Calcium and Phosphate Excretion in Preeclampsia, as Markers of Severe Disease

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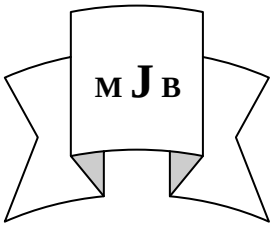

Abstract

Background: Preeclampsia is defined as hypertension of at least 140/90 mmHg recorded on at least two separate occasions and at least 4 hours apart and in presence of at least 300 mg protein in a 24 hour collection of urine, arising *de novo* after 20th week of pregnancy in a previously normotensive woman and resolving completely by the 6th postpartum week, and is often referred to as the disease of theories, and there are more questions than answers and is characterized by alterations in renal function, changes of electrolyte and water metabolism are common findings.

Aim of study: To evaluate the usefulness of urine calcium and phosphorus level as a makers of severity in preeclampsia.

Setting: Our study was carried out at AL-Zahraa Teaching Hospital of Maternity and pediatrics in AL-NAJAF city from April-September-2011 .

Patients and methods:

The total number of all cases was 100, among them 21 women with normal pregnancy , 26 cases with non proteinuric hypertension(whether essential or pregnancy-induced hypertension) , 20 cases with mild preeclampsia , 33 cases with sever preeclampsia .

Every patient was sent for urinary calcium and phosphate which both were determined by the Kramer-Tisdall and phosphomolibidic acid method, respectively.

Data are given as mean ± standard deviation. Student t- test and simple correlation analysis were used as statistical method to test the results .

Results: There was significant decrease in excretion of calcium and phosphorus in sever preeclampsia[$p< 0.001, p< 0.01$] respectively as a result of decrease glomerular filtration rate. While in mild cases and pregnancy induced hypertension cases there were no significant changes.

Conclusion: Urine calcium and phosphorus level are ≥significant determinant of severity of preeclampsia and may be considered as useful marker for predicting the level of renal impairment and time of delivery.

الخلاصة

مرض ما قبل الشنج هو مرض متعدد الاضطرابات خاص بفترة مابعد الاسبوع العشرين من الحمل و يتميز بارتفاع الضغط و وجود البروتين بالادرارحيبت تحدث عدة تغييرات في وظائف الكلى والايض للماء والاملاح في الجسم وهذه الدراسة توضح اهمية نسبة افراز الكالسيوم و الفسفور بالادرار في النساء الحوامل المصابات بمرض ماقبل الشنج، كعلامة لخطورة التلف او الضعف الكلوي وبالتالي تحديد موعد انهاء الحمل.

تم اجراء الدراسة في مستشفى الزهراء التعليمي للنسائية الاطفال في محافظة النجف الاشرف في شهر نيسان ٢٧ /٢٠١١ وكان عدد الحالات ١٠٠ حالة وقد كانت ٢١ حالة حمل طبيعي و٢٦ حالة بوجود ارتفاع ضغط الدم و٢٠ حالة بوجود مرض ماقبل الشنج البسيط و٣٣ حالة بوجود مرض ماقبل الشنج الشديد وقد تم اخذ عينات من ادرار كل مريضة لغرض ايجاد الكالسيوم والفسفور في الادرار وقد تم اجراء عمليات احصائية متعددة للحصول على النتائج وقد وجد ما يلي: هناك نقص ملحوظ في افراز الكالسيوم والفسفور في الحالات الشديدة و عدم وجود تغييرات ملموسة في الحالات البسيطة وحالات ارتفاع ضغط الدم و بالتالي يمكن الاعتماد على نسبة الكالسيوم والفسفور في تحديد شدة الخطورة في الحوامل اللاتي يعانين من هذا المرض لتحديد قصور الكلى ووقت الولادة .

Introduction

Preeclampsia is a multisystem disorder of unknown cause that is unique to human pregnancy. The incidence is reported to be between 2% and 7%, depending on the population[1].Criteria for the Diagnosis of Mild Preeclampsia SBP ≥140 mm Hg and/or DBP≥90 mm Hg on two occasions at least 6 hours apart, typically occurring after 20 weeks gestation and Proteinuria of 300 mg in a 24-hour urine collection or >1+ on two random sample urine dipsticks at least 6 hours apart (no more than 1 week apart)[2].

Sever preeclampsia is characterized by:systolic blood pressure ≥160mmHg or diastolic blood pressure of ≥110 mmHg on bed rest or more, the presence of significant proteinuria, with or without the presence of one of the following complications: Marked protein urea (is defined as 5g or more of protein in 24 hours urine collection), oliguria, Cerebral or visual disturbance, Pulmonary oedema or cyanosis, epigastric or right upper quadrant abdominal pain, Impaired liver function, Thrombocytopenia, Intrauterine growth retardation.Preeclampsia progress along a continuum from mild disease to severe preeclampsia, HELLP syndrome or eclampsia [3,4].

The pathophysiology of preeclampsia reflects alteration in the normal adaptations of pregnancy. Normal physiologic adaptations to pregnancy include increase blood plasma volume vasodilatation, and decreased systemic vascular resistance, elevated cardiac output, and decreased colloid osmotic pressure. The pathologic changes in the endothelial cells of the glomeruli are uniquely characteristic of preeclampsia, particularly in nulliparous women[5]. The main pathogenic factor is not an increase in BP but poor perfusion as a result vasospasm. Arteriolar vasospasm diminishes the diameter of blood vessels, which impedes blood flow to all organs and raises blood pressure function in organs such as the placenta, kidneys, liver and brain is deceased by as much as 40% to 60%.Renal plasma flow increases by 50-70% in pregnancy, and this change is most pronounced in the first 2 trimesters. The increase in renal plasma flow is one of the factors that lead to an increased glomerular filtration rate (GFR), which peaks around the 13th week of pregnancy and can reach levels up to 150% of normal. Therefore, both blood urea nitrogen and creatinine levels, the plasma markers of GFR, are decreased[6]. This decrease has clinical significance in that a normal blood urea nitrogen or creatinine level in a pregnant female may actually indicate underlying renal disease. One of these conditions is related to abnormal renal function and probably decreased urinary calcium excretion. Calcium and phosphate metabolism during normal pregnancy is characterized by minor changes in the serum levels of calcium and phosphate [7]. however urinary calcium and phosphate excretion increases While urinary calcium values in non pregnant women are about 100 - 250 mg/day, in the pregnant women they range between 350 - 620 mg/day [8,9]. Alongside the many studies reporting hypocalciuria in preeclampsia, there are others that have found no such correlation It is unclear whether the decrease of calcium is due to disordered renal function or is a compensatory mechanism in the pathogenesis of preeclampsia. It is concluded that both normal pregnancy and preeclampsia are accompanied by considerable alterations in calcium metabolism, that

parathyroid hormone(PTH) and calcitonin (CT) in both groups are mainly unchanged and at non pregnant level,[10]. and that the increase and decrease in renal calcium excretion in normal pregnancy and preeclampsia, respectively, may be attributed to the changes in kidney function.

Materials and Methods

This prospective study was carried at AL-Zahraa Teaching Hospital of Maternity and Pediatrics in AL-Najaf city-Iraq from April-September-2011. The 100 cases were divided into four groups:21 of normal pregnancy, 26 with hypertension(non proteinuric hypertension whether essential or pregnancy –induced hypertension) and 20 with mild preeclampsia and 33pregnant women with severe preeclampsia.The following inclusion criteria were followed: pre- eclampsia was diagnosed by blood pressure elevation equal to or more than 140/90 ml in combination with proteinuria++ and /or oedma after 20 weeks gestation in previously normotensive non proteinuric patient. Their agesranges from 14 - 44 years, parityranges between (nulliparous- 10), their gestation age from 27 -40 weeks, and a singleton pregnancy. Most of those patient with moderate to severe cases of non proteinuric hypertension, were treated by methyldopa with doses ranging from 750mg-2 grams for varying durations during pregnancy, for those whose blood pressure were fail to be controlled by methyldopa alone, some specialist chose hydralazin tablets in combination with methyldopa. The same was followed for treatment of moderate- severe preeclampsia, although for shorter duration of treatment because of earlier selection of termination of pregnancy.

The exclusion criteria were: Multiple pregnancy, history of essential hypertension,diabetes mellitus, Renal disease, hepatic disease, blood disorder, epilepsy and other medical diseases, and any history of drug intake other than supplement .

Blood pressure was recorded in the study in the sitting position with cuff that is large enough for the subjects arm on at least two occasions 6 hours apart, andKorotkoff phase 5[k5][9] disappearance of sound was used to detect the diastolic blood pressure. Proteinuria was diagnosed by collecting clean catch mid-stream urine sampled in clean dry container then urine protein was determined using the reagent strip [Albustix] reading at 2+[1 gm./l] or more was considered to be positive result for protein urea [significant protein urea] or equal to 1+[0.3gm/l] if the specific gravity is less than 10.30

Severe preeclampsia was defined as a blood pressure of≥ 160/110 mmHg with proteinuria (≥3 +),or the presence of symptoms of imminent eclampsia (headache, blurred vision, Epigastric pain/tenderness with or without the presence of preeclamptic/eclamptic complications(intrauterine growth restriction, elevated hepatic transaminases, low platelets, pulmonary oedema, oliguria, increased serum creatinine, convulsion). Venous blood was obtained after overnight fasting, and 24 hours urine collection, and centrifuged at 2000 g to remove the serum. Serum and urinary creatinine and urea levels were determined by the Jaffe and Kowarski methods and urinary calcium and phosphate were determined by the Kramer-Tisdall and phosphomolibdic acid methods, respectively[8]. Data are given as mean ± standard deviation. Student's t test and simple correlation analysis were used as statistical methods to test the results.

All analyses were performed using commercially available software (SPSS *version 18*). Significant differences of continuous variables were assessed by ONE WAY ANOV. Analysis (*F*-tests , $P \leq 0.01$). Category fetus weight outcome data were assessed by Chi squared(χ^2) test. A P-value ≤0.05 and ≤0.01was considered as statistically significant and highly significant at 1% and 5% respectively .

Results

Table 1 Comparison of Age, GA, Blood Pressure, and Blood parameters (platelets count)

Patient groups	*MEAN ±SD*	*PARAMETERS*				
		AGE(years)	**GA(weeks)**	**SYS. P(mmHg)**	**DIST. P(mmHg)**	**Platelet(x10^9)**
NORMAL	MEAN	27.28±6.14	37.93±1.18	117.4±9.95	76.7±9.13	232±15.0
HYPERTENSIVE	MEAN	27.93±6.387	37.09±1.94	155.0±15.56	96.9±9.70	227±18.1
MILD PRECLAMPSIA	MEAN	26.20±6.67	35.45±2.71	160.2±11.71	103.5±6.71	192±15.1
SEVER PRECLAMPSIA	MEAN	24.64±5.968	33.42±3.01	179.7±16.29	114.5±7.65	125±30.2
P value		0.067NS	0.000**	0.000**	0.000**	0.000**

- No significant differences were observed in the age of different groups.
- Lower gestational age (P<0.01) was recorded in the sever preeclamptic patients.
- Systolic and diastolic blood related significantly (P<0.01) with severity of preeclampsia.
- platelet count related significantly(P<0.01) with severity of preeclampsia.

Table 2 Renal function test.

Patient groups	*MEAN*	**BLOOD UREA mg/dl**	**S.CREAT mg/dl**	**S.URIC Acid**
NORMAL	MEAN	27.238	0.695	4.774
HYPERTENSIVE	MEAN	31.375	0.873	4.442
MILD PRECLAMPSIA	MEAN	32.000	0.868	5.409
SEVER PRECLAMPSIA	MEAN	39.929	1.111	6.474
P value		0.000**	0.001**	0.000**

All studied blood parameters showed significant relation with severity of preeclampsia. Blood urea, Serum creatinine and Serum uric acid were higher (P<0.01) in the sever preeclamptic women in comparison with other groups.

Table 3 Liver function test of studied groups.

Patient groups	*Liver function Test (mean +_SD)*			
	GPT(IU/L)	GOT(IU/L)	S.A.P.(IU/L)	T.S.B(mg/dl)
NORMAL(	5.762±1.97	14.333±9.9	175.143±23	0.761±0.104
HYPERTE	7.346±3.24	15.462±8.3	186.192±15	0.650±0.176
MILD	9.200±3.76	23.400±11.	187.200±23	0.780±0.278
SEVER	9.945±3.15	33.009±13.	205.091±92	1.185±1.051
P value	0.000**	0.000**	0.014*	0.000**

***significant at P≤0.05, **significant at P≤0.01, using ANOVA, values are presented as mean ±SD ; NS, not significant.**

Sever preclampsia women showed a higher value of GPT and GOT than other studied groups (P<0.01) and(P<0.05) respectively . The higher value GPT and GOT strongly significant in sever preeclampsia and Serum Alkaline Phosphatase (S.A.P.) showed significant differences between studied groups.

Table 4 Urine analysis of studied groups**.**

Patient groups		**Urinary calcium(mg/dl)**	**Urinary phosphorous(mg/dl)**	**Urinarycreatinine (mg/dl)**
NORMAL	mean	218.20	***1.03***	1.705
(21)	SD	32.045	0.152	0.784
HYPERTENSIVE	mean	198.92	0.892	1.431
(26)	SD	39.912	0.256	0.178
MILD PRECLAMPSIA	mean	110.28	0.439	0.972
(20)	SD	20.625	0.109	0.113
SEVER PRECLAMPSIA	**mean**	98.35	0.394	0.906
(33)	SD	15.597	0.147	0.125
P value		0.000**	0.00**	0.000**

Significant differences were observed in the urine calcium of different studied groups . In coclusion strong relation was obseved between decreasing of urine calcium and severity of preclampsia.

Urine phosphorus and creatinine showed significant differences between

studied groups . Mild and sever preclamptic womens showed lower value (P<0.01) of urine phosphorus and creatinine than other groups .

Table 5 Comparison of complication in the four studied groups.

Complications	NORMAL	HYPERTENSIVE	MILD PRECLAMPSIA	SEVER PRECLAMPSIA
Eclampsia	0	0	0	15(45.45%)
IUGR	0	0	0	7(21.21%)
HELLP syndrome	0	0	0	3(9.09%)
Pulmonary edema	0	0	0	2(6.06%)
Uncontrolled BP	0	2(23.07%)	0	2(6.06%)
placental abruption	0	0	1(5%)	3(9.09%)
sever oligohydramnios	0	0	0	2(6.06%)
fetal distress	0	1(3.84%)	1(5%)	0
% C/S	3(14.28)	9(34.61)	13(65)	28(84.84)

Of 39 cases of complications ,34(60.71%) cases occurred in the sever preeclampsic women and 2 (3.88%) cases in the mild preeclampsic women.

84.84% of sever preeclampsic women deliverd by C/S in comparison with 65% 0f mild preeclampsia and 34.61% of Hypertensive and only 14.28% in normal women.

Table 6 Comparison of Fetal weight (gm.)outcome

Groups	*Fetal Weight (kg)*	*G.A(weeks)*
Normal	***3.195±0.378***	**37.93±1.18**
Hypertensive	***2.950*±0.295**	**37.09±1.94**
Mild	***2.585*±0.387**	**35.45±2.71**
Sever	***2.115*±0.533**	**33.42±3.01**
P-value	***0.000*****	**0.000****

Sever preclampsia women have got lower fetal weight in comparison with other women, for a given gestational age. decreasing of fetal weight related significantly (P<0.01) with severity of preclampsia.

Discussion

Pre-eclampsia is often referred to as the disease of theories, and there is more questions than answers. Preeclampsia, is a vascular disorder of pregnancy and is a leading cause of maternal morbidity as well as perinatal morbidity and mortality. In our study table [1] we found lower gestational age was recorded in the sever preeclamptic patients these finding similar to study done by Gerther JM, Coustan DR, Kliger AS, Mallette, LE, Ravin N [13].

In normal pregnancy renal blood flow and filtration rate increase[8] while in Preeclampsia the renal blood flow and glomerular filtration rate decrease with development of toxemia [12],in our study table[2] the blood urea, serum creatinine and serum uric acid were higher in sever preeclamptic women in comparison with other group which result in the decrease of urea and creatinine excretion .These finding similar to study done by Hayachi T[13] he found the serum level of urea, creatinine and uric acid are elevated with sever preeclampsia [p<0.001 ,p< 0.01 ,p<0.01] respectively.

In our study [table 3] the sever preeclamptic women showed a higher value of GPT and GOT than other group [p<0.01] and [p<0.05] respectively. The higher value of GPT and GOT are strongly significant with the severity of preeclampsia, also serum Alkaline phosphatase showed significant differences between studied groups. these finding similar to study done by Taylor RN, Music TJ, Kuhn RM, Robert JM[2].

Renal excretion of calcium and phosphate increases during pregnancy due to increase in renal filtration rate[8]. Excretion usually increases during each trimester, with maximum level reached during the third trimester. Alteration of phosphate and most notably calcium excretion are common finding of hypertension and some renal disorder in general. There is a decrease in urinary calcium in preeclampsia [14] . Our finding [table 4] in preeclampsia confirm the result of sanchez-Rames , Yoshida and Tanfield [14-15] he found decrease level of calcium and phosphorus [p<0.001 , p<0.01] respectively .The reason for hypercalciuria in pregnancy is probably the increased glomerular filtration rate [15]. Pedersen et al. reported that fractional excretion of calcium in preeclampsia pregnant women was lower in the third trimester than it was in normotensive pregnant women [16]. Because parathyroid hormone and calcitonon

level were not altered in the patient with preeclampsia. It was concluded that decrease renal filtration rate and increase tubular reabsorption of calcium and phosphate may result in hypocalciuuria and hypophosphateuria in toxemia .In conclusion strong relation was observed between decreasing of urine calcium and severity of preeclampsia.

In table [5] we found most of the complication occurred in the sever preeclamptic women [e.g. eclampsia, HELLP syndrome, pulmonary oedema ,IUGR, sever oligohydramnios] these finding similar to many study e.g. Sibai BM, Ramadan MK, Chari RS, et al.

Also in our study table[6] the sever preeclamptic women have got lower fetal weight than other women .Decrease fetal weight related significantly with severity of preeclampsia.

Lower calcium excretion may result from dietary variation. All participants in our study were on a free range diet. Because we did not advise any of our patient to alter their diets, however, we believe it is unlikely that dietary calcium intake played an important role in our findings. As a conclusion, hypocalciuria and hypophosphateuria are important features of severe preeclampsia and probably are indirectly related to the altered renal function seen in toxemia of pregnancy.

Conclusion and Recommendation

Urine calcium and phosphorus level could be a significant determinant of severity of preeclampsia and might be considered as useful markers of predicting the level of renal impairment and thereby the time of delivery.

Further studies for larger number of patients may be needed to confirm our results, as it is a simple test and can be used as a routine predictive and diagnostic test for the severity of preeclampsia.

References

1. Wang Y, Walsh SW, Kay HH. Placental lipid peroxides and thromboxane are increased and prostacyclin is decreased in women with preeclampsia. Am J Obstet Gynecol 167:946-9, 2010.
2. Taylor RN, Musci TJ, Kuhn RM Roberts JM. Partial characterization of a novel growth factor from the blood of women with preeclampsia. J Clin Endocrinol Metab 70: 1285-91, 2011.
3. Institute of Medicine, Food and Nutrition Board. Dietary Reference Intakes: Calcium, Phosphorus, Magnesium, Vitamin D, and Fluoride. Washington, DC: National Academy Press, p146-189, 2011.
4. Bringhurst, F.R. et al. Hormones and disorders of mineral metabolism. In Wilson, J.D. et al Eds. Williams Textbook of Endocrinology, 9th Edition. Philadelphia: W.B. Saunders Company, p 1155-1210, 2011.
5. National Research Council, Food and Nutrition Board. Recommended Dietary Allowances, 10th Edition. Washington, DC: National Academy Press,: p 184-187, 2007.
6. Minerals. In Drug Facts and Comparisons. St. Louis, MO: Facts and Comparisons,: pages 27-51, 2000.
7. Silverman, H. et al. The Vitamin Book, 2nd edition. New York: Bantam Books, p 255-265, 2010
8. Mozdzien G, Schinninger M, Zazgornik J. Kidney function and electrolyte metabolism in healthy pregnant women. Wien. Med. Wochensch,2011.
9. Maikranz P, Holley JL, Parks JH, Lindheimer MD, Naragawa Y, Coe FL. Gestational hypercalciuria causes pathological urine calcium late super

saturation. Kidney Int 36: 108-13, 2009.
10. Samadi AR, Mayberry RM, Zaidi AA, et al. Maternal hypertension and associated pregnancy complications among African-American and other women in the United States. Obstet Gynecol;87:557-563, 2011.
11. Gerther JM, Coustan DR, Kliger AS, Mallette LE, Ravin N. Pregnancy as state of physiologic absorptive hypercalciuria. Am J Med 81: 451-5,2011.
12. Seligman SP. The role of nitric oxide in the pathogenesis of preeclampsia. Am J Obstet Gynecol 17(4): 944-8, 2010
13. Hayashi T: Uric acid and endogenous creatinine clearance studies in normal pregnancy and toxemia of pregnancy. Am J Obstet Gynecol**71**:859-870, 1956.
14. Taufield PA, Ales KL, Resnick LM Druzin ML, Gerther JM, Laragh JH. Hypocalciuria in preeclampsia. N Engl J Med 316 (12): 715-8, 2011.
15. Sanchez-Ramos L, Sandroni S, Andres FJ, Kaunitz AM. Calcium excretion in preeclampsia. Obstet. Gynecol 77:510-3, 2010.
16. Pedersen EB, Johannesen P, KristensenS. Calcium, parathyroid hormone andCalcitonin in normal pregnancy and preeclampsia. Gynecol Obstet Invest18: 156-64, 2009.