

A Hospital Based Prospective Study to Determine the Efficacy of Urinary Calcium to Creatinine Ratio in a Spot Urine Sample for the Prediction of Preeclampsia in Asymptomatic Pregnant Women Between 16 to 24 Weeks of Gestation

Pradeep Kumar Chouhan¹, Usha Rangey Chouhan^{2*}

¹Principal Specialist, Department of Medicine, Government District Hospital, Sirohi, Rajasthan, India

²Senior Specialist, Department of Obst. & Gynae., Government District Hospital, Sirohi, Rajasthan, India

Received: 27-10-2021 / Revised: 11-12-2021 / Accepted: 02-01-2022

Abstract

Background: Hypertensive disorder of pregnancy are one of the major causes of maternal and fetal mortality and morbidity. During pregnancy, by increasing intestinal absorption, decreasing renal calcium loss, and reabsorbing some calcium from the maternal skeleton the woman's body undergoes physiological compensation. The aim of this study to determine the efficacy of urinary calcium to creatinine ratio in a spot urine sample for the prediction of preeclampsia in a asymptomatic pregnant women between 16 to 24 weeks of gestation. **Materials & Methods:** A hospital based observational prospective study done on hundred primigravida / multigravida with singleton pregnancy within the gestational age of 16-24 weeks who came for regular ANC to OPD in government district hospital, Sirohi, Rajasthan, India during one year period. Urine Calcium-to-creatinine ratio (CCR) was calculated, and those with a ratio of less than or equal to 0.04 were considered as test positive. Descriptive statistics frequency analysis was used to describe about the data, percentage analysis were used for categorical variables and for continuous variables the mean & S.D were used. **Results:** The incidence of Preeclampsia was 15% and that of normotensive pregnant women was 85%. Amongst the patients who had a Urine Ca/Cr ratio <0.04, 15 women constituting 88.23% developed preeclampsia whereas 2 patients constituting 11.76% remained normotensive at term gestation. **Conclusion:** We conclude that a pregnant woman with a high risk factor such as nulliparity along with low urinary CCR is at high risk for development of preeclampsia. Therefore, a single urinary calcium to creatinine ratio may be an effective screening method for impending preeclampsia and may identify population at greater risk to be included in primary prevention programmes'.

Keywords: Hypertension, Pre-eclampsia, Normotensive, Urinary ca/Cr ratio.

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Introduction

Hypertensive disorder of pregnancy are one of the major causes of maternal and fetal mortality and morbidity and it affects almost 3% to 10% of all pregnancies all over the world[1].

Hypertension accounts for about 15 percent of antenatal hospitalization for pregnancy complications in the United States[2] and around 9% in developing countries[3]. Perinatal mortality rate is about 18% in hypertensive disorders[4].

Pregnancy is a physiological process that needs strict monitoring throughout gestational period to circumvent perilous complications like pregnancy induced hypertension (PIH), gestational diabetes etc[5]. Early detection and appropriate management of pregnancy may improve the outcome for both mother and the fetus. Although de novo preeclampsia and preeclampsia superimposed on chronic hypertension are leading causes of maternal and fetal morbidity, gestational hypertension (GH) (i.e. isolated de novo hypertension in the second half of pregnancy) and treated essential hypertension carry a good prognosis for both mother and fetus[6]. Preeclampsia is defined as hypertension after 20 weeks of pregnancy in a woman with edema and proteinuria without previous history of hypertension[7]. Preeclampsia is mainly the disease of the young primigravida, accordingly its incidence is higher in this group. Incidence of preeclampsia is around 2- 8% of all pregnancies, and is a major cause of maternal mortality and morbidity, perinatal mortality and morbidity and premature delivery[8].

*Correspondence

Dr. Usha Rangey Chouhan

Senior Specialist, Department of Obst. & Gynae., Government District Hospital, Sirohi, Rajasthan, India

E-mail: drushapradeepchouhanms@gmail.com

It has been found that nutritional deficiencies are far too common during pregnancy. An inadequate dietary intake before and during pregnancy, has been proved to be a risk factor not only for the mother but also for the fetus. There is evidence that indicates a role for micronutrients supplementation in preventing some pregnancy disorders.

Maternal calcium loss during pregnancy and lactation is a physiological part of fetal and neonatal development. During pregnancy, by increasing intestinal absorption, decreasing renal calcium loss, and reabsorbing some calcium from the maternal skeleton the woman's body undergoes physiological compensation. Renal function changes are seen in symptom free women in whom preeclampsia will finally develop.

Hypocalciuria during pregnancy is known as a risk factor for preeclampsia[9,10]. Low calcium intake has been hypothesized to cause increase in blood pressure by stimulating the release of parathyroid hormone and/or renin which leads to increased intracellular calcium concentration in vascular smooth muscle cells and causes vasoconstriction[11]. A well-known marker of renal function is Urine Creatinine. Excretion of Calcium & Creatinine is affected by the factors which influence functions of the kidneys.

There is hypercalciuria during normal pregnancy, although preeclampsia is associated with hypocalcaemia and low urinary calcium to creatinine ratio. This phenomenon occurs early enough and persists throughout gestation, so it is useful for early identification of patients at risk. The aim of this study to determine the efficacy of urinary calcium to creatinine ratio in a spot urine sample for the prediction of preeclampsia in a asymptomatic pregnant women between 16 to 24 weeks of gestation.

Materials & Methods

A hospital based observational prospective study done on hundred primigravida / multigravida with singleton pregnancy within the gestational age of 16-24 weeks who came for regular ANC to OPD in government district hospital, Sirohi, Rajasthan, India during one year period. A detailed clinical evaluation and relevant laboratory investigations was done for all subjects as per proforma. The relevant cases were followed till delivery.

Exclusion Criteria

- Multiple gestation
- Chronic hypertension
- Diabetes mellitus
- Congenital anomalies
- Thyroid disorders
- Intrauterine fetal death

Methods

Clinical examination of the patient was done. In the first trimester antenatal checkup, height and weight of the cases were recorded as preeclampsia results in excessive weight gain due to water retention. BMI was calculated. Lab investigation was sent for thyroid profile and blood sugar.

In early second trimester checkup, blood pressure was recorded by using sphygmomanometer by palpatory and auscultatory method in both supine and sitting position. Systolic blood pressure was taken by Korotkoff sound (phase I) and diastolic blood pressure was taken by Korotkoff sound (phase V). Spot urine sample was sent for estimation of urine calcium and urine creatinine ratio in those individuals with no comorbid condition. Patient was followed up till term and observed for any rise in the Blood pressure. A blood pressure reading of 140/90 mm Hg and more measured on 2 separate occasions each 6 hours apart, accompanied by proteinuria of at least 300mg per 24hrs, or at least "1+" on dipstick, was considered as preeclampsia. Then, relevant blood investigations like blood urea, creatinine, blood sugar, serum uric acid, liver function test, complete hemogram, and coagulation profile and urine albumin were done.

Method of Collection of Data

Random urine samples are collected from the study subjects as per protocol. The samples were stored in analyzer sample cups at 2-8°C

until analysis. The sample was centrifuged and analyzed using Accucare semi-auto analyzer. The following were analyzed

Urine Calcium Estimation

Urine calcium was measured by photometric color where the chelating agent used was Arsenazo III. The main principle behind this is that Calcium with Arsenazo III (1, 8- Dihydroxy-3, 6-disulpho-2, 7-naphthalene-bis (azo)-dibenzeneearsonic acid), at neutral pH, yields a blue colored complex. The calcium concentration in the sample is proportional to the intensity of the color formed.

Urine Creatinine Estimation

Urine creatinine was measured by photometric color by Accucare semi-automatic analyzer. The main principle behind this is that creatinine reacts with alkaline picrate to produce orange colored complex. Intensity of the color formed during the fixed time is directly proportional to the amount of creatinine present in the sample.

Urine Calcium-to-creatinine ratio (CCR) was calculated, and those with a ratio of less than or equal to 0.04 were considered as test positive[12].

Statistical analysis

IBM SPSS statistics software 22.0 Version was used to analyze the collected data. Descriptive statistics frequency analysis was used to describe about the data, percentage analysis were used for categorical variables and for continuous variables the mean & S.D were used.

Results

Among the 100 women within the study, 17% (17 women) belonged to the age group of 18-20 years. However, 52% (52 women) were in the age group of 21-25 years, 22% (22 women) were in the age group of 26-30 yrs. and 9% of women were above 30 years of age. Amongst the patients who developed preeclampsia 5 women (33.33%) belonged to age group of 18-20yrs, 53.33% (8 women) were between 21 – 25 yrs., 6.66% (3 women) were in the age group of 26-30yrs and 1 patient (6.66%) were above 30yrs of age. It was observed that 47% of the women in the study were primigravida as against to 53% of multiparous women. Amongst the 15 patients who developed preeclampsia 8 patients were primigravida constituting 53.33% whereas 7 women were multigravida constituting 46.66% (table 1).

Table 1: Demographic and biochemical profile of patients

Demographic and biochemical profile	No. of Patients (N=100)	Pre-eclampsia (N=15)
Age group (yrs)		
18-20 yrs	17	5
21-25 yrs	52	8
26-30 yrs	22	1
>30 yrs	9	1
Gravida		
Multi	53	7
Primi	47	8
Urine calcium/Creatinine ratio		
≤0.04	17	15
>0.04	83	0

The incidence of Preeclampsia was 15% and that of normotensive pregnant women was 85%. In the study, out of the 100 patients, 17 women (17% women) had a ratio of \leq 0.04 whereas 83 women (83%) had a ratio $>$ 0.4%. Amongst the patients who had a Urine Ca/Cr ratio $<$ 0.04, 15 women constituting 88.23% developed preeclampsia whereas 2 patients constituting 11.76% remained normotensive at term gestation (table 1).

Among the 100 women within the study, 59% (59 women) had a vaginal delivery, whereas 41% (41 women) underwent LSCS. Amongst the patients who underwent LSCS 6 patients (40%) were preeclamptic whereas 35 patients (41.17%) were normotensive. Amongst the patients who had a vaginal delivery 9 patients (60%) were preeclamptic whereas 50 patients (58.82%) were normotensive. This association was found to be statistically insignificant ($p >$ 0.05) (table 2).

Table 2: Comparison of outcomes in Pre-eclamptic patients in terms of vaginal delivery and cesarean section

Outcome	Total	Pre-eclampsia		P-value
		Yes	No	
LSCS	41	6	35	>0.05
Vaginal delivery	59	9	50	

It was observed that out of the 15 women who developed preeclampsia the mean birth weight of the babies was 2.29kg with a standard deviation of 0.16, whereas among the 85 normotensive pregnant women the mean birth weight of the babies was 2.88kg with a standard deviation of 0.39. This association was found to be statistically very significant (p value<0.05*) (table 3).

Table 3: Comparison of Preeclampsia with birth weight

Preeclampsia	Total	Mean±SD	P-value
Birth weight (kg)			<0.05*
Yes	15	2.29±0.16	
No	85	2.88±0.39	

Discussion

The challenge of any screening test for preeclampsia is to differentiate between those who are and will remain normotensive versus those who appear normal but will develop preeclampsia.

In a prospective study conducted by Rashmi Sinha, Indu Bhushan et al[13] found incidence of pre-eclampsia was 22% and the majority of subjects were of age group 21-30 yrs. (75%) which was consisted with our results. Moni SY, Ahmed KM, Siddika F, Habib A et al concluded that a spot single urine sample calcium/creatinine ratio may be an effective method for screening pregnant women for detection of pre-eclampsia. However they did not compare the occurrence of preeclampsia in different age groups[14].

The association between parity and PIH was not found to be statistically significant. Amongst the 15 patients who developed preeclampsia 8 patients were primigravida constituting 53.33% whereas 7 women were multigravida constituting 46.66%. Sheela CN, Beena SR, et al found 65% were primigravida and the rest were, multigravida[15].

Rani G et al found that when CCR alone is taken as high risk factor for prediction of PIH it was highly significant $P < 0.001$. However, they did not compare the occurrence of preeclampsia according to parity[16].

Urinary calcium excretion decreases in preeclampsia. To determine the predictability of preeclampsia with hypocalciuria, Kaleli B, Özeren M, Turan C et al investigated the calcium to creatinine ratio and the calcium level of a spot urine sample in 56 primigravida patients aged less than 25 years. Of these 56 cases, 44 remained normotensive and preeclampsia developed in 8 cases[17].

Amongst the patients who had a Urine Ca/Cr ratio <0.04 , 15 women constituting 88.23% developed preeclampsia whereas 2 patients constituting 11.76% remained normotensive at term gestation in our study. Akio Izumi HisanoriMinakami et al found that the urine calcium and creatinine ratio was significantly reduced in the 39 women who eventually developed proteinuria and 13 who developed preeclampsia[18].

Swapna VS, Jambale T, Jayaprakash MD et al found that the statistical analysis by unpaired t-test shows that the levels of urinary calcium were significantly decreased in the patients with preeclampsia[19].

To study the predictive value of urinary calcium excretion on occurrence of pregnancy-induced hypertension (PIH) Ye Y, Dai S, Geng X et al conducted a study in which twenty-four-hour urinary calcium excretion, urinary calcium concentration and urinary calcium/creatinine (Ca/Cr) ratio were determined in 184 normal pregnant women and 30 patients with PIH. It was observed that twenty-four-hour urinary calcium excretion, urinary calcium concentration, and Ca/Cr ratio in PIH group were significantly lower than that in normal pregnant group[20]. However both studies did not compare the mode of delivery in the normotensive and preeclamptic group.

Our study showed that birth weight of babies born to preeclamptic women was lower compared to the babies of normotensive women. To compare urinary calcium excretion in pre-eclampsia, gestational hypertension, and chronic hypertension with that in normotensive pregnancies, KristenM. et al[21] conducted a

systematic review and meta-analysis of hypocalciuria in preeclampsia. Twenty-one studies were included. Urinary calcium excretion was lower among women with pre-eclampsia than among those with normotensive pregnancies or chronic hypertension. However this study did not compare the birth weight of babies in the normotensive and preeclamptic group.

Conclusion

We conclude that a pregnant woman with a high risk factor such as nulliparity along with low urinary CCR is at high risk for development of preeclampsia. This test is cheap, simple, inexpensive, noninvasive and easy to perform. Therefore, a single urinary calcium to creatinine ratio may be an effective screening method for impending preeclampsia and may identify population at greater risk to be included in primary prevention programmes.

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Conflict of Interest: Nil Source of support: Nil