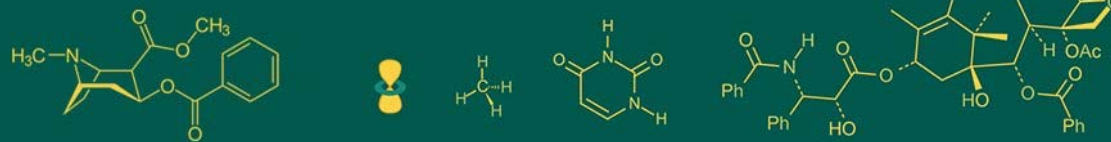


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Study of serum calcium and serum uric acid in preeclampsia cases in and around Chitradurga

Dr. Gaurang K Anandpara and Dr. Basavaraj V Savadi

Abstract

Pre-eclampsia is defined as development of hypertension and proteinuria (> 300mg urinary protein in 24hrs urine) after 20th week gestation. The present study aims to find correlation and usefulness of serum total calcium and uric acid level in preeclampsia. The study was conducted in Basaveshwara Medical College & Hospital from January 2014 to September 2014. Serum Calcium, Uric acid and 24 hours Urinary proteins were estimated. There was a significant decrease in Serum total calcium and increase in serum uric acid in cases of preeclampsia as compare to control group. There was a negative Correlation of serum total calcium and Positive correlation of Serum Uric acid with 24 hour Proteinuria, Systolic & Diastolic Blood Pressure. The study concluded that there was significant hypocalcaemia and hyperuricaemia associated with Preeclampsia. They have greater diagnostic and prognostic value and can be used as screening tests for early prediction of preeclampsia

Keywords: Preeclampsia, Serum Calcium, Serum Uric acid, Proteinuria, Preeclampsia

Introduction

Pregnancy is both desired and demanding for every woman of reproductive age. It is essentially a period that a woman's body undergoes adaptive physiological changes to accommodate fetal growth. One of the potential complications during this period is Preeclampsia. Preeclampsia is defined as the development of hypertension and proteinuria (> 300mg urinary protein in 24hrs urine) after 20th-week gestation. Hypertension is defined as a blood pressure greater than 140/90mm Hg or a rise in blood pressure of 30/15 mm Hg from the baseline confirmed by two measurements 6 hours apart. It is a hypertension disorder of unknown etiology characterized by proteinuria, coagulation abnormalities and different systemic manifestations [1]. Preeclampsia is a leading cause of maternal and perinatal morbidity and mortality worldwide [2]. The incidence of preeclampsia in developing nations is estimated to be 4–18% of total pregnancies. 9% of maternal mortalities in Asia and Africa are said to be due to hypertensive disorders in pregnancy [3]. Preeclampsia occurs in about 5–7% of pregnancies. It is known to affect the function of various organs involving metabolism. Early diagnosis and treatment help to reduce it to a minimum and therefore the importance of identifying the women at risk. The most important feature in toxemia of pregnancy is hypertension which is supposed to be due to the vasospastic phenomenon in kidney, uterus, placenta and brain.

Calcium plays an important role in muscle contraction and regulation of water balance in cells. It has been found that the lowering of serum total calcium and the increase in the concentration of cellular calcium may lead to elevation of blood pressure in preeclamptic pregnant females. Low calcium stimulates the parathyroid hormone which increases the intracellular calcium level leading to smooth vessel contraction and hypertension so dietary deficiency of calcium ions has been shown to have a harmful effect on the pregnant mother and growing fetus and possibly complicate preeclampsia [4]. Calcium supplementation has been reported to half the risk of development of preeclampsia [5].

Uric acid is an end product of purine degradation catalyzed by the enzyme xanthine oxidase. Hyperuricaemia is thought to be a biochemical feature of preeclampsia caused by early tubular retention of urate and disproportionate fall in uric acid clearance [6]. The increase in uric acid level appears to coincide with the increase in the blood pressure and precede the development of proteinuria. The serum level of uric acid rises as preeclampsia progresses; a level >5.5 mg/dl is a strong indicator of the disease and a level >7.8 mg/dl is associated with significant maternal morbidity and fetal morbidity including small-for-

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Gestational-age (SGA) infants and fetal death [7, 8]. The degree of uric acid elevation correlates with the severity of proteinuria and renal pathological changes. Uric acid levels have been used for early diagnosis of pre-eclampsia [9, 10]

The aim of the study was

- (1) To find the association between preeclampsia and serum total calcium.
- (2) To find the association between preeclampsia and serum uric acid.
- (3) To determine the usefulness of serum total calcium and serum uric acid as a biomarker in preeclampsia.

Materials and Methods

This was the Case-control study carried out in Basaveshwara Medical College Hospital & RC, Chitradurga from January 2014 to September 2014. A case-control comparative study was done with 50 cases of Pre-eclampsia and 50 normal pregnant women according to criteria. Detailed personal history, basic information and clinical history were taken from patients. Samples were collected when patient was admitted to Gynec Ward or visited in Gynec OPD.

Selection of study group (cases)

a. Inclusion Criteria

50 Primigravida with pre-eclampsia in the age group of 18-35 years, without any previous history of hypertension, dyslipidemia or other organ dysfunctions above gestational age of 20 weeks were enrolled in the study.

b. Exclusion Criteria

- History of Multigravida, Diabetic Mellitus, Kidney disease.
- History of chronic hypertension and proteinuria before conception or development of hypertension before 20 weeks of gestation
- Patients with chronic renal disease, pathological vaginal discharge
- Patients with a history of recurrent urinary tract infection
- Patients having a molar pregnancy.
- Patients who did not give consent.

Selection of control group

a. Inclusion Criteria

50 age-matched normal Primigravida pregnant women above gestational age of 20 weeks were enrolled in control group.

b. Exclusion Criteria

- History of Multigravida, diabetic mellitus, kidney disease
- History of chronic hypertension and proteinuria before conception or development of hypertension before 20 weeks of gestation
- Patients with chronic renal disease, pathological vaginal discharge
- Patients with a history of recurrent urinary tract infection
- Patients having a molar pregnancy.

- Patients who did not give consent.

Approval for the study was obtained from the Ethical Committee of the Hospital. Signed consent forms were also obtained from all participants before enrolling in the study. Demographic and general information such as age, number of pregnancy and gestational age were recorded.

Blood sample collection and processing

Blood samples were collected from the antecubital vein without stasis. 2 ml blood dispensed into the plain bulb. Serum samples were allowed to clot, afterwards serum was separated immediately by centrifugation at 3000 rpm for 5 minutes. The separated samples free from hemolysis were transferred into aliquots, labeled appropriately and analyzed in the Biochemistry laboratory for the estimation of serum uric acid and serum total calcium.

Estimation of Serum total calcium

Serum total calcium was determined by the Arsenazo method in a fully auto analyzer Erba 200 machine.

Estimation of Serum Uric acid

Serum Uric acid was determined by the Uricase enzymatic method in a fully auto analyzer Erba 200 machine.

Urine sample collection and processing

All patients underwent 24-hour urine collection as the gold standard for diagnosis of Preeclampsia, 24 hours urine was collected in all patients with assistance of nursing staff, Each container was marked with patient's name, number of container and collection time.

Estimation of 24 hours Urinary Protein

The urinary protein level was determined by the Pyrogallol method in the semi auto analyzer Erba Chem 5 Plus machine.

Statistical analysis

The results were documented, entered and analyzed through Excel 2007. Results are reported as mean \pm standard deviation. Mean and standard deviation was calculated for age, gestation age, 24-hour proteinuria, serum total calcium and serum uric acid. Chi square test and odds ratio were calculated for serum total calcium and serum uric acid levels in both groups. Pearson's correlation (r) was used to study the correlation between the study parameters. Probability value (p) < 0.05 was considered as statistically significant.

Results

Table 1: Comparison of variables between cases and controls

Variables	Cases(n=50) [Mean \pm SD]	Controls(n=50) [Mean \pm SD]
Patient age (years)	22.58 \pm 3.81	22.98 \pm 3.65
Gestational Age (weeks)	28 \pm 4	27 \pm 5
BMI (kg/m ²)	26.92 \pm 2.64	23.58 \pm 4.35
Systolic Blood Pressure (mm Hg)	150 \pm 8	120.12 \pm 6.01
Diastolic Blood Pressure (mm Hg)	98.96 \pm 7.76	74.28 \pm 3.23
Serum total calcium(mg/dl)	8.37 \pm 0.52	9.43 \pm 0.59
Serum Uric acid(mg/dl)	7.04 \pm 0.91	4.61 \pm 1.05
24 hour Urinary Proteinuria (gm)	1.620 \pm 1.365	0.139 \pm 0.054

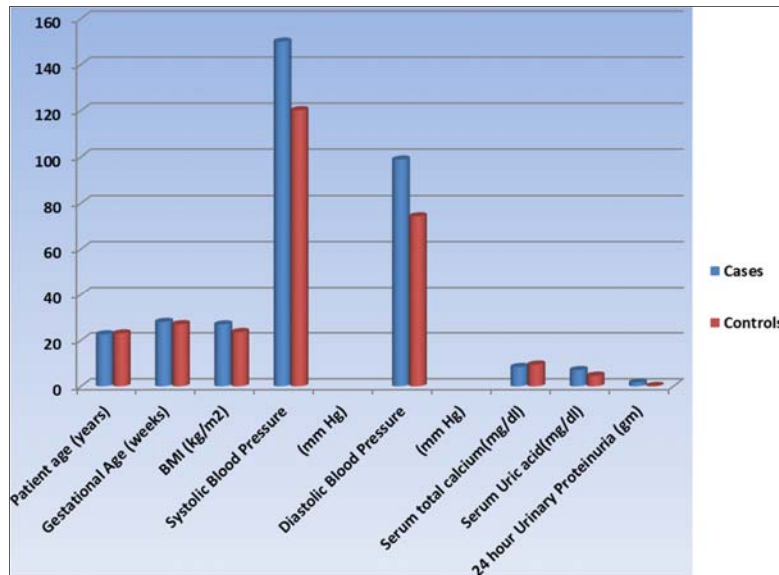


Fig 1: Comparison of variables between cases and controls

Table 1 and Figure 1 shows the variables of cases and controls including age, gestational age, BMI, Blood Pressure, serum calcium, serum Uric acid and 24 hrs Proteinuria.

The results of serum total calcium and serum uric acid were having normal reference range as below:

1. Serum Total Calcium: 9-11 mg/dl
2. Serum Uric acid: 2.5-6 mg/dl

Table 2: Comparison of serum total calcium and serum uric acid in Mild Preeclampsia and Severe Preeclampsia

Biochemical Parameter	Severe Pre eclampsia (n=12) [Mean + SD]	Mild Pre eclampsia (n=38) [Mean + SD]
Serum total calcium (mg/dl)	7.77±0.30	8.56±0.41
Serum Uric acid (mg/dl)	8.15±0.52	6.69±0.70

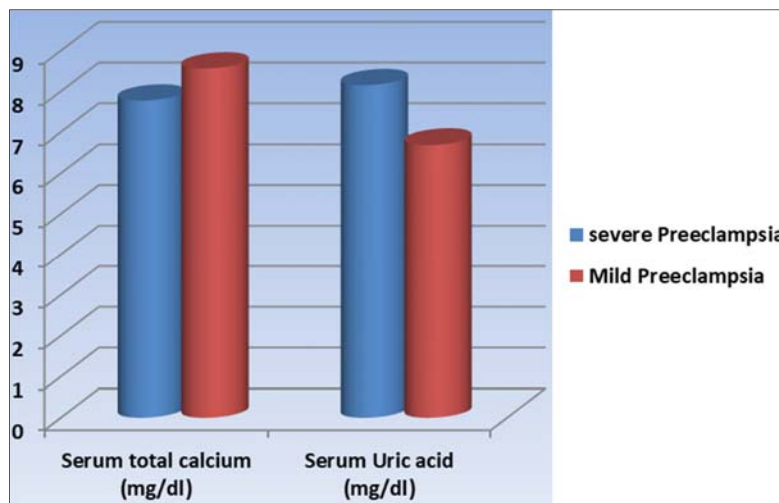


Fig 2: Comparison of serum total calcium and serum uric acid in Mild Preeclampsia and Severe Preeclampsia

Table 2 and figure 2 shows that mean serum total calcium is significantly lower in severe pre eclampsia as compare to mild eclampsia and serum uric acid is significantly higher in severe pre eclampsia as compare to mild eclampsia.

Table 3: Distribution of serum calcium among cases and controls

Serum calcium	Cases	Control	Total
7.0-9.0(mg/dl)	40	19	59
9.0-11.0(mg/dl)	10	31	43

Chi square value=18.23 and p value is <0.0001. The result is significant at p<0.05. Odds ratio is 6.52.

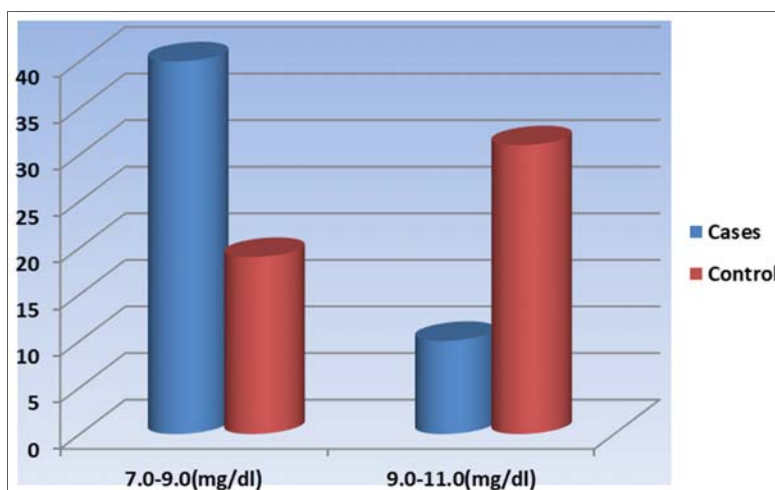


Fig 4: Distribution of Uric acid among cases and controls

Table 3 and figure 3 shows the distribution of serum calcium in cases and controls which show highest number of patients having hypocalcaemia belongs to pre eclampsia group.

Table 4 and figure 4 shows the distribution of serum uric acid in cases and controls which show highest number of patients having hyperuricaemia belongs to pre eclampsia group.

Table 5: Correlation of Serum Total Calcium, Serum Uric acid with Systolic & Diastolic blood Pressure and 24 hour Proteinuria

Parameters	24 Hour Proteinuria	Systolic BP	Diastolic BP	p Value
Serum total Calcium	-0.51	-0.46	-0.43	<0.05
Serum Uric Acid	0.56	0.76	0.53	<0.05

Table 5 shows the negative Correlation of Serum total Calcium with 24 hours Proteinuria, Systolic & Diastolic Blood Pressure and Positive correlation of Serum Uric acid with 24 hours Proteinuria, Systolic & Diastolic Blood Pressure.

Discussion

Our case-control study was carried out in Basaveshwara Medical College Hospital & RC, Chitradurga from January 2014 to September 2014.

Our study showed that there was no significant difference between maternal age, gestational age and body mass index between 2 groups. As expected, the systolic and diastolic blood pressure were significantly higher in the preeclampsia group [Table 1]. The results of our study showed that mean serum total calcium was 8.37 mg/dl in preeclampsia women and 9.43 mg/dl in control group ($p < 0.05$), so serum total calcium was significantly low in preeclamptic women as compared to control group. Mean serum Uric acid was 7.04 mg/dl in preeclamptic women and 4.61 mg/dl in control group ($p < 0.05$), so serum uric acid was significantly higher in preeclampsia women as compared to control group [Table 1].

On analyzing in detail, it showed that Out of 50 cases of preeclampsia, 12 had severe preeclampsia and 38 had mild preeclampsia [table 2]. Mean serum total calcium was 7.77 mg/dl in severe preeclampsia and 8.56 mg/dl in mild preeclampsia respectively [Table 2]. It shows that the serum total calcium was significantly decreased in severe preeclampsia as compare to mild preeclampsia. These results are in accordance with previous studies of Sukonpan K, *et al* and Kisters K, Barenbroka M, *et al*. They observed an inverse relationship between serum total calcium values and incidences of preeclampsia [11, 12].

In 50 cases of preeclampsia, 40 women (80%) had mean serum total calcium in between 7-9 mg/dl and the rest of 10 women (20 %) had mean serum total calcium in between 9-11 mg/dl. In normal pregnant women (controls), 19 women (38%) had mean serum total calcium in between 7-9 mg/dl, rest of 31 (62%) women had mean serum total calcium in between 9-11

Mg/dl. The odds ratio is 6.52 in our study which indicates that women with hypocalcaemia are 6 times more likely to develop preeclampsia [Table 3].

In our study, there was a significant negative correlation between Serum total calcium and systolic blood pressure, diastolic blood pressure and 24 hour proteinuria (correlation coefficient is -0.46, -0.43 and -0.51 respectively). which suggests that strong association between deficiency of calcium and progression of preeclampsia. our findings are similar to [13, 14, 15, 16, 17] [Table 5].

Calcium metabolism is under immense dynamism during pregnancy. Expectant mothers need to store about 30-50 gm of calcium during the course of pregnancy, of which 25 gm is needed by the fetus. Eighty percent of the total fetal calcium is deposited during the third trimester. The transport of ionized calcium from mother to the fetus increases about 50 mg/day at 20 weeks of gestation to a maximum of about 350 mg/day at 35 weeks of gestation [18]. The protective effect of calcium on blood pressure can be explained in part by the influence of calcitrophic hormones on intracellular calcium. Unavailability of this element due to deficiency or decreased concentration may be a predisposing factor in the development of pre-eclampsia or a contributory factor in its pathogenesis. The limitation of this study was that the dietary intake of calcium was not taken into consideration.

In cases of preeclampsia, mean serum uric acid was 8.15 mg/dl in severe preeclampsia and 6.69 mg/dl in mild

preeclampsia [Table 2]. It shows that the serum uric acid was significantly increased in severe preeclampsia as compare to mild preeclampsia.

In 50 cases of preeclampsia, 28 women (56%) had mean serum uric acid in between 6-8 mg/dl and rest of 22 women (44 %) had mean serum uric acid < 6 mg/dl. In normal pregnant women (controls), 06 women (12%) had mean serum uric acid in between 6-8 mg/dl, rest of 44 (88%) women had mean serum uric acid < 6 mg/dl [table 3,4]. The odds ratio is 9.33 in our study which indicates that women with hyperuricaemia are 9 times more likely to develop preeclampsia. This result is comparable with study of Lim *et al* which showed the mean serum uric acid values for women with preeclampsia (6.2 ± 1.4 mg/dl) were significantly higher than controls (4.3 ± 0.8 mg/dl, $p < 0.05$)^[19], Williams *et al* concluded that uric acid levels are significantly elevated in women with gestational hypertension and preeclampsia as compared to normotensive pregnant women which is similar to our study^[20].

In our study, there was a significant positive correlation of between serum uric acid and systolic blood pressure, diastolic blood pressure and 24-hour proteinuria (correlation coefficient is 0.76, 0.53 and 0.56 respectively) [Table 5].

During pregnancy maternal serum uric acid levels initially fall, with a subsequent rise to pre-pregnancy levels near term^[21]. In the third trimester, there is a rise in uric acid which may be related to an increase in fetal uric acid production or a decrease in uric acid clearance^[22]. Elevated serum uric acid levels due to decreased renal urate excretion are frequently found in women with preeclampsia. Besides the reduced clearance, hyperuricaemia in pre-eclampsia may be due to increased uric acid production caused by trophoblast breakdown, cytokine release and ischemia^[23]. Uric acid can promote endothelial dysfunction, damage and inflammation, which leads to oxidation. So, pre-eclampsia, which is characterized by widespread endothelial dysfunction and inflammation, might be propagated by uric acid^[24].

Conclusion

The study concluded that there is significant association of hypocalcaemia and hyperuricaemia in Preeclampsia and their levels vary with severity of disease progression. Hyperuricaemia is one of the earliest and consistent observations of preeclamptic patients. Hypocalcaemia may have a role in etiology of preeclampsia and dietary interventions with calcium reduce the risk of preeclampsia. This study helps in to establish preventive strategy for preeclampsia by calcium supplementations.

The estimations of serum total calcium and uric acid have greater diagnostic and prognostic value in preeclampsia. Their estimations are relatively cheap and can be used as a screening test for early prediction of preeclampsia.

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Conflict of interest: None.

Contribution of authors: Each author contributes equally to prepare this manuscript.

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