSUP M. Djamil, RSIA Cicik and RSIA Siti Hawa who have not received MgSO4 treatment, while normal pregnant women also take blood 1 time at the polyclinic. Researchers check blood pressure first, before taking blood. The researcher explained informed consent to respondents, respondents signed informed consent that had been read and approved then the researcher conducted interviews and filled out questionnaires related to respondent characteristic data. Blood sampling is taken as much as 3 cc to check serum cortisol levels and serum levels of 25 (OH) D. Blood taken is stored in EDTA tubes (yellow color) and / or heparin. Then, the serum sample was brought and sent to the Biomedical Laboratory of FK Unand immediately. Serum samples of subjects that have been put into a microtube are put into a cool box which has been prepared ice cold pack to maintain the temperature in the cool box is still safe in the range of 2-6°C on the way to the Biomedical Laboratory of FK Unand. Blood that has been taken is then centrifuge to obtain the subject's blood serum so that it has a longer

resistance and is not damaged quickly. Using the 25-hydroxyvitamin D ELISA kit (25(OH)D) from Biochem Diagnosis Canada (DBC). After the sample entered the Biomedical Laboratory of FK Unand, the subject's serum sample was stored at a temperature of less than -100 C to be stored for a longer time (± 6 months). Each micro tube of serum samples is given a code according to the respondent code. After laboratory analysis tests, data analysis was carried out using statistical tests Independent sample T test if normally distributed and Mann Whitney test if not normally distributed to obtain the final results of the study.

RESULT AND DISCUSSION

This research has been carried out from February 2021 to August 2022 with research sites at Dr. M Djamil Padang Hospital, RSIA Cicik Padang and Siti Hawa Padang Hospital and the Biomedical Laboratory of the Faculty of Medicine, Andalas University. The sample studied amounted to 54 respondents consisting of 27 respondents with a diagnosis of preeclampsia and 27 respondents with a diagnosis of normal pregnancy.

This study used Kolmogorov Smirnov's normality test (data ≥ 50 samples). At serum cortisol levels and serum 25 hydroxyvitamin D levels, the results obtained p = 0.200 (p> 0.05) so that data on serum cortisol levels and serum 25 hydroxyvitamin D levels can be concluded normally, the data can be continued with bivariate analysis using the Independent Sample T test.

The most characteristic results of respondents with high school education level were 17 people (63.0%) in the preeclampsia pregnancy group and 15 people (55.6%) in the normal pregnancy group. In employment status, 21 (77.8%) mothers did not work in the preeclampsia pregnancy group and 20 (74.1%) mothers did not work in the normal pregnancy group. A total of 8 people (29.6%) mothers with second and third pregnancies as many as 9 people (33.3%) in the preeclampsia pregnancy group, and 10 people (37.0%) mothers with third pregnancies in the normal pregnancy group.

In this study, it was known that pregnant women in the preeclampsia group had the most BMI of obesity (≥27.5 kg / m2) as many as 13 people (48.1%), as well as normal pregnancy groups had the most BMI of obesity (≥27.5 kg / m2) as many as 11 people (40.7%). Most family members in the preeclampsia pregnant women group smoked as many as 24 people (88.9%), as well as the normal pregnancy group also known to many family members smoke, as many as 21 people (77.8%).

In this study it was also known that in the pre-eclampsia pregnancy group did the most activities outside the home less than one hour as many as 17 people (63%), as well as in the normal pregnancy group did the most activities outside the home less than one hour, which was as many as 11 people (40.7%). The average age of pregnant women in the preeclampsia pregnancy group was (33.30 ± 5.84) years and in the normal pregnancy group was (31.11 ± 4.76) years. Average blood pressure in the preeclampsia pregnancy group was (162.78 ± 12.62) mmHg and in the normal pregnancy group was (117.85 ± 11.04) mmHg.

Based on the results of the study, the average serum cortisol levels in preeclampsia and normal pregnancies can be seen in table 2. Based on Table 2. The average results of serum cortisol levels in preeclampsia pregnancy were (48.70 ± 15.23) µg / dL. Average normal pregnancy serum cortisol levels were (38.62 ± 8.27) µg/dL

Table 1. Average Serum Cortisol Levels Preeclampsia Pregnancy and Normal Pregnancy

Average Serum Levels	Preeclampsia Pregnancy Group (Mean ± SD)	Normal Pregnancy Group (Mean ± SD)
Kortisol (µg/dL)	48,70 ± 15,23	38,62 ± 8,27

Based on the results of the study, the average level of 25 serum Hidroxyvitamin D in preeclampsia and normal pregnancy can be seen in table 3. Based on Table 3. The average results of 25 serum Hidroxyvitamin D in preeclampsia pregnancy were (13.63 ± 6.12) ng / ml. Average levels of 25 normal pregnancy serum Hidroxyvitamin D were (14.49 ± 3.94) ng/ml.

Table 2. Average Serum Hydroxyvitamin D Levels in Preeclampsia and Normal Pregnancy

Average Serum Levels	Preeclampsia Pregnancy Group (Mean ± SD)	Kelompok Kehamilan Normal (Mean ± SD)
25 Hydroxyvitamin D (ng/ml)	13,63±6,12	14,49 ± 3,94

Based on the results of the study, differences in serum cortisol levels in preeclampsia and normal pregnancy can be seen in table 4. Based on Table 5.4, the results of differences in serum cortisol levels of preeclampsia pregnancy were (48.70 ± 15.23) µg / dL and normal pregnancy serum cortisol namely (38.62 ± 8.27) µg / dL with a difference in values of 10.08 µg / dL. Based on the results of statistical tests using the Independent T-test, the result of p-value = 0.004 (p< 0.05) means that there is a difference in serum cortisol levels between preeclampsia pregnancy and normal pregnancy.

Table 3. Differences in Serum Cortisol Levels in Preeclampsia Pregnancy and Normal Pregnancy

Up to Serum	Preeclampsia Pregnancy Group (Mean ± SD)	Normal Pregnancy Group (Mean ± SD)	Difference (Δ)	p-*value*
Kortisol (µg/dL)	48,70 ± 15,23	38,62 ± 8,27	10,08	0,004

Based on the results of the study, the difference in serum levels of 25 Hidroxyvitamin D in preeclampsia and normal pregnancy can be seen in table 5. Based on Table 5., the results of differences in serum levels of 25 Hidroxyvitamin D in preeclampsia pregnancy were (13.63 ± 6.12) ng / ml and 25 Hidroxyvitamin D serum normal pregnancy namely (14.49 ± 3.94) ng / ml with a difference in values

of 0.86 ng / ml. Based on the results of statistical tests using the Independent T-test, the result of p-value = 0.539 (p> 0.05) means that there is no difference in serum levels of 25 Hidroxyvitamin D between preeclampsia pregnancy and normal pregnancy.

Table 4. Differences in Serum 25 Hydroxyvitamin D Levels in Preeclampsia and Normal Pregnancy

Up to Serum	Preeclampsia Pregnancy Group (Mean ± SD)	Normal Pregnancy Group (Mean ± SD)	Difference (Δ)	p-*value*
25 *Hidroxyvitamin* D (ng/ml)	13,63 ± 6,12	14,49 ± 3,94	0,86	0,539

The results showed that the characteristics of respondents based on education were the most with high school education level, namely 17 people (63.0%) in the preeclampsia pregnancy group and 15 people (55.6%) in the normal pregnancy group. According to research by Fairbrother et al., (2017) the education factor of pregnant women affects the occurrence of preeclampsia.

The level of education also determines whether or not a person absorbs and understands pregnancy knowledge and the risks of complicating it that they get. This can be used as a basis for distinguishing methods in proper absorption. Education is needed so that a person is responsive to nutritional problems in the family and can take action as soon as possible (Suhardjo et al., 1985).

In employment status, 21 people (77.8%) were non-working mothers / IRT (Housewives) in the preeclampsia pregnancy group and 20 people (74.1%) were not working mothers / IRT in the normal pregnancy group. Work is related to the physical activity load of mothers who work as housewives tend to do activities and spend time at home.

Activities that are often done by housewives such as starting from cleaning the house, preparing school children, ironing clothes, cooking, washing clothes and various other activities that often trigger stress due to monotonous or repetitive activities every day. This can be seen from the statistical results that serum cortisol levels are higher in housewives with preeclampsia pregnancy with average serum cortisol levels of 49.7 (µg / dL) while serum cortisol levels in housewives with normal pregnancy groups are 37.66 (µg / dL).

Mothers who work as housewives are busy with work inside the house causing mothers to rarely move outside the home so that little sun exposure. This can be seen from the statistical results in this study it is known that most mothers with work as housewives have an average level of 25 serum hydroxyvitamin D (25 (OH)D) lower than working mothers, where the average level of 25 serum hydroxyvitamin D in housewives of normal pregnancy group is 12.79 ng / ml, while the average level of 25 serum hydroxyvitamin D in housewives with preeclampsia pregnancy group is 10.93 ng / ml.

Research has been conducted by Wulandari, R and Firnawati, A (2012), which states that there is a significant relationship between maternal work and the incidence of preeclampsia with p = 0.001; OR = 4.173 (CI = 1.709-10.188). Research by Indriani et al., (2012), also proved that employment status has a significant relationship with preeclampsia. In addition, there is also a significant relationship between work and stress levels in pregnant women.3

In this study, it is known that pregnant women in the preeclampsia group have the most obesity BMI (≥27.5 kg / m2) as many as 12 people, while in the group of normal pregnant women mothers with obesity BMI (≥27.5 kg / m2) only as many as 4 people. Based on the statistical results of the research conducted, it is known that mothers with obesity can increase the average serum cortisol levels where the average serum cortisol levels in pregnant women in the normal group are 50.5 µg / dl, while serum cortisol levels in the preeclampsia pregnancy group are 56.8 µg / dl. Conversely, pregnant women with obesity can reduce serum levels of 25 hydroxyvitamin D, where the average vitamin D levels in the normal pregnancy group are 10.16 ng / ml, while the average vitamin D levels of the preeclampsia pregnancy group are 9.47 ng / ml.

Women who were overweight and obese were 1.4 and 1.8 times more likely to have preeclampsia than women with normal BMI, while women who were underweight were likely to have preeclampsia 0.7 times. There was a positive association between increased body mass index before pregnancy and the risk of developing preeclampsia, with an odd ratio of 1.8 for obese women with a BMI above 30 compared to women of normal weight, a BMI between 20 and 24.9 (Mrema et al., 2018).

In this study also found most family members in the group of preeclampsia pregnant women smoked as many as 24 people, as well as the normal pregnancy group also known to many family members smoke, as many as 21 people. In research states that smoking during pregnancy causes oxidative stress which has an impact on the occurrence of vitamin D deficiency and secondhand smoke during pregnancy has a synergistic impact on gestational hypertension. Vitamin D deficiency and smoking are important interrelated factors for hypertension in pregnancy or preeclampsia. Mothers exposed to secondhand smoke had significantly higher systolic, diastolic and proteinuria blood levels (Yıldız et al., 2022).

In this study, both in the preeclampsia pregnancy group and the normal pregnancy group, pregnant women mostly experienced exposure to high cigarette smoke. In this study, from statistical results it was proven that exposure to cigarette smoke can reduce serum hydroxyvitamin D levels where, the average serum hydroxyvitamin D levels in mothers exposed to normal pregnancy cigarette smoke were 12.81 ng / ml, while the average vitamin D levels in mothers who were not exposed to cigarette smoke in normal pregnancy was 20.37 ng / ml. Similarly, the average serum level of 25 hydroxyvitamin D in the preeclampsia group exposed to cigarette smoke was 12.11 ng / ml, while the level of 25 serum hydroxyvitamin D in preeclampsia mothers who were not exposed to cigarette smoke was 25.72 ng / ml.

In this study, it was also known that in the preeclampsia pregnancy group did the most activities outside the home for less than one hour as many as 17 people (63%), as well as in the normal pregnancy group the most did activities outside the home for less than one hour, which was as many as 11 people (40.7%). According to research, high daily exposure to solar radiation during pregnancy is associated with a reduced risk of hypertension in pregnancy (Preeclampsia).

Clinical studies have shown that sunlight, especially UVB, has a direct effect on vascular health and reduces blood pressure through the release of nitric oxide from the skin. UVB exposure for 20 minutes led to a reduction of 3.5 mmHg in mean systolic artery pressure and 4.9 mmHg in diastolic blood pressure in healthy adults. Best exposed to sunlight at 10.00 to 13.00 wib. Lack of sun exposure is associated with reduced serum levels of 25 hydroxyvitamin D (Botyar & Khoramroudi, 2018).

In line with the results of research that has been conducted where statistical results obtained the levels of 25 serum hydroxyvitamin D were lower in respondents with activities outside the home less than one hour in the normal pregnancy group at 11.9 ng / ml while serum 25 hydroxyvitamin D levels in the normal pregnancy group with activities outside the home for more than one hour at 18.14 ng / ml. The average serum hydroxyvitamin D level in the group with preeclampsia pregnancy with less than one hour activity was 9.95 ng / ml, while the average serum 25 hydroxyvitamin D levels in preeclampsia pregnancy with activity more than one hour was 19.87 ng / ml.

A significant correlation was found between maternal serum 25 hydroxyvitamin D levels and exposed body surface area ($r = 0.36$, $p < 0.002$) or percentage of body surface exposed ($r = 0.39$, $p < 0.001$) and radiation intensity ($r = 0.15$, $p = 0.029$).3 The body surface area that must be minimally exposed to the sun is sun exposure on the face, arms, and hands (15-20% of the body surface area). There are significant differences in vitamin D synthesis between full-body exposure versus face-to-foot sun exposure (Judistiani et al., 2019).

Pregnant women without hijab are advised to have continuous exposure for approximately 37.5 minutes per day, while for hijabi women the recommended duration is about 64.5 minutes per day.15 Skin exposure to sunlight for 7-25 minutes from 10.00 am to 3 pm, at least twice a week, where the face, arms, and legs are exposed without sunscreen application should be sufficient to induce vitamin D synthesis (Ilmiawati et al., 2020). The characteristics of the fabric used, such as color, thickness, and weaving mode, can affect its effectiveness in blocking UVB absorption by the skin. Dark cloth is twice as effective at absorbing UVB radiation as white cloth.16

The average age of pregnant women in this study in the preeclampsia pregnancy group was (33.30 ± 5.84) years and in the normal pregnancy group was (31.11 ± 4.76) years. Average blood pressure in the preeclampsia pregnancy group was (162.78 ± 12.62) mmHg and in the normal pregnancy group was (117.85 ± 11.04) mmHg. Age is one of the dominant risk factors associated with the incidence of preeclampsia.16

Pregnant women aged 35 years or older have a four times greater chance of developing preeclampsia than those aged 25-29 years. Likewise, those women aged

30-34 were about three times more likely to develop preeclampsia than those aged 25-29. Mothers aged >35 years experience changes in their bodies such as impaired aging of organs, decreased kidney function, decreased liver function, increased blood pressure and diabetes mellitus, so the likelihood of finding diseases during pregnancy such as preeclampsia will increase (Yıldız et al., 2022).

In this study, most respondents found multigravida pregnant women with the same number of gravids more than two, both in the normal pregnancy group of 24 respondents (88.8%) and in multigravida mothers in the preeclampsia pregnancy group of 23 respondents (85.18%). Based on the results of research conducted by Brunton, et al (2020) stated that parity was identified as a predictor of anxiety and psychological disorders.17

According to research by Madhavanprabhakaran et al., (2015) states that primiparous mothers with younger ages have higher levels of pregnancy-related anxiety compared to older multiparous mothers. Stress during pregnancy is common in mothers who give birth for the first time (primipara) due to lack of adaptation to physical (biological) and psychological changes during pregnancy.18

In contrast to research conducted by Polo-Kantola et al.,(2017) states that older age during pregnancy causes depression and higher psychological disorders. Psychological disorders lead to an increase in cortisol levels. Stress in multiparous mothers can arise due to experiences through the previous process of pregnancy and childbirth. In addition, stress levels can occur because multigravida mothers have an older age during pregnancy is significantly associated with worsening sleep quality and insomnia during pregnancy. Polo-Kantola et al., (2017) also reported that multiparous mothers had poorer sleep quality than primiparous mothers. Sleep disorders and psychological disorders cause elevated cortisol levels (Bublitz et al., 2018; Polo-Kantola et al., 2017)In this study, it was found that multigravida mothers had a higher average cortisol than mothers with primigravida where the average cortisol levels in primigravida mothers in the normal pregnancy group were 33.08 µg / dL while the average cortisol levels in multigravida were 39.31 µg / dL. While in the preeclampsia pregnancy group with primigravida had an average cortisol level of 48.01 µg / dL, then in the preeclampsia pregnancy group with multigravida the average cortisol level was 48.82 µg / dL.

Based on Table 2. The average results of serum cortisol levels in preeclampsia pregnancy were (48.70 ± 15.23) µg / dL. The average serum cortisol level of normal pregnancy is (38.62 ± 8.27) µg / dL.Based on the results of statistical tests using the Independent T-test, the result of p-value = 0.004 ($p < 0.05$) which means there is a difference in serum cortisol levels between preeclampsia pregnancy and normal pregnancy. In this study, it was seen that the difference in average serum cortisol levels in the preeclampsia pregnancy group and the normal pregnancy group looked much different, namely 10.08 µg / dL. From these results, it can be seen that there are differences in cortisol levels, where serum cortisol levels are higher in the preeclampsia group than in the normal pregnancy group.

This result is in line with the results of research by Bärebring et al., (2019) which reported that there is a significant positive correlation or relationship between cortisol

and the incidence of preeclampsia. Where the higher cortisol levels, the higher the risk of preeclampsia. In this study, the average cortisol level in the group of hypertensive pregnant women or preeclampsia was 111.50 μg / dL. An increase in serum cortisol of 16.9 μg / dL was associated with an increase in systolic and diastolic 0.7 and 0.4 mmHg.21

Research by Liu et al., (2020) also states that cortisol levels in the preeclampsia pregnancy group are higher than in the normal pregnancy group.22 Prenatal glucocorticoid exposure is associated with higher increases in systolic and diastolic blood pressure. High cortisol levels can be a risk factor for cardiovascular disease, with elevated plasma glucose and triglyceride levels and elevated systolic blood pressure (Eberle et al., 2021; Wang et al., 2013)

High exposure to glucocorticoids/cortisol during prenatal activity can cause regulation of Lipoxin A (LXA 4) levels to decrease in preeclampsia patients. Cortisol levels are negatively correlated with LXA4 levels. Where the higher cortisol levels, LXA 4 levels will also decrease, this can suppress the expression of 11β-HSD2 which is a glucocorticoid inhibitor enzyme in the placenta. Lipoxin A4 (LXA4) is one of the most important endogenous anti-inflammatory, derived from arachidonic acid (AA) which involves enzymatically double lipoxygenase of arachidonic acid by lipoxygenase (LOXs), serving as a "stop signal" in inflammation. LXA 4 provides an important role in the recovery of hemostasis and the cessation of inflammation (Liu et al., 2020).

Preeclampsia can occur due to systemic inflammatory conditions in which oxidative stress and endothelial dysfunction and spiral artery remodeling can inhibit fetal growth, but it can also be caused by a deficiency of polyunsaturated fatty acids (PUFAs) and mediator anti-inflammatory products including lipoxins, resolvin, protectin, and maresin (Liu et al., 2020).

This is in line with research conducted by Leff-Gelman et al., (2020) which states that high cortisol is associated with psychological disorders such as anxiety disorders or chronic stress. Psychological disorders such as major anxiety disorder have a positive correlation during pregnancy. Where the higher the level of anxiety and stress levels, the serum cortisol levels will also increase (Zhang et al., 2018).

A lot of evidence suggests that preeclampsia is also associated with glucocorticoid/cortisol exposure caused by stress and anxiety disorders. Preeclampsia can increase stress and trigger elevated cortisol levels.26

This is in line with the research that has been done, where this study limits normal cortisol levels based on the standard cortisol kit used. The normal limits of cortisol are related to the division of AM and PM time. If sampling in the morning (AM) time range, the normal limit value range of cortisol is 3.95-27.23 μg/dL with an average value of 15.59 μg/dL. Meanwhile, if sampling in the day-night (PM) range, the normal limit range of cortisol is 1.45-10.41 μg / dL with an average value of 5.93 μg / dL

The results of the study that the researchers have conducted obtained samples in the normal pregnancy group in the morning range (AM) as many as 11 respondents with an average cortisol level of 41.77 μg / dL, while sampling normal

pregnant women during the day to night (PM) as many as 16 respondents with an average cortisol level of 36.46 μg / dL. Then in the preeclampsia pregnancy group in the morning range (AM) as many as 16 respondents with an average cortisol level of 54.83 μg / dL, while sampling of normal pregnant women during the day to night (PM) as many as 11 respondents with an average cortisol level of 39.78 μg / dL

This shows that pregnant women in the normal pregnancy group and the preeclampsia pregnancy group have cortisol levels higher than normal limits which is a marker that pregnant women experience stress levels, but the average cortisol levels are higher in the preeclampsia pregnancy group than the normal pregnancy group which is a marker that has a higher stress level preeclampsia group.

Fairbrother et al., (2017) research reported that women with risky pregnancies such as preeclampsia have 5.2 times greater anxiety and stress than women with normal pregnancies.27 This is in line with research that researchers have conducted, mothers with risky pregnancies such as preeclampsia are more easily stressed or anxious than normal pregnancies because the obstetric problems they experience require a level of care and doctor visits and medical treatment More often, so this can cause financial constraints and treatment time which more often triggers psychological problems.

Therefore, it needs good emotional and instrumental support from family and health workers for pregnant women so that psychological problems do not occur both in early pregnancy and before delivery which can increase the production of cortisol levels in pregnant women. Researchers argue that in this study, the preeclampsia group had cortisol levels higher than the group with normal pregnancy because it was triggered by stress that occurred in early pregnancy and then the stress persisted due to poor obstetric conditions, such as obesity problems, as well as exposure to cigarette smoke which can increase the occurrence of oxidative stress, this stress is what causes an increase in cortisol levels and affects vasoconstriction, vascularization disorders that have an effect on increasing blood pressure in mothers

Based on Table 5. The results of differences in serum levels of 25 Hydroxyvitamin D in preeclampsia pregnancy were (13.63 ± 6.12) ng / ml and 25 Hydroxyvitamin D normal pregnancy serum namely (14.49 ± 3.94) ng / ml with a difference in values of 0.86 ng / ml. Based on the results of statistical tests using the Independent T-test, p-value = 0.539 ($p > 0.05$) which means there is no difference in serum 25 Hydroxyvitamin D levels in preeclampsia pregnancy with normal pregnancy.

According to guidelines from the Institute of Medicine (IOM) in (Raia-Barjat et al., 2021) recommend vitamin D (25(OH)D) concentrations for women in pregnancy above 20 ng/mL, then it is said to be deficient if serum 25 hydroxyvitamin D (25(OH)D) levels are below 20 ng/mL (50 nmol/L). Based on the results of research that researchers have conducted, 25 serum hydroxyvitamin D levels of preeclampsia pregnancy were obtained (13.63 ± 6.12) ng / ml and 25 hydroxyvitamin D levels of normal pregnancy serum namely (14.49 ± 3.94) ng / ml. This showed that the average vitamin D levels in both groups of pregnant women

were < 20 ng / mL. This showed that subjects in this study in both normal pregnancy and preeclampsia groups experienced vitamin D deficiency (Wang et al., 2013).

Vitamin D deficiency during pregnancy is associated with many short- and long-term problems for both the fetus and the mother. Vitamin D during pregnancy plays three main roles. First, stimulation of calcium absorption, a process necessary for the accrual of fetal bone minerals during the last trimester of pregnancy. Secondly, vitamin D contributes to fetal tolerance, being an allograft during pregnancy. A third important role is its involvement in various transcriptional regulations (Raia-Barjat et al., 2021).

Vitamin D deficiency in mothers during pregnancy is a common and public health problem at the global level. Vitamin D deficiency (25-hydroxyvitamin D serum (25(OH)D) < 20 ng/mL) is a major widespread public health problem among the general population and is particularly prevalent in pregnant women, The overall average prevalence rate of vitamin D deficiency in pregnant women and newborns is 54% and 75%, respectively. Globally, it is estimated that one billion people are deficient in vitamin D (Morales-Suárez-Varela et al., 2022).

In this study it is known that most pregnant women experience vitamin D deficiency both in the preeclampsia pregnancy group and the normal pregnancy group. In this study, researchers assumed the cause of vitamin D deficiency in pregnant women in the city of Padang was caused by lack of sun exposure in pregnant women where in the pre-eclampsia pregnancy group most did activities outside the home for less than one hour as many as 17 people (63%), as well as in the normal pregnancy group did the most activities outside the home for less than one hour, which was as many as 11 people (40.7%).

In this study, sun exposure was also influenced by employment status as many as 21 people (77.8%) non-working mothers / IRT (housewives) in the preeclampsia pregnancy group and 20 people (74.1%) non-working mothers / IRT in the normal pregnancy group. Work is related to the load of physical activity. Pregnant women who work as housewives more often spend time and do physical activities in the house, ranging from cleaning the house, preparing school children, ironing clothes, cooking, washing clothes and various other activities so that pregnant women rarely leave the house, especially at 10.00-13.00. In addition, many pregnant women wear hijab when doing outdoor activities every day, or cover the entire surface of their body when outdoors even using umbrellas, besides that there are still many pregnant women who are afraid of exposure to sunlight for fear of black.

In the opinion of researchers in this study found low levels of vitamin D in pregnant women in the preeclampsia group and normal pregnancy group also due to other factors such as lack of food intake of vitamin D, nutritional status or body mass index. It is known that pregnant women in the preeclampsia group have the most BMI with obesity (≥27.5 kg / m2) as many as 13 people (48.1%), as well as normal pregnancy groups have the most BMI obesity (≥27.5 kg / m2) as many as 11 people (40.7%). Mothers with obesity are prone to vitamin D deficiency. Vitamin D is a fat-soluble vitamin and fat accumulation throughout the body will interfere with the transport and conversion of previtamin D3 into vitamin D3. Thus, overweight or

obese individuals experience a decreased capacity for vitamin D synthesis (Morales-Suárez-Varela et al., 2022).

Based on the difference in the average levels of 25 serum hydroxyvitamin D from the two groups, it was seen that the average difference in serum 25 hydroxyvitamin D levels was not too far with a value of 0.86 ng / ml, so there was no significant difference between preeclampsia pregnancy and normal pregnancy with p = 0.539.This result is in line with the research of Serrano et al., (2018) which stated that the concentration of serum 25 hydroxyvitamin D levels (25(OH)D) was significantly lower in the group preeclampsia (mean 29.99 ng/mL) compared with normal pregnancy group (mean 33.7 ng/mL) (Serrano et al., 2018). The study of Gholami et al., (2022) also reported that the average serum hydroxyvitamin D levels of the preeclampsia case group (16.60 ±5.87 ng/mL) were lower than the normal pregnancy group (19.06±7.48 ng/mL), in this study found no correlation between vitamin D deficiency and predictive factors for preeclampsia (H. Gholami et al., 2022). Meanwhile, according to research Raia-Barjat et al. (2021) reported that pregnant women with vitamin D or 25(OH)D levels of <12 ng/mL had a 2.4-fold higher risk for early preeclampsia at the age of <35 weeks than patients with vitamin D or 25(OH)D levels of >30 ng/ml (Raia-Barjat et al., 2021)

In the opinion of researchers, there is no significance of the difference in serum 25 hydroxyvitamin D levels in the normal pregnant group and the preeclampsia pregnancy group because nutritional status can be seen from the Body Mass Index. In this study many pregnant women are obese, and underweight. Obesity can cause vitamin D to get trapped. Obesity can lead to increased cytokine-mediated inflammation and oxidative stress, increased shear stress, dyslipidemia, and increased sympathetic nerve activity as major pathways that may be involved in the pathogenesis of preeclampsia, while an underweight BMI can also cause oxidative stress, endothelial disorders and can also trigger preeclampsia (Raia-Barjat et al., 2021).

In this study, statistical results stated that there was no significance of vitamin D levels with the incidence of preeclampsia, also related to the design of research methods and inclusion and exclusion criteria that were still less strict in considering covariables that could be related to the variables studied. This is in line with the meta-analysis research of Aguilar-Cordero et al., (2020) which states that there is no significance or relationship between vitamin D levels and the incidence of preeclampsia due to the design of the research methodology. We recommend that vitamin D supplementation trials are well-designed by establishing strict exclusion criteria, studying each pathology independently and excluding obvious covariables that may affect research results (Aguilar-Cordero et al., 2020).

This study has several limitations, namely the lack of strictness in determining exclusion criteria. There are other factors that can affect vitamin D levels are not assessed such as lifestyle changes, skin pigmentation, season, culture culture, amount and frequency of consumption of vitamin D supplements. In addition, this study also did not assess the level of stress associated with increased cortisol levels

CONCLUSION

There is a difference in serum cortisol levels between preeclampsia and normal pregnancies. There was no difference in serum 25 hydroxyvitamin D levels between preeclampsia and normal pregnancies. It is expected that future researchers will continue this study by assessing the intake of vitamin D levels, the relationship of cigarette smoke exposure, pre-pregnancy body mass index with the incidence of vitamin D deficiency and preeclampsia with stricter inclusion and exclusion criteria. It is expected that future researchers will use cohort design in assessing the effect of vitamin D supplementation and psychological stress on pregnancy on the incidence of preeclampsia. It is expected for health workers to improve the quality of ANC by not only focusing on obstetric or physical problems but also must pay attention to psychological problems besides that it is also expected for health workers to provide education about the importance of vitamin D levels in pregnant women.

BIBLIOGRAPHY

Aguilar-Cordero, M. J., Lasserrot-Cuadrado, A., Mur-Villar, N., León-Ríos, X. A., Rivero-Blanco, T., & Pérez-Castillo, I. M. (2020). Vitamin D, preeclampsia and prematurity: A systematic review and meta-analysis of observational and interventional studies. *Midwifery*, *87*, 102707.

Bärebring, L., O'Connell, M., Winkvist, A., Johannsson, G., & Augustin, H. (2019). Serum cortisol and vitamin D status are independently associated with blood pressure in pregnancy. *The Journal of Steroid Biochemistry and Molecular Biology*, *189*, 259–264.

Black, J. M., & Hawks, J. H. (2014). *Keperawatan medikal bedah: manajemen klinis untuk hasil yang diharapkan*. Elsevier (Singapore).

Botyar, M., & Khoramroudi, R. (2018). Ultraviolet radiation and its effects on pregnancy: A review study. *Journal of Family Medicine and Primary Care*, *7*(3), 511.

Bublitz, M. H., Bourjeily, G., D'Angelo, C., & Stroud, L. R. (2018). Maternal sleep quality and diurnal cortisol regulation over pregnancy. *Behavioral Sleep Medicine*, *16*(3), 282–293.

Chappell, L. C., Cluver, C. A., & Tong, S. (2021). Pre-eclampsia. *The Lancet*, *398*(10297), 341–354.

Dror, R. O., Dirks, R. M., Grossman, J. P., Xu, H., & Shaw, D. E. (2012). Biomolecular simulation: a computational microscope for molecular biology. *Annual Review of Biophysics*, *41*, 429–452.

Eberle, C., Fasig, T., Brueseke, F., & Stichling, S. (2021). Impact of maternal prenatal stress by glucocorticoids on metabolic and cardiovascular outcomes in their offspring: a systematic scoping review. *PLoS One*, *16*(1), e0245386.

Fairbrother, N., Young, A. H., Zhang, A., Janssen, P., & Antony, M. M. (2017). The prevalence and incidence of perinatal anxiety disorders among women experiencing a medically complicated pregnancy. *Archives of Women's Mental Health*, *20*, 311–319.

Farapti, F., & Fadilla, C. (2021). Further Understanding about the Mechanism of Vitamin D on Blood Pressure. *Indonesian Journal of Pharmacy/Majalah Farmasi Indonesia*, *32*(3).

Gholami, H., Fayazi, S., Shirshekan, M., Motamed, N., & Tofighi, S. (2022).

Comparison of Serum level of Vitamin D in Pregnant Women with Preeclampsia and a Control Group in Ayatollah Mousavi Hospital in Zanjan. *Journal of Obstetrics, Gynecology and Cancer Research*, *7*(4), 335–340.

Gholami, M., Ghaziani, R. K., & Eskandari, Z. (2022). Three-dimensional fractional system with the stability condition and chaos control. *Mathematical Modelling and Numerical Simulation with Applications*, *2*(1), 41–47.

Ilmiawati, C., Oviana, A., Friadi, A., & Reza, M. (2020). Sunlight exposed body surface area is associated with serum 25-hydroxyvitamin D (25 (OH) D) level in pregnant Minangkabau women, Indonesia. *BMC Nutrition*, *6*(1), 1–7.

Judistiani, R. T. D., Nirmala, S. A., Rahmawati, M., Ghrahani, R., Natalia, Y. A., Sugianli, A. K., Indrati, A. R., Suwarsa, O., & Setiabudiawan, B. (2019). Optimizing ultraviolet B radiation exposure to prevent vitamin D deficiency among pregnant women in the tropical zone: report from cohort study on vitamin D status and its impact during pregnancy in Indonesia. *BMC Pregnancy and Childbirth*, *19*(1), 1–9.

Karrar, S. A., & Hong, P. L. (2022). Preeclampsia. In *StatPearls [Internet]*. StatPearls Publishing.

Leff-Gelman, P., Flores-Ramos, M., Carrasco, A. E. Á., Martínez, M. L., Takashima, M. F. S., Coronel, F. M. C., Labonne, B. F., Dosal, J. A. Z., Chávez-Peón, P. B., & Morales, S. G. (2020). Cortisol and DHEA-S levels in pregnant women with severe anxiety. *BMC Psychiatry*, *20*(1), 1–14.

Liu, H., Huang, W., Chen, L., Xu, Q., Ye, D., & Zhang, D. (2020). Glucocorticoid exposure induces preeclampsia via DampeningLipoxin A4, an endogenous anti-inflammatory and proresolving mediator. *Frontiers in Pharmacology*, *11*, 1131.

Madhavanprabhakaran, G. K., D'Souza, M. S., & Nairy, K. S. (2015). Prevalence of pregnancy anxiety and associated factors. *International Journal of Africa Nursing Sciences*, *3*, 1–7.

Morales-Suárez-Varela, M., Uçar, N., Soriano, J. M., Llopis-Morales, A., Sanford, B. S., & Grant, W. B. (2022). Vitamin D-Related Risk Factors for Maternal Morbidity and Mortality during Pregnancy: Systematic Review and Meta-Analysis. *Nutrients*, *14*(19), 4124.

Mrema, D., Lie, R. T., Østbye, T., Mahande, M. J., & Daltveit, A. K. (2018). The association between pre pregnancy body mass index and risk of preeclampsia: a registry based study from Tanzania. *BMC Pregnancy and Childbirth*, *18*(1), 1–8.

Polo-Kantola, P., Aukia, L., Karlsson, H., Karlsson, L., & Paavonen, E. J. (2017). Sleep quality during pregnancy: associations with depressive and anxiety symptoms. *Acta Obstetricia et Gynecologica Scandinavica*, *96*(2), 198–206.

Prawirohardjo, S. (2014). Ilmu kebidanan sarwono prawirohardjo. *Jakarta: PT. Bina Pustaka Sarwono Prawirohardjo*.

Purswani, J. M., Gala, P., Dwarkanath, P., Larkin, H. M., Kurpad, A., & Mehta, S. (2017). The role of vitamin D in pre-eclampsia: a systematic review. *BMC Pregnancy and Childbirth*, *17*(1), 1–15.

Raia-Barjat, T., Sarkis, C., Rancon, F., Thibaudin, L., Gris, J.-C., Alfaidy, N., & Chauleur, C. (2021). Vitamin D deficiency during late pregnancy mediates placenta-associated complications. *Scientific Reports*, *11*(1), 20708.

Salustiano, R., Neto, E., & Martinez, M. (2013). The unbalanced load cost on transformer losses at a distribution system. *22nd International Conference and Exhibition on Electricity Distribution (CIRED 2013)*, 1–3.

Serrano, N. C., Guío, E., Quintero-Lesmes, D. C., Becerra-Bayona, S., Luna-Gonzalez, M. L., Herrera, V. M., & Prada, C. E. (2018). Vitamin D deficiency and pre-eclampsia in Colombia: PREVitD study. *Pregnancy Hypertension*, *14*, 240–244.

Sherwood, L. (2014). Fisiologi Manusia; Dari sel ke sistem (Introduction to Human Physiology). *Penerbit Buku Kedokteran EGC*.

Suhardjo, L. J. H., Deaton, B. J., & Driskel, J. A. (1985). Pangan, Gizi dan Pertanian. *UI-Pers Jakarta*.

Usman, A. S. H. H. (2021). Risiko peningkatan hormon kortisol pada hipertensi gestasional. *Jurnal Ilmiah Obsgin: Jurnal Ilmiah Ilmu Kebidanan & Kandungan P-ISSN: 1979-3340 e-ISSN: 2685-7987*, *13*(4), 182–192.

Wang, Z., Lin, Y. S., Dickmann, L. J., Poulton, E., Eaton, D. L., Lampe, J. W., Shen, D. D., Davis, C. L., Shuhart, M. C., & Thummel, K. E. (2013). Enhancement of hepatic 4-hydroxylation of 25-hydroxyvitamin D3 through CYP3A4 induction in vitro and in vivo: implications for drug-induced osteomalacia. *Journal of Bone and Mineral Research*, *28*(5), 1101–1116.

Yıldız, S., Tammo, Ö., & Tammo Sr, Ö. (2022). Comparison of Vitamin D Levels and Related Factors in Pregnant Women and Neonates Exposed to Second-Hand Smoke. *Cureus*, *14*(8).

Zhang, D., Zeng, J., Miao, X., Liu, H., Ge, L., Huang, W., Jiao, J., & Ye, D. (2018). Glucocorticoid exposure induces preeclampsia via dampening 1, 25-dihydroxyvitamin D3. *Hypertension Research*, *41*(2), 104–111.

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