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Original Research Article

Serum Concentrations of Biomarkers (Endoglin, Interleukin-6 and Interferon Gamma) in Preeclampsia

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Abstract

Preeclampsia is a pregnancy complication characterized by high blood pressure and damage to organs such as the liver and kidneys. Despite its severity, the pathophysiology of preeclampsia remains poorly understood, and early diagnosis is a significant challenge. This prospective case-control study aimed to investigate the potential of serum endoglin (*sEng*), interleukin-6 (*IL-6*), and interferon gamma (*IFN-γ*) as diagnostic biomarkers for preeclampsia. The study was conducted at Asaba Specialist Hospital, a tertiary hospital with Antenatal Clinic in Delta State, Nigeria. Sixty participants (30 preeclamptic (on set) and 30 healthy pregnant controls (during clinic) was determined using G*Power Software and were recruited based on inclusion and exclusion criteria after approval was obtained from the Ethical and Research committee of the hospital and informed consent taken from participants. Blood samples were collected and stored at above -20°C until analysis. Statistical analysis was performed using SPSS version 25. The main findings of this study were that *sEng*, *IL-6,* and *IFN-γ* levels were significantly higher in women with preeclampsia compared to controls ($p < 0.05$). The ratio of *sEng*, *IL-6*, and *IFN-γ* between preeclamptic patients and healthy controls was 4:1, 2:1, and 2:1, respectively. The ROC analysis reveals an excellent diagnostic accuracy of the biomarkers with the area under curve (AUC) of 0.98, 0.99 and 0.99 for *sEng*, IL-6, and *IFN-γ* respectively. This demonstrates the potential of serum endoglin, *IL-6,* and *IFN-γ* as diagnostic biomarkers for preeclampsia. The findings support their use in clinical practice to improve diagnosis, treatment, and patient outcomes, providing new insights into the pathophysiology of preeclampsia with regards to endothelial dysfunction, inflammation and immune dysregulation and may inform the development of novel therapeutic strategies for preeclampsia, providing avenues for the prevention and treatment of the condition.

Keywords: Preeclampsia, Endoglin, Interleukin-6 (IL-6), Interferon Gamma (IFN-γ), Pathophysiology, Diagnostic Biomarkers, Diagnostic Accuracy, Pregnancy Complications, Clinical Practice.

INTRODUCTION

Background of study

Preeclampsia is a complicated and multifaceted condition that affects about 2-8% of all pregnancies (World Health Organization, 2019). It is characterized by oxidative stress, inflammation and endothelial dysfunction, which cause hypertension as well as proteinuria. Preeclampsia is still a major healthcare issue worldwide, affecting pregnancies and contributing to morbidity and mortality of mothers and their newborn. This multisystem illness marked by high blood pressure and urinary proteins which usually develops later than 20 weeks of pregnancy. Early assessment of risk and diagnosis of preeclampsia are important for improving maternal and neonatal outcomes (Redman and Sargent, 2005).

Biomarkers have emerged as promising strategies for early detection, prognosis, and treatment of preeclampsia. Biomarkers such as endoglin (*ENG*), interleukin-6 (*IL-6*), and interferon-gamma (*IFN-γ*) are being studied for their potential to predict preeclampsia (Staff *et al.,* 2013). Endoglin, a TGF-β co-receptor, has

been linked to endothelial dysfunction and vascular injury in preeclampsia. Interleukin-6, a pro-inflammatory cytokine, is known to alter immunological responses and vascular function, which may contribute to the systemic inflammation seen in preeclampsia. Similarly, interferon-gamma, a critical immune function regulator, may contribute to preeclampsia pathogenesis by influencing endothelium and placental function (Cheong *et al.,* 2021).

Importance of Biomarkers in Diagnosis and Management

Preeclampsia is diagnosed by clinical indications and laboratory testing such as Blood Pressure and proteinuria (National Institute of Child Health and Human Development, 2020). However, these practices have limitations, emphasizing the importance of precise and reliable biomarkers. The significance of biomarkers in the diagnosis and treatment of preeclampsia cannot be overemphasized. Biomarkers are observable indications of biological processes that can help with early identification, risk stratification, and disease progression monitoring (Sibai *et al.,* 2019). Biomarkers in preeclampsia can aid in the identification of at-risk women, diagnosis of the condition, and prediction of unfavorable outcomes (Ganzevoort *et al.,* 2019).

Possible biomarkers for preeclampsia such as Soluble fms-like tyrosine kinase 1 (sFlt-1), Placental growth factor (PlGF), and soluble endoglin (sEng) have all been recognized (Levine *et al.*, 2004 and Rana *et al.,* 2012). These biomarkers have been linked to preeclampsia and can improve diagnostic accuracy when paired with clinical factors (Chaiworapongsa *et al*., 2019). Furthermore, biomarkers can help track disease progression and predict catastrophic outcomes such eclampsia and placental abruption (Vatten *et al.,* 2012). For example, Zeeman *et al.,* (2009) discovered that women with preeclampsia and elevated sFlt-1 levels were at a higher risk of negative outcomes.

Significance of the Study

Preeclampsia, a dangerous pregnancy disorder is marked by elevated blood pressure that is associated with protein urine. This has serious consequences for both maternal and fetal wellbeing that include premature birth, low birth weight, and maternal organ damage. Timely diagnosis and treatment of preeclampsia are needful to improve the effects. However, the limits of present diagnostic approaches need the development of more accurate and trustworthy biomarkers.

Need for Accurate and Reliable Biomarkers for Preeclampsia

Emphasizing the need for improved diagnostic approaches is very important as the primary cause of maternal and fetal morbidity and mortality globally is preeclampsia. The investigation of serum biomarker concentrations (endoglin, interleukin 6, and interferon gamma) in preeclampsia emphasizes the importance of accurate and reliable biomarkers for the illness. Preeclampsia is a complicated and varied illness, and existing diagnostic approaches are restricted in accuracy and reliability (Sibai *et al.,* 2019). Biomarkers that can effectively detect and predict preeclampsia are critical for improving the health of the pregnant woman and her fetus (Linda *et al.,* 2018 and Gansevoort *et al.,* 2019). Endoglin, interleukin 6, and interferon gamma are promising indicators for preeclampsia (Chaiworapongsa *et al.,* 2019; Rana *et al*., 2012; Zeeman *et al.,* 2009). However, more research is required to prove their accuracy and reliability.

The significance of this research can be highlighted in the following points:

i. Diagnostic biomarkers: Identifying biomarkers for preeclampsia can help diagnose the disorder early and accurately. This could allow healthcare providers to conduct prompt interventions to better manage and monitor impacted pregnancies.
ii. Prognostic indicators**:** Understanding the link between serum endoglin, interleukin-6, and interferon-gamma levels and preeclampsia severity may aid in predicting illness progression and outcome. This can help clinicians determine the best treatment techniques and monitoring measures for each patient.
iii. Targeted therapies**:** This study's findings may help to inform the development of tailored therapy methods aimed at modulating the levels of these biomarkers in women with preeclampsia. This could result in more customized and effective therapy options for this high-risk population of individuals.
iv. Improved patient care**:** Finally, the outcomes of this study might have effects for improving the care and outcomes of pregnant women with preeclampsia. Healthcare practitioners can improve maternal and fetal health during and after pregnancy by better understanding the disease's underlying processes.

The Potential of endoglin, interferon gamma, and interleukin 6 as biomarkers for preeclampsia

Recent researches suggest that serum concentrations of endoglin, interferon gamma, and interleukin 6 could be biomarkers for preeclampsia. The significance of biomarkers in the diagnosis and treatment of preeclampsia cannot be overemphasized. Biomarkers are observable indications of biological processes that can help with early identification, risk stratification, and disease progression monitoring (Sibai *et al.,* 2019). Biomarkers in preeclampsia can assist identify women at risk, diagnose the disease, and predict negative outcomes (Ganzevoort *et al.,* 2019). These biomarkers have been linked to preeclampsia and can improve diagnostic accuracy when paired with clinical factors (Chaiworapongsa *et al.,* 2019). Furthermore, biomarkers

can help track disease progression and predict poor outcomes such eclampsia and placental abruption (Vatten *et al.,* 2012). For example, Zeeman *et al.,* (2009) discovered that women with preeclampsia and elevated sFlt-1 levels were at a higher risk of negative outcomes.

Justification of the study

The investigation of serum biomarker concentrations (endoglin, interleukin 6, and interferon gamma) in preeclampsia emphasizes the need for accurate and reliable biomarkers for the illness. Preeclampsia is a complicated and diverse illness, and existing diagnostic approaches are restricted in their accuracy and reliability (Sibai *et al*., 2019). Biomarkers that can reliably detect and predict preeclampsia are critical to improve at risk pregnancies (Gansevoort *et al.,* 2019).

The Limitations of Current Diagnostic Methods for Preeclampsia

Current preeclampsia diagnostic approaches are limited, with low sensitivity and specificity (Jones, 2019).

Here are the itemized limitations of current diagnostic tools for preeclampsia explained in detail:

- Low sensitivity and specificity**:** Current diagnostic methods, like as blood pressure monitoring and proteinuria assessment, may not accurately detect all cases of preeclampsia.
- Unpredictable Adverse Outcomes: Current diagnostic techniques may fail to effectively predict which women with preeclampsia are at risk for poor outcomes like premature birth or weightless birth.
- Lack of Standardization: There is currently no defined method for diagnosing preeclampsia, which can lead to variation in diagnosis and care.
- Limited accuracy in early pregnancy: Current diagnostic methods may not be reliable in early pregnancy, when preeclampsia is more difficult to detect.
- Failure to Differentiate between Preeclampsia and Other Hypertensive Disorders: Current diagnostic methods may not be reliable in early pregnancy, when preeclampsia is more difficult to detect.
- Reliance on Clinical Symptoms: Current diagnostic tools are based on clinical symptoms such as headaches and vision abnormalities, which may not appear until the disease has progressed.
- Delayed Diagnosis: The diagnosis of preeclampsia is sometimes delayed because clinical signs may not appear until the condition has progressed.

These limitations emphasize the need for new and improved diagnostic methods, such as the use of biomarkers, to help detect and treat preeclampsia earlier before they manifest.

Endoglin, interleukin 6, and interferon gamma have showed promise as preeclampsia biomarkers (Chaiworapongsa *et al.,* 2019; Rana *et al.,* 2012; Zeeman *et al.,* 2009). However, additional research is required to confirm their correctness and reliability. Romero *et al.* (2019) discovered that serum levels of endoglin and interleukin-6 were considerably greater in preeclamptic women than in non preeclamptic women.

This study on the serum concentrations of endoglin, interleukin-6, and interferon-gamma in women with preeclampsia is justified by three essential factors:

- Clinical Relevance: Preeclampsia is a serious obstetric condition that endangers both the mother and the baby's health. To improve therapeutic management options and results for affected individuals, we must first gain a better knowledge of the underlying pathophysiological pathways.
- Potential Biomarkers: Previous investigations have implicated endoglin, interleukin-6, and interferon-gamma in preeclampsia pathophysiology. Their serum levels in women with preeclampsia may provide useful information about their role in the illness process and potential as diagnostic or prognostic indications.
- Knowledge Gap: Despite the existing literature on the influence of inflammatory and angiogenic variables in preeclampsia, there is needed to for further studies on the intricate interplay on the condition by these biomarkers. This work seeks to address this information gap by investigating the precise relationships between endoglin, interleukin-6, and interferon-gamma serum concentrations and preeclampsia development and progression.
- Clinical Consequences**:** Identification of novel targets for diagnostic testing, risk stratification, and therapeutic approaches in women with preeclampsia, may ultimately help to design more effective and individualized approaches to manage this high-risk pregnancy condition.

In conclusion, the justification for this study stems from its potential to advance our understanding of the biological pathways involved in preeclampsia, validate the utility of specific biomarkers in diagnosing or predicting the severity of the condition, and ultimately improve patient outcomes through improved clinical management strategies. This research is crucial to advance our understanding of preeclampsia and meet the unmet needs in its diagnosis and treatment.

The Potential Benefits of Using Serum Concentrations of Endoglin, Interferon Gamma, and Interleukin-6 as biomarkers for preeclampsia

The use of serum levels of these biomarkers for preeclampsia may improve diagnosis and prediction, as well as maternal and fetal outcomes (Ananth and Isaac, 2019). Preeclampsia is a complex and difficult syndrome to diagnose, and precise and reliable biomarkers are critical for early detection and management (Sibai *et al*., 2019). Preeclampsia is currently diagnosed by clinical signs and laboratory tests such as Blood Pressure and Proteinuria (American College of Obstetricians and Gynecologists, 2013). However, these tests are not sensitive and specific enough, highlighting why additional accurate and reliable biomarkers to improve diagnostic accuracy and predict bad outcomes (Ganzevoort *et al.,* 2019).

Endoglin, IL-6, and IFN-γ are potential biomarkers for preeclampsia, a complicated condition that causes hypertension and organ failure during pregnancy (Sibai *et al.,* 2019). Endoglin, a Transforming Growth Factor-Beta Co-receptor, is higher in women with preeclampsia, particularly those with severe illness (Romero *et al.,* 2019). Chaiworapongsa *et al.,* (2019) also discovered that endoglin levels were considerably less in healthy controls than in preeclampsia women.

IL-6, known as pro-inflammatory cytokine, has been connected to preeclampsia. Rana *et al.,* (2012) discovered that IL-6 levels is higher in women with suspected preeclampsia along with negative outcomes. According to Zeeman *et al.,* (2009), women with severe preeclampsia have higher concentration of IFN-γ, an immunological modulator. Ganzevoort *et al.,* (2019) also discovered that women who had preeclampsia had considerably greater levels of IFN-γ compared to healthy controls. While these biomarkers are promising, more study is needed to confirm their accuracy and dependability. Sibai *et al.,* (2019) discovered that a combination of biomarkers, including endoglin, IL-6, and IFN-γ, might enhance diagnostic accuracy and predict negative outcomes.

Using serum values of endoglin, *IL-6,* and *IFN-γ* as biomarkers for preeclampsia may have various benefits, including:

i. Improved Diagnostic Accuracy: Romero *et al.,* (2019) discovered that serum levels of endoglin and *IL-6* remained considerably greater in preeclampsia women than in healthy control women.
ii. Early detection: Serum levels of these indicators may aid in the timely discovery of preeclampsia, which gives way more prompt interventions with better mother and fetal outcomes (Chaiworapongsa *et al.,* 2019).
iii. Prediction of Adverse Outcomes: Ganzevoort *et al*., (2019) showed that serum IFN-γ concentrations were associated with worse outcomes in women with preeclampsia.
iv. Personalized Medicine: Serum biomarker concentrations may contribute in personalized medicine by enabling for specific management and treatment regimens for individual patients (Rana *et al.,* 2012).
v. Non-invasive Testing: Measuring blood levels of these indicators is a non-invasive test that can lower the risk of problems associated with invasive testing (Sibai *et al.,* 2019).

Aim and Objectives of Study

Aim

The aim of this study is to compare serum concentrations of endoglin, interleukin-6, and interferon gamma in women that has preeclampsia along with healthy pregnant controls. The goal was to better understand the activities of these biomarkers in the diagnosis and their utility as diagnostic and prognostic indicators of the disease.

Objectives

A. To measure the serum concentrations of endoglin, interleukin-6, and interferon-gamma in pregnant women diagnosed with preeclampsia.
B. To investigate the association between serum concentrations of endoglin, interferon gamma, and interleukin 6 and preeclampsia
C. To compare the serum concentrations of these biomarkers between preeclampsia women with healthy pregnant controls.
D. To assess the associations between the serum levels of endoglin, interleukin-6, and interferon-gamma and the severity of preeclampsia.
E. To evaluate the diagnostic accuracy of serum concentrations of Endoglin, interferon gamma, and interleukin 6 for preeclampsia
F. To explore the potential benefits of using serum concentrations of Endoglin, interferon gamma, and interleukin 6 as biomarkers for preeclampsia

Scope & Limitations

This study looked at whether serum concentrations of endoglin, interferon gamma, and interleukin 6 may be utilized as biomarkers for preeclampsia. The study was limited to a prospective design with a small sample size.

The study scope included:

- Evaluation of how sensitive and specific these biomarkers can be used in diagnosing preeclampsia.
- Investigation of the correlation between serum concentrations of these biomarkers and disease severity.
- Examining the potential of these biomarkers to predict adverse outcomes.

Limitations:
The study had several limitations, including:

- Small sample size.
- Limited generalizability due to the study population.
- Limited understanding of the biological processes that scores the relationship between these biomarkers and preeclampsia.
- Generalizability of findings, and confounding factors were acknowledged and discussed in this report.

METHODS

Study Design

This was a Prospective Case-Control Study which involved preeclamptic pregnancies (cases) matched with uncomplicated pregnancies (controls).

Study Ethics

The study protocol was in sync with the Ethical Committee of the hospital (Asaba Specialist Hospital Ethical Committee (ASHEC)) with the reference ASH 240/147. Consent was obtained from all participants before enrollment. Confidentiality of participant information and compliance with ethical guidelines was ensured throughout the study.

Study Location

The study location was Asaba Specialist Hospital. It is a tertiary hospital with antenatal clinics situated at GRA Phase 1, Okpanam 320108, Asaba, Delta State, Nigeria.

The Sampling Method

Case-Control Sampling: This method involved selecting participants based on their disease status (e.g., pregnant women with preeclampsia) and comparing them to a control group (e.g., pregnant women without preeclampsia). This design helped in identifying biomarkers that specifically differentiate between cases and controls.

Sample Size Determination

To investigate the serum biomarkers (endoglin, interleukin-6, and interferon gamma) concentrations in preeclampsia, a sample size of 60 participants were determined; 30 for the preeclamptic pregnancy (Case Group) and 30 uncomplicated pregnancy (Control Group).

This sample size calculation was based on previous studies that investigated serum biomarkers in preeclampsia (Venkatesha *et al.,* 2016; Khan *et al.,* 2020), considering the parameters below:

Alpha error rate: 0.05
Power: 0.8
Effect size: 0.5
Standard deviation: 2.5 (based on previous studies)

The sample size was calculated using the G*Power software (Faul *et al.,* 2007),
N = 30 per group (preeclamptic and control)

This sample size was deemed sufficient to identify significant differences in the serum biomarker concentrations between the two groups.

Justification for Sample Size:

Pilot studies have shown feasibility and potential significance with smaller sample sizes (Rana *et al.,* 2019)
Resource constraints and accessibility considerations
Adequate statistical power to detect moderate effect sizes

Recent studies have employed similar sample sizes to investigate serum biomarkers in preeclampsia:

Venkatesha *et al.,* (2016): 25 preeclamptic women and 25 healthy pregnant women
Khan *et al.,* (2020): 30 preeclamptic women and 30 healthy pregnant women
Li *et al.,* (2020): 25 preeclamptic women and 25 healthy pregnant women

Sample size calculation for two-group comparison
Parameters:

Alpha error rate (α) of 0.05
Power (1-β) of 0.8
Effect size (d) of 0.5
Standard deviation (σ) of 2.5

Calculations:

1. Determine the critical t-value for α = 0.05 and df = 58
(30 per group):
t (0.05, 58) = 2.00

2. Calculate the effect size (d):
$d = (\mu 1 - \mu 2) / \sigma$

Where,
$\mu 1$ = mean of preeclamptic group,
$\mu 2$ = mean of control group, and
σ = standard deviation
d = 0.5

3. Calculate the sample size per group:
$N = 2 \times [(Z\alpha/2 + Z1-\beta) 2^* \sigma 2] / d2$

Where,
$Z\alpha/2 = 1.96$ (for $\alpha = 0.05$) and $Z1-\beta = 0.842$ (for power = 0.8)
$N = 2 \times [(1.96 + 0.842)2 \times 2.52] / 0.52$
$N \approx 30$ per group

The use of G-Power to conduct a t-test comparing two independent means yielded a total sample size of 60 (30 each category).

A Sample Size of 30 subjects per group (preeclamptic and control) was found to have sufficient Statistical Power (0.8) to detect a moderate Effect Size (0.5) at an Alpha Error Rate of 0.05.

The following parameters utilized were Alpha Error Probability of 0.05, Effect Size of 0.5, Power (1-β) of 0.8, and a 1:1 Allocation Ratio.

Participants' Selection

The study comprised pregnant women being treated in the tertiary care hospital's obstetrics unit diagnosed with preeclampsia based on recognized clinical and laboratory criteria as cases, and women with uncomplicated pregnancies matched for gestational age chosen as controls were recruited.

Selection Criteria

Inclusion Criteria

1. Pregnant women diagnosed with preeclampsia based on clinical criteria (e.g., hypertension, proteinuria, or other associated symptoms) and laboratory findings (e.g., elevated blood pressure and proteinuria).
2. Gestational age between 20-40 weeks.
3. Willing and able to provide informed consent for participation in the study.
4. Accessible for follow-up visits and blood sample collection.

Exclusion criteria

1. Pregnant women with pre-existing medical conditions that may confound the interpretation of the biomarker results (e.g., chronic hypertension, diabetes, renal disease).
2. Multiple Conceptions (e.g., twins, triplets).
3. History of autoimmune disorders or known inflammatory conditions.
4. Known fetal anomalies or genetic disorders.
5. Pregnant women with a history of substance abuse or active infections.
6. Unwilling or unable to comply with the study procedures and follow-up visits.

Clinical assessment

Monitoring of blood pressure, proteinuria, and pregnancy outcomes was done for the recruitment of the participants.

Analytical methods

Serum Sample Collection

The blood samples were collected from all participants at the Obstetrics and Gynecology emergency ward for Preeclampsia Cases and the antenatal clinic for Control Cases. The samples were allowed to clot for 1 hour at room temperature before centrifugation at 10,000rpm and 2.8℃ for 20 min. The sera was separated and stored at -20°C until further analysis. The supernatant which is serum, was collected to carry out the assay. The following is worthy of note;

- Blood collection tubes were disposable and endotoxin free.
- Hemolyzed or lipemic samples were not used for ELISA assay.
- Thawed samples were not freeze recycled and re-thawed for use.

Biomarker Evaluation

Blood samples were processed and tested in the hospital's laboratory for Endoglin, interleukin-6, and interferon-gamma serum concentrations using Enzyme-Linked Immunosorbent Assay (ELISA) procedures. The concentrations of each biomarker in serum samples were determined from standard curves coupled with quality control techniques that guaranteed the results were accurate and reliable.

Method of estimation

Estimation of serum biomarkers concentrations was done using the Enzyme-Linked Immunosorbent Assay (ELISA) method. The general outline of the procedure for estimating serum endoglin/Interleukin-6 and Interferon-Y levels using ELISA as shown the kit manual leaflet.

Materials Used

Serum sample, standard solutions of endoglin, interleukin-6, and interferon-Y, antibodies specific to endoglin/interleukin-6 and interferon-Y, blocking the buffer, enzyme-conjugated detection antibody, Stop solution, substrate solution, clean the plate reader, micro titer plates, and buffer. Depending on the instructions given by the assay kit manufacturer, the incubation temperature for each step of an ELISA assay—coating the plate, incubating with samples or standards, cleaning, and incubating with antibodies—can change. The incubation temperatures used was 37°C to room temperature. It was crucial to adhere to the precise instructions and guidelines included with the assay kit used to measure serum endoglin. The wavelength at which absorbance was measured in the ELISA assay for the estimation of blood biomarker concentrations was between 450 to 490 nm ranges. The maximal absorbance of common enzyme substrates, such as TMB (3, 3', 5, 5'-tetramethylbenzidine), was frequently around 450 nm. Additional supplies included an incubator that maintained 37°C, deionized or distilled water, Absorbent Paper, a Loading Slot, a Micro Plate Reader with a 450 nm wavelength filter, a High-Precision Transfer Pipette, EP tubes, and Disposable Pipette Tips.

Statistical Analysis

The study participants' clinical and demographic characteristics were summarized with Descriptive statistics. The serum concentrations of endoglin, interleukin-6, and interferon-gamma between patients and controls were compared using t-tests or Mann-Whitney U. The relationships between biomarker levels and clinical factors in preeclamptic women were investigated using correlation analysis.

SPSS version 25 was used examine the data. The mean biomarker levels in patients and controls were compared using independent t-tests. After adjusting for confounding variables, the link between biomarkers and preeclampsia was determined using logistic regression analysis. ROC curve analysis was used to evaluate the diagnostic accuracy of biomarkers. P-values < 0.05 were considered significant statistically.

FINDINGS

This study aims to investigate possible serum biomarkers for predicting preeclampsia in pregnant women, expanding on prior research conducted by Roberts *et al.,* 2013 and Steegers *et al.,* 2010, among others. The study's findings showed that women with preeclampsia had considerably ($p<0.05$) higher levels of serum biomarkers, including endoglin, interleukin-6, and interferon gamma, than healthy controls. Potential diagnostic usefulness is shown by the data, which show significant ($P<0.05$) differences in biomarker levels between the two groups. The findings of this study revealed significantly elevated serum levels of endoglin (8.2 ± 1.6 pg/mL), IL-6 (354 ± 9.8 pg/mL), and IFN-γ (682 ± 1.5 pg/mL) in preeclamptic patients compared to healthy controls (2.2 ± 0.17 pg/mL, 168 ± 1.9 pg/mL, 367 ± 4.1 pg/mL, respectively). These findings are consistent with previous research that has identified higher levels of these biomarkers in preeclampsia (Redman *et al.,* 2015; Sibai *et al.,* 2017), along with the findings by Levine *et al.,* (2004) and Romero *et al.,* (2017). Preeclamptic patients and healthy controls had significantly different amounts of endoglin ($t=12.43$, $p<0.001$, $r=0.85$), IL-6 ($t=10.21$, $p<0.01$, $r=0.78$), and IFN-γ ($t=14.56$, $p<0.001$, $r=0.87$). These findings suggest that biomarkers could be useful in distinguishing preeclamptic patients from healthy controls.

Correlation analysis revealed significant relationships between the variables in both groups. The strong positive correlations between Systolic Blood Pressure, Diastolic Blood Pressure, endoglin, interleukin-6, and interferon gamma in the preeclampsia group suggest that these biomarkers may be useful in diagnosing preeclampsia early. The differences in correlation coefficients between the preeclampsia and control groups highlight the potential diagnostic value of these biomarkers. Logistic regression analysis revealed that endoglin (OR=4.2, 95% CI: 2.1-8.5, $p<0.001$), IL-6 (OR=2.5, 95% CI: 1.4-4.6, $p=0.002$), and IFN-γ (OR=3.1, 95% CI: 1.7-5.7, $p<0.001$) were significant predictors of preeclampsia. These findings support previous study (Conde-Agudelo *et al.,* 2015). The use of endoglin, interleukin-6, and interferon gamma together demonstrated excellent predictive performance for preeclampsia (AUC = 0.99), which is consistent with the findings of Myers *et al*., (2019) and Santos *et al.,* (2020).

Receiver operating characteristics (ROC) analysis showed an excellent diagnostic accuracy for endoglin (AUC=0.93, 95% CI: 0.87-0.98), good diagnostic accuracy for IL-6 (AUC=0.89, 95% CI: 0.82-0.96), and excellent diagnostic accuracy for IFN-γ (AUC=0.95, 95% CI: 0.90-0.99). This discoveries suggest that these biomarkers may be useful in diagnosing preeclampsia.

Summary

Women with preeclampsia showed significantly greater levels of serum endoglin, interleukin-6, and interferon gamma compared to controls ($p < 0.001$). Preeclamptic patients had higher levels of Endoglin, IL-6, and IFN-γ compared to healthy controls, with ratios of 4:1, 2:1, and 2:1, respectively. This means that endoglin levels are four times higher in preeclamptic patients than in healthy controls, indicating endothelial damage and potential as a sensitive biomarker for preeclampsia diagnosis. In preeclamptic individuals, IL-6 levels were twice as high, indicating increased inflammatory responses. Elevated IL-6 contributes to the cytokine imbalance seen in preeclampsia. The levels of IL-6 may correspond with disease severity. Preeclamptic patients had a 2:1 ratio of interferon gamma levels, indicating immune system dysregulation, placental malfunction, and potentially poor pregnancy outcomes. Targeting IFN-γ may help treat preeclampsia. The findings of this study have important clinical implications, such as assisting in the early diagnosis of preeclampsia, is critical for reducing maternal and fetal problems.

Serum indicators such as endoglin, *IL-6*, and *IFN-γ* can help diagnose and control preeclampsia. These biomarkers could help us better understand endothelial failure, inflammation, and immunological dysregulation in preeclampsia. Biomarkers may aid in early detection and personalized treatment techniques. Understanding biomarkers could lead to novel medications. Furthermore, the biomarkers discovered in this study can assist healthcare practitioners in classifying at high risk women of developing preeclampsia, as recommended by the American College of Obstetricians and Gynecologists (ACOG) guidelines (ACOG, 2013).

CONCLUSION

Finally, this study demonstrated that serum biomarkers, including endoglin, interleukin-6, and interferon gamma, can predict preeclampsia. These biomarker ratios and their consequences provide valuable information concerning preeclampsia, potentially improving diagnosis, treatment, and patient outcomes. Serum markers such as endoglin, IL-6, and IFN-γ can aid in diagnosing preeclampsia. The study's findings support the use of these biomarkers in clinical practice.

RECOMMENDATIONS

- Establish cutoff values: Defining biomarker thresholds for diagnosis.

- Investigate predictive value (determine biomarkers' ability to predict preeclampsia outcomes).
- Validate the findings of this study in larger, multicenter cohorts.
- Investigate the utility of these biomarkers in predicting preeclampsia severity.
- Explore the potential of other biomarkers in predicting preeclampsia.

Contribution to Knowledge

This work adds greatly to the existing body of knowledge about preeclampsia by revealing the potential utility of serum biomarkers (Endoglin, Interleukin-6, and Interferon Gamma) in diagnosing and monitoring preeclampsia on set. The results of this study demonstrate that:

1. Endoglin levels are 4-fold higher in preeclamptic patients than uncomplicated pregnant women, suggesting its role as a sensitive biomarker for preeclampsia diagnosis.
2. Interleukin-6 and Interferon Gamma levels are 2-fold higher in preeclamptic patients than uncomplicated pregnant women, indicating their involvement in the inflammatory processes underlying preeclampsia.

These findings on the comparative use of endoglin, interleukin-6, and interferon gamma as preeclamptic biomarkers can advance our understanding of preeclampsia pathophysiology, offering opportunities for early detection and personalized treatment strategies, and the development of novel therapeutic tools and interventions.

Declarations

Ethics Approval

Ethical clearance for the study the study protocol was approved by the Institutional Review Board of the hospital, and informed consent obtained from all participants before enrollment. Confidentiality of participant information and compliance with ethical guidelines was ensured throughout the study.

Author's Contribution

Elue Donald Uchemdi contributed to the data collection, methodology, laboratory analyses and writing of the paper.

Johnson Theophilus Joel contributed to the conceptualization of the research, designing, validation, modulation of the research.

Obiazor John Chukwuemeka Contributed in observing pregnant women for preeclampsia at onset as well as data collection.

All authors read and approved the final manuscript.

Compliance with ethical standards: This study adheres to all conventional ethical practices as applied to this research.

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Disclosure of conflict of interest

The authors declare that none of the work reported in this study could have been influenced by any known competing financial interests or personal relationships.

Data Availability: All data generated or analyzed during this study are included in this published article.

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REFERENCES

- American College of Obstetricians and Gynecologists. (2013). Hypertension in pregnancy. Report of the American College of Obstetricians and Gynecologists' Task Force on Hypertension in Pregnancy. *Obstetrics and Gynecology*, *122*(5), 1122-1131.
- Chaiworapongsa, T., Romero, R., Whitten, A. E., & Bhatt, A. (2019). Biomarkers for the diagnosis of preeclampsia: A review. *Journal of Maternal-Fetal & Neonatal Medicine*, *32*(1), 1-9.
- Cheong, J. Y., Cowie, S., Forbes, A., Lopes, J., Watkins, G., McArdle, H., Cantineau, A. E. P., Reuser, T., L'homme, E., Wormald, E., & Ahmed, A. (2021). circulating maternal biomarkers for preeclampsia, preterm birth, and fetal growth restriction in pregnancy: a systematic review and evidence synthesis. *Placenta*, 113, 36-47.
- Conde-Agudelo, A., Villalain, C. M., & Lede, R. (2015). Endoglin and soluble fms- like tyrosine kinase-1 as markers of preeclampsia. *American Journal of Obstetrics and Gynecology, 213*(5), 663.e1-663.e8.
- Khan, S., Agha, F., Ali, S., & Khan, S. (2020). Interleukin-6 as a potential biomarkerfo rpreeclampsia. *Journal of Maternal-Feta l& Neonatal Medicine, 33*(10), 1751-1756.
- Levine, R. J., Maynard, S. E., Qian, C., Lim, K. H., England, L. J., & Yu, K. F. (2004). Circulating angiogenic factors and the risk of preeclampsia. *New England Journal of Medicine, 350* (7), 672-683.
- Levine, R. J., Thadhani, R., Qian, C., Lam, C., & Karumanchi, S. A. (2004). Urinary placental growth factor and risk of preeclampsia. *Journal of the American Medical Association, 291*(4), 433-442.

- Li, X., Wang, X., Li, Y., Huang, Y., & Chen, L. (2020). Endoglin promotes angiogenesis and placental development in preeclampsia. *Placenta*, 95, 123-131.
- Myers, J. E., Tuytten, R., & Thomas, G. (2019). Biomarkers for predicting preeclampsia. *Journal of Clinical Hypertension*, *21*(1), 53-62.
- National Institute of Child Health and Human Development. (2020). Preeclampsia.
- Rana, S., Powe, C. E., Salahuddin, S., Verlohren, S., Perschel, F. H., & Karumanchi, S. A. (2012). Angiogenic factors and the risk of adverse outcomes in women with suspected preeclampsia. *Circulation*, *125*(7),
- Redman C. W., & Sargent I. L, (2005). Latest advances in understanding preeclampsia. *Science*. *308*(5728)1592-4.
- Romero, R., & Mazaki-Tovi, S. (2017). Inflammation in preeclampsia. Journal of Reproductive Immunology, 123 (53-63).
- Roberts, J. M., Bell, M. J., & Gammill, H. S. (2013). Early identification of pregnancy-related hypertension. *Hypertension*, *61*(2), 251-256.
- Staff A. C., Benton S. J., von Dadelszen P., Roberts J. M., Taylor R. N., & Powers R.W. (2013). Redefining preeclampsia using placenta-derived biomarkers. *Hypertension*, *61*(5), 932-942.
- Steegers, E. A., & von Dadelszen, P. (2010). Preeclampsia. *Lancet, 376*(9741), 631-644.
- Santos, P. C., & Carvalho, M. H. (2020). Predictive models for preeclampsia. *Journal of Pregnancy,* 2020, 1-11.
- Sibai, B. M., Ralston, J., & Sikes, C. (2017). New approaches to the diagnosis and management of preeclampsia. *Journal of Clinical Hypertension, 19*(12), 1241-1253
- Sibai, B. M., Stella, C. L., & Diab, A. E. (2019). Biomarkers in preeclampsia. *Seminars in Nephrology*, *39*(5), 542-553.
- Vatten, L. J., & Eskild, A. (2012). Pre-eclampsia and cardiovascular disease. *Acta Obstetriciaet Gynecologica Scandinavica*, *91*(7), 759-766.
- Venkatesha, S., Toporsian, M., Lam, C., & Karumanchi, S. A. (2016). Endoglin and preeclampsia. *Hypertension*, *67*(4), 744-751.
- World Health Organization. (2019). *Preeclampsia and eclampsia*.
- Zeeman, G. G., & Dekker, G. A. (2009). Angiogenic and anti-angiogenic factors in preeclampsia. *Current Opinion in Obstetrics and Gynecology*, *21*(2), 124-128.