A COMPARATIVE STUDY OF SERUM URIC ACID,C REACTIVE PROTEIN AND SERUM CALCIUM IN PREECLAMPTIC AND NORMOTENSIVE PREGNANT WOMEN.

Dr. Anurag Gundi[1*], Dr. Somnath Salgar[2]

Abstract

Background: The Absence Of Reliable Biochemical Markers That Can Accurately Diagnose Preeclampsia Is Mostly To Blame For The Poor Maternal And Perinatal Outcomes In Preeclampsia Patients. In This Study, We Aimed To Find The Association Of Serum Uric Acid, C- Reactive Protein And Serum Calcium With Preeclampsia.

Methodology: Prospective Case Control Study Was Conducted In B J Medical College And Sassoon General Hospital, Pune Over 12 Months. Total 76 Antenatal Women Of >24 Weeks Of Gestation With 38 Cases Having Preeclampsia And 38 Normotensive Females Were Enrolled And Were Compared For Maternal Uric Acid, Calcium Levels And C Reactive Protein. Independent Sample T Test Was Used To Compare Averages And Roc Analysis Was Done For Prediction Of Preeclampsia.

Result: The Mean Value Of Crp (7.6 ±1,2 Mg/L), Serum Uric Acid (8.75±1.38 Mg/Dl) Was Significantly Higher In The Preeclamptic Group Whereas Serum Calcium In The Preeclamptic Women Was Significantly Lower (7.45±1.5 Mg/Dl) Compared To The Normotensive Women <0.001). AUC For Uric Acid Was (0.853) (P<0.001) And It Was Found 73.3 % Sensitive And 87.7% Specific. AUC For Serum Calcium Was (0.809) (P<0.001) And It Was Found 76.1 % Sensitive And 93.1% Specific. AUC For All Three Parameters Taken Together Was Found 0.919 (P<0.001) And It Was Found 100 % Sensitive And 86.4% Specific In Prediction Of Preeclampsia.

Conclusion: Maternal hyperuricemia, high CRP and hypocalcaemia were associated with preeclampsia. all three biochemical markers taken together showed highest accuracy in prediction of preeclampsia.

Keywords: Hyperuricemia, CRP, Hypocalcemia, preeclampsia.

[1*]Junior Resident 3, Department Of Biochemistry, BJ Government Medical College And Sassoon Hospital,Pune

[2]Md Biochemistry And Professor and Head of the Department, Department Of Biochemistry, BJ Government Medical College And Sassoon Hospital,Pune

***Corresponding Author:** Dr.Anurag Gundi *, Department Of Biochemistry, BJ Government Medical College And Sassoon Hospital,Pune. Mobile No: +91-9518516981, Email ID: anuraggundi1996@gmail.com

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INTRODUCTION

Preeclampsia is a pregnancy-related condition that is characterized by the onset of hypertension and proteinuria after 20 weeks of gestation. It is a systemic syndrome that poses significant health risks to both the mother and the fetus, and can lead to serious complications and even death.[1]

Preeclampsia is often associated with increased levels of uric acid in the blood,[3] which is a marker of oxidative stress, tissue injury, and renal dysfunction. This suggests that uric acid may identify a more severe form of preeclampsia. However, increased serum uric acid is also an independent risk factor for cardiovascular diseases and can mediate altered vascular function and inflammation. The hyperuricemia observed in preeclampsia is believed to result from decreased renal excretion secondary to tissue ischemia and oxidative stress. Hyperuricemia can induce endothelial damage, cause hypertension, and lead to vascular disease.[4]

In preeclampsia, changes in serum calcium, including increased intercellular calcium and hypocalciuria, may contribute to enhanced vascular contractility and elevated blood pressure in the mother. [5] Hypocalciuria in Pregnancy Induced Hypertension (PIH) may be due to abnormal calcium uptake and decreased efflux by the renal tubular cells, leading to decreased serum calcium and increased intracellular calcium, which can also contribute to elevated blood pressure in the mother.[5,6]

Another marker, C-reactive protein (CRP) assay is a simple, quick, and affordable test that requires a simple blood draw and can be used to measure the severity of preeclampsia. CRP values indicates the severity of endothelial cell injury, which is one of the major factors responsible for initiating preeclampsia. Furthermore, the half-life of CRP is 19 hours, and it is cleared from the bloodstream quickly as preeclampsia resolves, making it a good biomarker for the severity of the disease.[7]

The primary objective of this study is to assess the predictive value of three biomarkers, namely serum uric acid, CRP, and serum calcium, for the development of eclampsia in pregnant women. The study will evaluate the levels of these markers in both preeclamptic and normotensive pregnant women and determine their relationship with the onset of eclampsia. The findings of this study can potentially be used to develop a predictive model for the early identification of women at high risk of developing eclampsia, leading to prompt management and improved maternal and fetal outcomes.

MATERIALS AND METHODS

This prospective case-control study was conducted at the Department of Biochemistry in collaboration with the Department of Obstetrics of B.J. Medical College and Sassoon General Hospital, Pune, India. The inclusion criteria for the study were singleton pregnant patients diagnosed with preeclampsia based on specific criteria. These criteria included a blood pressure reading of greater than 140/90, determined by history and clinical examination, along with significant proteinuria (3+) after 20 weeks of gestation. Antenatal women who were in good health, with normal blood pressure readings and without proteinuria on dipstick test, were enrolled as controls.

There were several exclusion criteria for the study. Patients with preeclampsia along with other pregnancy complications, or any other systemic disease such as liver or renal disease, were excluded. Cases with essential hypertension and patients who were not willing to sign an informed consent form were also excluded. Cases with incomplete data were excluded from the study as well.

Procedure

The study involved taking informed written consent from all participants and recording demographic features such as age, gestation, parity, and medical and family history. A physical examination was conducted and blood pressure was measured twice in each participant using a manual mercury sphygmomanometer. Urine analysis was done using a dipstick test to determine the degree of proteinuria. Participants with normal blood pressure and no proteinuria were considered as controls, while those with high blood pressure and proteinuria were enrolled as cases.

Sample collection and laboratory analysis

After overnight fasting blood sample was drawn from antecubital vein using sterile needle and syringe early in the morning for serum calcium and uric acid measurement.

For serum calcium the blood sample was allowed to clot and then centrifuged at 3000 revolutions per minute for 10 min. Serum calcium levels were then measured by O- Cresol Phthalein Complexone (OCPC) method on Biochemistry analyzer. Serum uric acid was measured by enzymatic colour test using Uricase and Peroxidase enzymes. Serum C reactive protein was performed on Biochemistry analyzer using latex agglutination method.

Sample Size Calculation: Sample size estimation was done using online sample size calculator openpi.com based on the Fleiss method with continuity correction by considering two-sided confidence level of 95% and a power of 90%. The ratio of controls to cases is assumed to be 1. The hypothetical proportion of controls with exposure was 10%, and the hypothetical proportion of cases with exposure was 50% which yielded the sample size of 38 for cases and 38 for controls.

Statistical Analysis Method

Primary data was collected using paper-based Case Report forms. Collected data was entered in the Microsoft Excel spreadsheets 2016. Statistical analysis was performed on IBM SPSS STATISTICS VERSION 20. Categorical variables were taken in the form of frequencies and percentages and cross tabulations were done for the chosen parameters and column proportions were compared using Chi square test. Average values were compared using independent sample t test. A receiver operating characteristic curve (ROC curve) using test variables as the biochemistry markers and the outcome variable as preeclampsia was plotted to find the diagnostic ability of a markers. Area under the curve was identified. Sensitivity and specificity was calculated using cross tables. P value < 0.05 was considered significant and p value < 0.01 was considered highly significant.

Observations and Results

A total of 38 preeclamptic women (49.3%) and 38 normotensive women (50.7%) were enrolled in the study. The median age of the participants (n=76) was 29 years and the average age was comparable.

The percentage of primigravida females was significantly higher in preeclamptic group (54.05%) compared to normotensive group (28.95%) ($p<0.05$) however the parity among the two groups was comparable.

The percentage of Obese females (BMI > 30 Kg/m2) was significantly higher in preeclamptic group (21.6%) compared to normotensive females,(2.6%) ($p<0.05$).

Hyperuricemia (serum uric acid> 7mg%) was found in 89.2% of the preeclamptic women compared to 31.6% of the normotensive females,($p<0.0001$).

Increased CRP (> 6mg/dl) was found in 97.3 % of the preeclamptic women compared to 23.8 % of the normotensive females,($p<0.0001$).

Hypocalcemia (Serum calcium < 9 mg/dl) was found in 94.6% of the preeclamptic women compared to 28.9 % of the normotensive females,($p<0.0001$).

Area under the curve for CRP was (0.91) ($p<0.001$) and it was found 79.6 % sensitive and 93.5% specific in prediction of preeclampsia.

Area under the curve for Uric Acid was (0.853) ($p<0.001$) and it was found 73.3 % sensitive and 87.7% specific in prediction of preeclampsia.

Area under the curve for Serum calcium was (0.809) ($p<0.001$) and it was found 76.1 % sensitive and 93.1% specific in prediction of preeclampsia.

Area under the curve for all three parameters taken together was found 0.919 ($p<0.001$) and it was found 100 % sensitive and 86.4% specific in prediction of preeclampsia.

Table 1: Characteristics of the Study Population

Baseline Characteristics		Preeclamptic (N=38)		Normotensive (N=38)		P
		Number	%	Number	%	
Age	≤ 25 years	13	34.2	8	21.1	0.23
	26-30 years	8	21.1	10	26.3	
	31-35 years	11	28.9	12	31.6	
	Above 35 years	6	15.8	8	21.1	
BMI (Kg/m2)	Under weight (≤ 18.5)	1	2.6	0	0	0.012
	Normal (18.6 to 25)	16	42.1	21	55.26	
	Over weight (25.1 to 30)	13	34.2	16	42.11	
	Obesity (> 30)	8	21.1	1	2.63	
Gestational Age	26-30 weeks	16	42.1	13	34.2	0.18
	31-35 weeks	22	57.9	25	65.8	
Parity	0	13	34.21	20	52.63	0.16
	1	12	31.58	13	34.21	
	3	9	23.68	3	7.89	
	3	3	7.89	2	5.26	
	4	1	2.63	0	0.00	
Gravidity	1	11	28.9	20	54.1	0.046
	2	12	31.6	13	35.1	
	3	9	23.7	3	8.1	
	4	4	10.5	1	2.7	
	5	3	7.9	0	0.0	

Table 2: Comparison of Biomarker levels between cases and controls

Biomarker	Levels	Preeclamptic (N=38)		Normotensive (N=38)		P
		Number	%	Number	%	
URIC ACID Ranges (in Mg/dl)	Normal (3 - 7)	5	13.2	26	68.4	<0.0001
	Increased (>7)	33	86.8	12	31.6	
CRP (in mg/dl)	Normal (upto 6)	2	5.3	29	76.3	<0.0001
	Increased (>)	36	94.7	9	23.7	
CALCIUM (in mg/dl)	Normal (> 9)	3	7.9	27	71.1	<0.0001
	Decreased (≤ 9)	35	92.1	11	29	

Table 3: Comparison of average Biomarker values in normotensive Vs.Preelcmaptic Females

		URIC ACID IN mg%	CRP (mg/dl)	CALCIUM IN mg/dl
Normotensive (N=38)	Mean	6.09	3.30	9.17
	SD	1.93	2.77	1.54
Preeclamptic (N=38)	Mean	8.75	7.66	7.45
	SD	1.38	1.20	1.15
Total	Mean	7.40	5.45	8.32
	SD	2.15	3.06	1.61
P		<0.0001	<0.0001	<0.0001

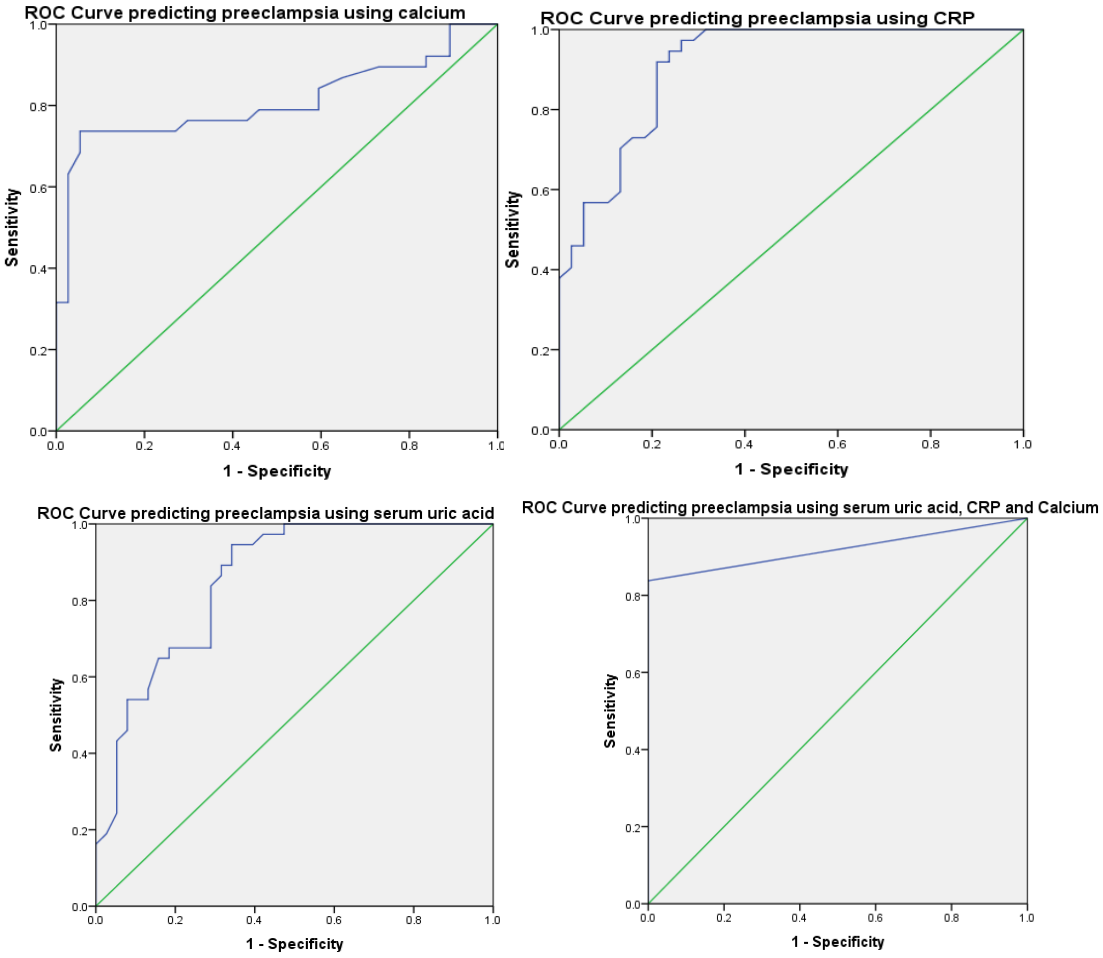

Table 3: Diagnostic Accuracy of Biomarkers using Receiver Operating Curve analysis in prediction of Preeclampsia.

Test Result Variable(s)	Area Under The Curve (AUC)	95% CI for AUC		Sensitivity	Specificity	P
		Lower Bound	Upper Bound			
CRP (Normal- upto 6 mg/dl)	0.91	0.846	0.974	79.55%	93.55%	<0.0001
URIC ACID in mg% (Normal = 3 - 7)	0.853	0.768	0.938	73.33%	86.67%	<0.0001
CALCIUM in mg/dl (Normal= 9-11)	0.809	0.703	0.915	76.09%	93.10%	<0.0001
All three biomarkers	0.919	0.847	0.991	100%	86.36%	<0.0001

DISCUSSION

Pre-eclampsia presents a significant challenge for obstetricians due to its potential to cause multi-organ dysfunction. It is commonly observed in women who have not previously given birth and is characterized by liver function abnormalities and renal function impairment resulting from endothelial cell injury. This condition has numerous associated complications, and it is crucial to minimize its consequences for both the mother and neonate. The current study demonstrates significant alterations in various biochemical parameters, including serum uric acid, calcium, and CRP in preeclamptic women characterized by increased blood pressures, proteinuria and pedal edema compared to normal values in normotensive women.

The study found that normotensive women had a mean gestational age of 30.16±5.97 years, while preeclamptic women had a mean gestational age of 33.19±3.16 years. A recent study by Ugwuanyi et al. [8] reported a mean gestational age of normotensive women as 38.65±1.65 and preeclamptic women as 36.92±4.72. Chaudhary et al. [9] found that the mean gestational age of preeclamptic women was 36.00±2.90 and normotensive women was 31.17±4.30.

In the present study, the mean value of serum uric acid level in the preeclamptic women was significantly higher (8.75±1.38 mg/dl) than in the normotensive women (6.09±1.93 mg/dl) ($p< 0.001$). It is comparable to the study conducted by Wadhwani *et al.(*2021), the mean serum uric acid level was found to be 6.98 ± 1.85 in preeclamptic women and it was 4.55 ± 1.38 in normotensive women.[10] Similarly, in a study conducted by Dhunguna Arun *et al*.(2017) the mean serum uric acid level was found to be 6.14 ± 0.85 in preeclamptic women and the mean serum uric acid level in normotensive women was found to be 4.01±0.62.[6]

In the present study, increased CRP (> 6mg/dl) was found in 97.3 % of the preeclamptic women compared to 23.68% of the normotensive females,($p<0.0001$). Similarly, in a study conducted by Renu *et al*.(2022) showed that 97.83% of the subjects with eclampsia had CRP levels > 6mg/L. In severe preeclampsia group 93.62% subjects and 75.44% subjects in non-severe pre-eclampsia group had CRP levels > 6mg/L. However 92% subjects in control groups had CRP levels < 6mg/L. This difference was found to be highly significant.[11]

In the present study, 97.3 % of the preeclamptic women compared to 23.68% of the normotensive females,($p<0.0001$). Similarly, in a study conducted by Renu *et al*.(2022) showed that 97.83% of the subjects with eclampsia had CRP levels > 6mg/L. In severe preeclampsia group 93.62% subjects and 75.44% subjects in non-severe pre-eclampsia group had CRP levels > 6mg/L. However 92% subjects in control groups had CRP levels < 6mg/L. This difference was found to be highly significant.[11]

In the present study, the mean value of serum calcium in the preeclamptic women was significantly lower 7.45±1.5 mg/dl compared to the normotensive women (9.17± 1.54 $p <0.001$). Hypocalcemia (Serum calcium < 9 mg/dl) was found in 94.6% of the preeclamptic women compared to 28.9 % of the normotensive females,($p<0.0001$). In another study conducted by Dhunguna Arun *et al*.(2017), the mean serum calcium was found to be 8.10±0.56 and in normotensive women, it was 9.59 ±0.62.[6] Another study conducted by Jain *et al*.(2021) showed that the mean serum calcium levels in normotensive women was 9.44±0.82 and in the preeclamptic women was found to be 8.06±0.54. (14)

In this study three Biomarkers were studied simultaneously in this study. Many studies have studied these biomarkers earlier but either in isolataion or in different combinations. This is one of its kind study which has simultaneously studied three parameters which together provide sensitivity of 100%.

AUC for Uric Acid was (0.853) ($p<0.001$) and it was found 73.3 % sensitive and 87.7% specific in prediction of preeclampsia which was similar to Corominas *et al.(*2022) who showed that the AUC was 0.918 [95% CI 0.858–0.979) for the preeclampsia group. The sensitivity and specificity was found to be 90.9% and 83.6%, respectively.[13]

AUC for Serum calcium was (0.809) ($p<0.001$) and it was found 76.1 % sensitive and 93.1% specific in prediction of preeclampsia which was comparable to by El-Maghraby et al.(2022) who reported AUC of 0.898; with the sensitivity of 80 % and specificity of 84 %.[14]

Feto-maternal outcomes could not be studied as all the subjects did not choose to get readmitted for the delivery at the same institute this was one of the limitations of the study.

As per the inclusion criteria, females were enrolled in the third trimester of the pregnancy therefore biochemistry markers were evaluated in the third trimester only. There is scope for future researchers to evaluate these biochemistry markers in the first or second trimester and prospectively follow the cases till delivery to evaluate their prediction

potential for preeclampsia which is common mostly in last trimester.

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