

## Study Of Urinary Calcium/Creatinine Ratio(CCR) In A Spot Sample Of Urine For Early Prediction Of Preeclampsia.

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### Abstract

**Objectives:** To assess the efficacy of calcium /creatinine ratio (CCR) as a diagnostic test for the prediction of preeclampsia, and also to determine the changes in urinary excretion of calcium in preeclampsia and normotensive women. **Material & Methods:** Urinary calcium creatinine ratio was determined in a spot urine sample in 145 asymptomatic pregnant women between 20-28 weeks of gestation, who attended the antenatal OPD at Rama Medical hospital and Research Centre, Ghaziabad. The results were analyzed by Chi square test and t test to find the significant association of findings of preeclampsia and CCR. Area under Receiver Operator Curve (ROC) was used to find the predictive values of CCR at less than or equal to 0.04.

**Results:** It was found that CCR had a sensitivity, specificity, Positive Predictive Value (PPV) and Negative Predictive Value (NPV) of 81.2%, 96.4%, 86.6%, and 94.7% respectively with a statistical accuracy of 93.1% and p value of <0.001 (strongly significant). It was found to be a good test for prediction of preeclampsia.

**Conclusion:** CCR at 0.04 in spot urine sample being a good test for prediction of preeclampsia can be recommended as a screening test in all asymptomatic pregnant women.

**Keywords:** Calcium/creatinine ratio(CCR), Preeclampsia, Receiver Operator Curve(ROC)

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### I. Introduction

Hypertensive disorders of pregnancy, such as preeclampsia(PE) and pregnancy-induced hypertension(PIH) are a major cause of maternal morbidity<sup>1</sup>. About 5-7 % of the pregnancies are affected by preeclampsia and hypertensive disorders<sup>2</sup>. It accounts for more than 40% of pre-mature deliveries and associated complications of pre-maturity<sup>3</sup>.

Prediction of Preeclampsia in a patients extremely essential so that early detection and timely intervention and treatment will prevent the complications of pregnancy induced hypertension to occur. A variety of biochemical and biophysical markers, based primarily on rationales implicated in the pathology and pathophysiology of hypertensive disorders due to pregnancy have been proposed for the purpose of predicting the development of preeclampsia later in pregnancy.<sup>4,5</sup> But none of them have been proved ideal either because of high incidence of false positivity or their complexity in result interpretation. It has been found that decreased urinary excretion of calcium may be considered as an useful tool for the early diagnosis of preeclampsia<sup>6</sup>. Reduction in urinary calcium excretion was observed at 10-24 weeks of gestation in patients who later developed preeclampsia and persisted throughout pregnancy<sup>7</sup> When used as a single test, the urinary Ca:Cr ratio is a better predictor of preeclampsia than the urinary microalbuminuria concentration<sup>8,9,10,11</sup>. The present study was to estimate the urinary calcium and creatinine ratio in prediction of preeclampsia. It is a simple and noninvasive screening test which is also cost effective.

The aims of this study were to investigate whether hypocalciuria precedes preeclampsia in pregnant women with gestational hypertension, and whether the urinary calcium/creatinine ratio is a reliable predictor of emerging preeclampsia in such a group.

### II. Material & Methods

A prospective study was conducted on total 145 pregnant women between 20 to 28 weeks of gestation who have been admitted in the Department of Obstetrics and Gynaecology, Rama Medical hospital and Research Centre, Ghaziabad from august 2014 to october 2015. Out of these 32 developed preeclampsia based on defined criteria. The study population of 145 pregnant women was divided into two groups; Preeclamptic and Normotensive.

These women were aged between 18-30 years with mean age of 24 years and who were following up with their regular antenatal checkups. A detailed clinical history regarding age, parity, socioeconomic status, past, family and personal history are taken with general physical and obstetric examination. Women with H/o chronic hypertension BP >140/90 mm of Hg, Diabetes mellitus, renal disease are excluded from the study.

None of the women received any antihypertensive medication until study samples were taken. The presence of PIH was defined as the recorded blood pressure more than 140/90 mm of Hg or a rise of 30 mm of Hg in systolic pressure or of 15mm of Hg in diastolic pressure ( measured twice 6 hrs apart at bed rest ) associated with proteinuria or oedema or both.

**Analytical methods:** The early morning first urine sample from all patients was collected in calcium free vials. 10ml of urine was collected and 0.2ml HCL was added to prevent calcium salt precipitation. Calcium and creatinine levels were estimated in the collected samples. Urinary Calcium was estimated by Arsenazo Dye method and Urinary Creatinine was estimated by Jaffe's Kinetic method by Erba's Semi autoanalyzer.

All the collected data was reviewed and analysed for urinary calcium-creatinine ratio (CCR) in both the study and control groups. Cut off for CCR is taken as  $\leq 0.04$  <sup>10</sup>.

So, the ratio is given as 
$$\frac{\text{Urinary Calcium(mg/dl)}}{\text{Urinary Creatinine(mg/dl)}}$$

**Statistical Analysis:** Results are expressed as mean±SD. *Chi square test* and 't' test were used for analysis of results.  $p < 0.05$  was considered as statistically Significant. The best cutoff point was established by analyzing the receiver operating characteristics(ROC) curves. Sensitivity, specificity and positive and negative predictive values were calculated. Diagnostic accuracy of the test was also analyzed.

### III. Results

In this prospective study 145 pregnant women of gestation 20 to 28 weeks were enrolled. Out of these 32 developed preeclampsia based on defined criteria. Thus the incidence of preeclampsia in study is 22%. The study population of 145 pregnant women was divided into two groups; Preeclamptic and Normotensive. Anthropometric, Clinical and laboratory characteristics of patients are presented in table 1. The majority of subjects were of age group 21-30 yrs(75%). Result showed the maternal mean age 24.16±5.14 years and 23.87±5.62 years in normotensive and preeclamptic respectively which is almost similar. It was observed that most of them i.e. 73 (64.6%) out of 113 normotensive and 21 (65.6%) out of 32 preeclamptic women were nulliparous. Family history of preeclampsia found insignificant (pvalue 0.15) among both groups. No significant difference was seen in mean blood pressures of both groups at first visit of test performed, but significant difference was noted in mean systolic and diastolic blood pressure in the subsequent follow up (p value < 0.05).

**Table.1:** Anthropometric, clinical and laboratory characteristics

Parameters	Normotensive group(n=113)	Preeclamptic group (n=32)	p value
Maternal age(yrs)	24.16±5.14	23.87±5.62	>0.05
SBP(mm Hg)	108±9.23	167±12.23	<0.05
DBP(mm Hg)	67.35±7.83	102±6.74	<0.05
BMI(Kgm <sup>-2</sup> )	27.25±6.78	29.34±10.43	>0.05
Serum calcium(mg/dl)	9.06±0.52	8.91±0.43	<0.05
Serum creatinine(mg/dl)	0.77±0.12	0.88±0.45	<0.05

Table :2. shows urinary calcium , urinary creatinine and urinary CCR values in case and control groups. Urinary calcium excretion in preeclamptic group was 3.8±2.03mg/dl which was found to be significantly low (p value < 0.01) as compared to control 22.2±4.33 mg/dl. The mean urinary creatinine levels in cases with development of preeclampsia was 129.32±11.21 mg/dl and without preeclampsia was 116.32±13.29 mg/dl (p value < 0.01). The preeclamptic women had urinary CCR of 0.043±0.021 while normotensive pregnant women had 0.234±0.09 values. The mean difference of 0.191 was observed between normotensive and preeclamptic. Statistical analysis showed that the urinary calcium/creatinine ratio in preeclamptic pregnant women were significantly lower than normotensive pregnancies. The p value was highly significant (p < 0.001).

**Table.2:** Comparison of urinary calcium, urinary creatinine & urinary calcium to creatinine ratio between normotensive pregnant women and Preeclamptic patients

Parameters	Normotensive group(n=113 )	Preeclamptic group(n=32)	p value
Urine calcium(mg/dl)	21.2±4.33	3.8±2.03	<0.01
Urine creatinine(mg/dl)	129.32±11.21	116.32±13.29	<0.01
Urine calcium/creatinine ratio	0.234±0.09	0.043±0.021	<0.001

With the use of R.O.C.(Receiver operator curve) of a urine calcium:creatinine ratio threshold value of 0.04 was taken as cut off for prediction of preeclampsia . The chosen cut off point < 0.04 for CCR was considered abnormal and cut off > 0.04 for CCR considered normal. At these cut off values the number of true positive, true negative, false positive and false negative were determined.

**Table: 3.**Distribution of Subjects according to urinary calcium creatinine ratio (CCR)

Group	CCR>0.04	CCR≤0.04
Normotensive patients (n=113)	109(96.4%)	4(3.53%)
Preeclamptic group (n=32)	6(18.7%)	26(81.25%)

In study group, among 32 patients with PIH, 26 patients (81.25%) had CCR≤0.04 and 6 patients (18.7%) had CCR>0.04, whereas in control group 109 patients (96.4%) had CCR >0.04 and 4 patients (3.53%) had CCR≤0.04. (table:3)

**Table : 4.**Result of statistical analysis

Test	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)	Accuracy (%)	pvalue (%)
CCR	81.2	96.4	86.6	94.7	93.1	<0.001

It is evident from the table:4 that the ratio of calcium/creatinine(≤0.04) test has sensitivity of 81.2%, specificity of 96.4%, positive predictive value of 86.6%, negative predictive value of 94.7%., and diagnostic accuracy of 93.1%.

#### IV. Discussion

Pregnancy induced hypertension is a multifaceted syndrome with involvement of several important organs<sup>12,13</sup>. PIH is also associated with endothelial dysfunction<sup>14,15,16</sup>. In this study preeclamptic women had reduced urinary excretion of calcium compared with those normal pregnancies. Our observation that preeclamptic women have hypocalciuria is in accord with reports from other investigators<sup>8, 17,18</sup>. These authors suggested that the urinary calcium excretion levels may serve as a diagnostic tool for differentiating between the various forms of hypertension in pregnancy. The aetiology of hypocalciuria in preeclamptic patients is unknown. It has been speculated that hypocalciuria may result from decreased dietary intake, decreased intestinal absorption, increased calcium uptake by the fetus and placenta, or intrinsic renal tubular dysfunction<sup>19</sup>. Taufield et al<sup>17</sup> observed marked hypocalciuria in patients with hypertensive disorders of pregnancy and suggested increased distal tubular reabsorption of calcium as a possible mechanism. Pedersen et al<sup>20</sup> in a longitudinal study reported that urinary calcium excretion was considerably lower in the third trimester of preeclamptic pregnant women than in both pregnant subjects and non-pregnant controls. They suggested that these changes could partly be related to a decrease in the glomerular filtration in pre-eclampsia. The authors also found reduced tubular reabsorption in preeclamptics because the fractional excretion was decreased, a finding which is similar to ours in this study. While others noted decrease of urinary creatinine in pre eclampsia, Mittal Shilpa et al.,<sup>21</sup>, Kazemi AFN et al.,<sup>22</sup> and Moni SY et al.,<sup>23</sup> observed increase in urinary creatinine in preeclamptic patients. In this study an attempt was made to show the relation between low urinary calcium creatinine ratio (CCR) and P.I.H. With the use of R.O.C (Receiver operator curve) urinary Ca: Cr ratio threshold value of 0.04 was taken as cut off for prediction of preeclampsia. It is observed that the urinary excretion of calcium and creatinine is increased during pregnancy but when pregnancy is associated with high risk factors like hypertension, their excretion is reduced resulting in development of preeclampsia. So, we find a definite relationship between low calcium creatinine ratio and development of preeclampsia. The mean level of Urinary Calcium: Creatinine ratio was significantly decreased (p < 0.001) in preeclamptic cases as compared to control group. On statistical analysis it was found that CCR could be taken as high risk factor for prediction of PIH as it has highly significant p value of <0.001, sensitivity of 81.2%, specificity of 96.4%, positive predictive value of 86.6%, negative predictive value of 94.7%. The diagnostic accuracy of test was found 93.1%. These findings are coinciding with results of Ozcan et al<sup>16</sup> 1995. Ozcan et al<sup>16</sup> who investigated the predictive value of decreasing calcium to creatinine ratio in a spot urine sample as early as in 1995 reported that it might be an effective marker for preeclampsia. Saudan<sup>24</sup>, has reported a sensitivity of only 68% and specificity of 70% and Izumi et al<sup>25</sup> found that it had limited value in prediction of preeclampsia, but the threshold value used was 0.10 by the former and Izumi et al carried out the test in early pregnancy, at less than or equal to 12 weeks of gestation. Kazerooni<sup>26</sup> evaluating between 20-24 weeks of gestation and Karetal<sup>10</sup> evaluating the predictive value of CCR at less than or equal to 0.04 between 20-34 weeks of gestation (similar to our study), have reported that it was a satisfactory test for prediction of preeclampsia and could be an effective method for screening asymptomatic women for preeclampsia. Rodriguez et al<sup>8</sup> evaluated the role of decreasing calcium creatinine ratio and micro albuminuria in prediction of preeclampsia as early as in 1988 and have concluded that these tests may be useful screening tools in prediction of preeclampsia. Our findings are similar to these investigators.

## V. Conclusion

We can conclude that a single estimation of calcium to creatinine ratio at less than or equal to 0.04, in a spot urine sample, in asymptomatic pregnant women between 20-24 weeks of gestation is a good predictor of future development of preeclampsia and therefore can be recommended as a screening test for all pregnant women as it justifies the cost.

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