
OBSTETRICS

The accuracy of using random urinary protein-to-creatinine ratio for prediction of significant proteinuria for diagnosis of preeclampsia in hypertensive pregnancies

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ABSTRACT

Objective To evaluate the accuracy of random urinary protein-to-creatinine ratio for prediction of significant proteinuria for diagnosis of preeclampsia in hypertensive pregnancies.

Material and method Regardless of the urinary protein dipstick determinations, women who had hypertension after 20 weeks of gestation, admitted in obstetric ward, Chonburi Hospital, were studied prospectively. Random mid-stream urine specimen were obtained for protein-to-creatinine ratio determination before initiation of a 24-hour urine collection. With the criterion of 24-hour proteinuria of at least 300 mg as a significant proteinuria, the sensitivity, specificity and accuracy of a random urinary protein-to-creatinine ratio of > 0.25 for prediction of significant proteinuria were analyzed and a receiver operating characteristic curve was constructed to determine the optimal cutoff value for prediction of significant proteinuria for diagnosis of preeclampsia in hypertensive pregnancies.

Results Sixty-seven patients completed the study. Forty-five percent of the study population had significant proteinuria. The optimal cutoff value is 0.25 which yielded sensitivity, specificity and accuracy of 90.0%, 75.9% and 85.1% respectively. Three patients who had false-negative test results were mild preeclampsia. Seven false-positive test results had 24-hour urine protein levels that ranged from 134 to 288.2 mg.

Conclusion In hospitalized patient, the random urinary protein-to-creatinine at a cutoff of > 0.25 revealed a high accurate prediction of significant proteinuria for diagnosis of preeclampsia in hypertensive pregnant women. This test could be useful in the screening, assessment, and follow-up of proteinuria in hypertensive pregnancy and avoidance of the problems of associated with 24-hour urine collection.

Keywords: Protein-to-creatinine ratio, Proteinuria, Preeclampsia

Introduction

Hypertensive disorders occur in 12% to 22% of pregnancies and contribute significantly to maternal and neonatal morbidity and mortality⁽¹⁾. The development of proteinuria is an important sign of preeclampsia and is associated with a high rate of maternal and fetal complications among hypertensive pregnancies^(2,3). The “gold standard” definition of significant proteinuria for diagnosis of preeclampsia is based on total protein excretion in 24 hours and is most commonly accepted as > 300 mg per 24 hours⁽⁴⁾. However, the 24-hour urine collection is a time-consuming and cumbersome process leading to delay diagnosis and inaccurate result from incomplete collection of the specimen^(5,6). A rapid and reliable method for diagnosis of significant proteinuria could assist clinicians to make decisions regarding delivery and to use magnesium sulfate for seizure prophylaxis. A quicker and easier method to evaluate the quantity of urinary protein is the urinary dipstick. But studies showed that this test was a poor predictor of 24-hour urine total protein level⁽⁷⁾ and one study showed that 66% of the patients with negative or trace protein on dipstick had significant proteinuria of ≥ 300 mg per 24 hours⁽⁸⁾.

Many studies in non pregnant women showed that random urinary protein-to-creatinine ratio could reliably correspond with the 24-hour urine protein and a ratio of less than 0.20 reflected insignificant proteinuria⁽⁹⁻¹¹⁾. Several studies have found a strong linear correlation between the random urinary protein-to-creatinine ratio and the 24-hour urine protein in hypertensive pregnant women^(5,6,12-14). One prospective study in Thai population demonstrated highly accurate prediction of significant proteinuria when the random urinary protein-to-creatinine ratio at a cutoff of ≥ 0.25 ⁽¹⁵⁾. However, this study included only the pregnant women who had hypertension and had urine protein $\geq 1+$ by dipstick. But the previous studies revealed that dipstick urinalysis had a significant false negative rate⁽¹⁶⁾ and a dipstick of negative to trace should not be used to rule out significant proteinuria⁽⁸⁾. Therefore this study might miss preeclamptic women and yielded a limitation in

clinical application. So the present study was designed to evaluate the accuracy of the random urinary protein-to-creatinine ratio in prediction of significant proteinuria for diagnosis of preeclampsia by including hypertensive pregnant women, regardless of the urinary protein dipstick determinations and using 24-hour urine protein as a gold standard.

Materials and Methods

The study was performed at Chonburi hospital in Chonburi, a tertiary care center, Thailand, between March 1, 2006 and March 30, 2007, with institutional ethical board approval. Pregnant women who were admitted in the obstetric ward with a suspicion of preeclampsia were studied prospectively. The inclusion criteria were : (1) pregnant women who had a sustained blood pressure of either ≥ 140 mmHg systolic or ≥ 90 mmHg diastolic after 20 weeks' gestation with previously normal blood pressure ; or (2) pregnant women who had chronic hypertension without proteinuria before 20 weeks' gestation and had ≥ 1 of following clinical finding : a new-onset urine protein $\geq 1+$ by dipstick or a sudden increase blood pressure.

Preeclampsia is categorized as mild, severe and superimposed preeclampsia upon chronic hypertension. Mild preeclampsia is defined as the presence of blood pressure $\geq 140/90$ mmHg and proteinuria ≥ 300 mg/24 hours or $\geq 1+$ dipstick in over 20 weeks' gestation⁽¹⁷⁾. Severe preeclampsia is defined as having one or more of the following criteria : blood pressure of at least 160/110 mmHg , proteinuria of at least 2 g per 24 hours, or at least 2+ on dipstick testing, cerebral or visual disturbance, epigastric pain, elevated ALT or AST, thrombocytopenia, microangiopathic hemolysis, serum creatinine >1.2 mg/dl unless known to be previously elevated⁽¹⁷⁾. Superimposed preeclampsia upon chronic hypertension in the present study was determined by evidence of new onset proteinuria ≥ 300 mg/24 hours in hypertensive women but no proteinuria before 20 weeks' gestation⁽¹⁷⁾.

Women with underlying primary/secondary

renal disease or urinary tract infection were excluded. To participate in the study, the patients had to give written informed consent. Each eligible participant was asked to collect a random mid-stream urine sample in daytime and before bedtime (except the first voided morning urine) for protein-to-creatinine ratio determination. Then they were advised for bed rest and instructed to collect urine for 24 hours. The volume of the 24-hour urine was recorded and 10-ml aliquot of thoroughly mixed urine were removed for measurement of protein and creatinine concentration and excretion. All urine specimens were collected without preservatives, refrigerated at 4°C and analysed in the next day. The subjects who could not completely collect a 24-hour urine sample were excluded. The 24-hour urine specimens were considered inadequate and excluded from this analysis if total creatinine excretion were < 10 mg/kg of prepregnancy weight per day because the urine creatinine provides a rough estimation of the completeness of a timed urine collection⁽¹⁸⁻²⁰⁾.

Proteinuria on 24-hour urine collection was defined as “significant” (≥ 300 mg) or “severe” ($\geq 2,000$ mg), and mild proteinuria was defined as 300 to 1,999 mg. Urinary protein quantitation was determined by the dyne-binding colorimetric method which utilized pyrogallol red-molybdate complex⁽²¹⁾, and urine creatinine level was determined by the Jaffy rate method⁽²²⁾. All tests were done on the Au 640® Clinical System (Japan) by the laboratory service of Chonburi Hospital.

Statistical analysis was performed with the SPSS® statistical package version 11.5. Descriptive statistic were used for demographic and baseline data and summarized as mean/median or percent. To evaluate the correlation between the random urinary protein-to-creatinine ratio and the 24-hour urine protein excretion, a simple linear regression with calculation of a Pearson correlation coefficient (r) was used. The accuracy of a random urinary protein-to-creatinine ratio for detection of significant proteinuria was evaluated with the use of the 24-hour urine protein as the gold standard test. The

sensitivity, specificity, positive and negative predictive values were analyzed by using a random urinary protein-to-creatinine ratio more than or equal to 0.25 as a cutoff⁽¹⁵⁾, as well as with other cutoff point. A receiver operating characteristic (ROC) curve was then constructed to determine the optimal cutoff and the area under the curve was calculated. A prior sample size calculation indicated 66 subjects required for constructing 95 percent confidence interval with maximum allowable error 0.10 at a sensitivity of 0.96 and specificity of 0.92⁽²³⁾.

Results

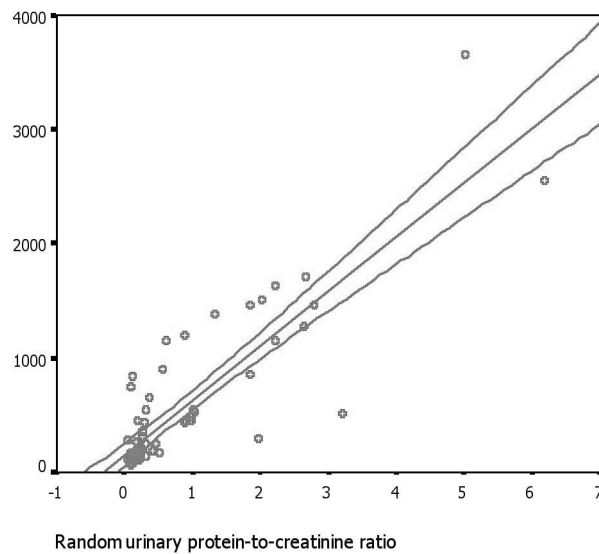
Seventy-six pregnant patients were recruited. Nine patients initially enrolled were excluded in the final analysis. Of these 9 patients, 5 patients were excluded because of their 24-hour urine creatinine excretion were < 10 mg/kg/day, and 4 patients were delivered before the completion of the 24-hour urine collection, leaving 67 women in the study.

The maternal age ranged from 21 to 43 years (mean \pm SD = 30.0 \pm 6.23 years). The gestational age at recruitment ranged from 23 to 39 weeks (mean \pm SD = 33.3 \pm 5.03 weeks). Eighty-seven percent (58/67) of the patients were in the third trimester and only 13.4 % (9/67) in the second trimester. Twenty-nine patients (43.3%) were nulliparous. Sixty-five patients had singleton and two patients had twin pregnancies. Diagnosed by clinical finding (eg. elevated blood pressure, persistent headache) and urine dipstick, the majority of patients (41 in 67cases) had gestational hypertension, 15 patients had mild preeclampsia, 6 patients had severe preeclampsia and 5 patients had superimposed preeclampsia.

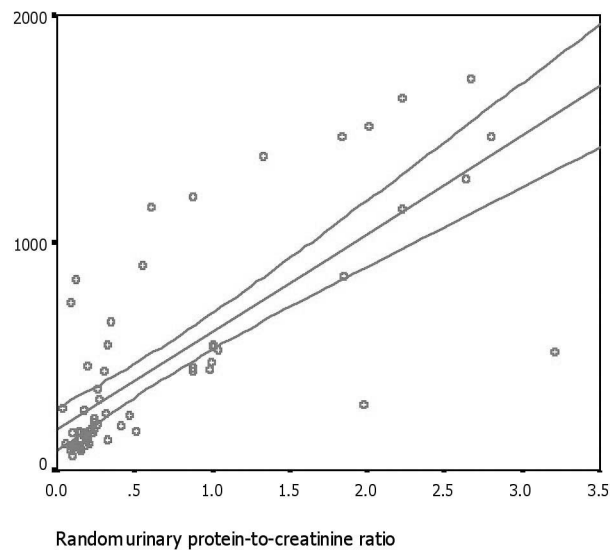
The mean 24-h urine protein excretion was 539.5 mg and the mean random urinary protein-to-creatinine ratio was 0.80. Forty-five percent (30/67) of the patients had significant proteinuria as determined by the 24-hour urine collection and 3% (2/67) had a 24-hour urine excretion ≥ 2 grams. The random urinary protein-to-creatinine ratio was strongly correlated with 24-hour urine excretion with a correlation coefficient of 0.859 (Fig. 1A). The correlation coefficient was reduced from 0.859 to

0.762 if 2 cases with 24-hour proteinuria of more than 2 grams were not included. However, there was no statistically significance difference, when

comparing this value to the correlation coefficient with all the participants included (Fig. 1B)



A.



B.

Fig. 1 Correlation between 24-hour protein excretion and the random urinary protein-to-creatinine ratio. **A.** All participants ($n = 67$, $r = 0.859$). **B.** Cases of 24-hour urine protein of less than 2 grams ($n = 65$, $r = 0.762$). Each circle represents 1 patient. The middle line indicates the regression; the upper and lower line indicate the 95% CI.

The ROC curve for prediction of significant proteinuria by random urinary protein-to-creatinine ratio is shown in Figure 2. The area under the ROC curve is 0.877. The sensitivity, specificity, PPV and NPV and 95% confidence intervals for various cutoffs are shown in Table 1. The optimal cutoff is 0.25 which yielded a sensitivity of 90.0%, specificity of 75.9%, PPV of 79.4%, NPV of 90.0%, positive likelihood ratio of 4.76, negative likelihood ratio of 0.12 and accuracy of 85.1%. With the use of this cutoff, there were three cases of false-negative test

results and seven cases of false-positive test results. The cases with the false-negative test results had 24-hour urine protein of 456, 739.7 and 839 mg. The cases with the false-positive test results had 24-hour urine protein that ranged from 134 to 288.2 mg.

ROC curve analysis and the optimal cutoff in a subgroup of 24-hour urine protein level of less than 2 grams were also calculated. The area under the curve and cutoff value were 0.868 and 0.25 respectively, not different from the values of all women included in the study.

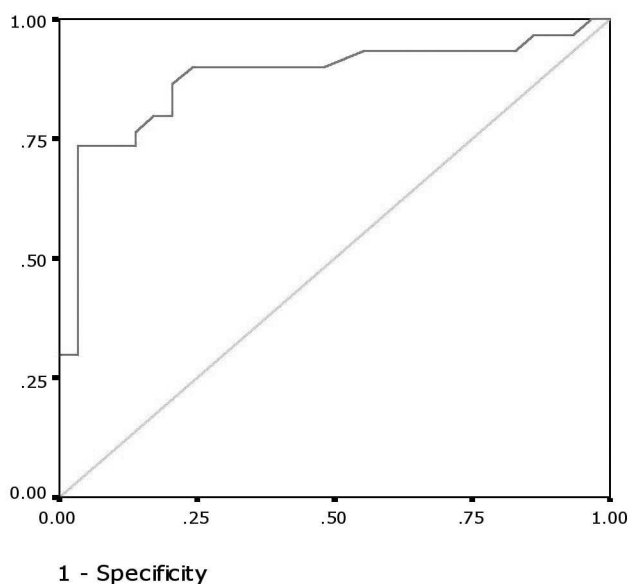


Fig. 2 ROC curve for various cutoffs for the random protein-to-creatinine ratio as a predictor of significant proteinuria.

Table 1 Characteristics of the random urinary protein-to-creatinine ratio for the detection of significant proteinuria with the use of various cutoffs

Cutoff	Sensitivity % (95%CI)	Specificity % (95%CI)	PPV % (95%CI)	NPV % (95%CI)
0.185	93.3 (87.3, 99.3)	44.8 (32.9, 56.7)	62.2 (50.6, 73.8)	90.9 (84.0, 97.8)
0.200	90.0 (82.8, 97.2)	51.7 (39.7, 63.7)	64.3 (52.8, 75.8)	88.0 (80.2, 95.8)
0.220	90.0 (82.8, 97.2)	58.6 (46.8, 70.4)	67.5 (56.3, 78.7)	88.8 (81.2, 96.4)
0.235	90.0 (82.8, 97.2)	62.1 (50.5, 73.7)	71.1 (60.2, 82.0)	89.7 (82.4, 97.0)
0.250	90.0 (82.8, 97.2)	75.9 (65.7, 86.1)	79.4 (69.7, 89.1)	90.9 (84.0, 97.8)
0.265	86.7 (78.6, 94.8)	79.3 (69.6, 89.0)	81.3 (72.0, 90.6)	88.6 (81.0, 96.2)
0.285	83.3 (74.4, 92.2)	79.3 (69.6, 89.0)	80.6 (71.4, 90.0)	86.1 (77.8, 94.4)
0.305	80.0 (70.4, 89.6)	79.3 (69.6, 89.0)	80.0 (70.4, 89.6)	83.8 (75.0, 92.6)
0.315	80.0 (70.4, 89.6)	82.8 (73.8, 91.8)	82.8 (73.8, 91.8)	84.2 (75.5, 92.9)

PPV = Positive predictive value ; NPV = Negative predictive value

Discussion

This study demonstrated a strong correlation between the random urinary protein-to-creatinine ratio and the quantitation of 24-hour proteinuria in hypertensive pregnant women. A random urinary protein-to-creatinine ratio was hypothesized to be preferable to a random protein alone because the ratio of two stable excretion rates, creatinine and protein, would cancel out the time factor and thus provide a better estimate of 24-hour protein excretion⁽⁹⁾.

The present study suggested that the random urinary protein-to-creatinine ratio is a high accurate test for prediction of significant proteinuria in hypertensive pregnancies as demonstrated by an area under the ROC curve of 0.877. In determining what the best cutoff was, the cutoff should be dependent on the implication of the test⁽²⁴⁾. Therefore, the authors believed that it was important to maximize sensitivity, given the potential consequences of missing the diagnosis of preeclampsia. A cutoff value of ≤ 0.185 had the sensitivity of 93.3% but the specificity was only 44.8%. To maximize the specificity while maintaining a sensitivity of $\geq 90\%$, the optimal cutoff of ≥ 0.25 was set in the population (in which the prevalence of preeclampsia was 45%). This yielded a sensitivity, specificity, positive and negative predictive values of 90.0%, 75.9%, 79.4% and 90.9%, respectively. Because the present study included the hypertensive pregnant women even they had negative urine protein by dipstick, therefore most participants were gestational hypertension.

For twenty-six patients (26/67) who were initially diagnosed as preeclampsia by clinical finding and urine dipstick, the optimal cutoff value for prediction of significant proteinuria was 0.53 which had a poor negative predictive value (42.6%), but it had a high positive predictive value (94.7%), with a sensitivity of 75% and a specificity of 81%.

The recent prospective study from Yamasmit, et al.⁽¹⁵⁾ showed an excellent correlation and a high accuracy to use the random urinary protein-to-creatinine ratio for discriminating between

insignificant and significant proteinuria by using the same definition of significant proteinuria as the present study. The different target population may explain the more correlation in Yamasmit's study than found in the present study because urinary protein excretion is dependent on body-surface area⁽²⁵⁾ and, creatinine production and excretion are also related to body size^(18,20). Thus it is possible that the good correlation expected between the urinary protein-to-creatinine ratio and quantitative protein excretion in individual patients may also hold for groups of patients. The prevalence of significant proteinuria in Yamasmit's study was higher than those in the present study because they recruited the hypertensive pregnant women who had to have urine protein $\geq 1+$ by dipstick. Many studies revealed that dipstick urinalysis had a significant false-negative rate^(8,16), thus the use of this test for recruitment may lose the preeclamptic women. However, using the same optimal cutoff of ≥ 0.25 , the accuracy, the sensitivity and the specificity in Yamasmit's study were higher than those in the present study. The lower prevalence of preeclampsia in the present population (45% versus 69% in Yamasmit's study) may explain the lower PPV found in the present study⁽²⁶⁾.

The retrospective study of Rodriguez-Thompson, et al.⁽⁶⁾ reviewing the medical records of 138 women who completed both a random urinary protein-to-creatinine ratio and a 24-hour urine collection for evaluation of preeclampsia, revealed the correlation coefficient value ($r = 0.80$) and the area under the ROC curve (0.923) in Rodriguez-Thompson's study were as high as the results of our study and suggested that the best cutoff ≥ 0.19 that yielded the sensitivity of 90% and the specificity of 70% (in which the prevalence of significant proteinuria was 55%). Although the present optimal cutoff seemed to be higher than Rodriguez-Thompson's study, the performance of the test was actually not different by 95%CI comparison.

Only two subjects of the present study had severe proteinuria (these were diagnosed as severe preeclampsia), thus it was unable to assess the

cutoff value of the random urinary protein-to-creatinine ratio for the diagnosis of severe proteinuria. It would be difficult to enroll an adequate number of the participants for this analysis because severe proteinuria is low prevalence and the participants usually delivered before completion of the 24-hour urine collection.

The present study was designed to collect the random urine samples before the 24-hour urine because it most closely resembles how the test would be used in practice and the results of random urine specimen can be altered if patients remain at bed rest during the collecting period. The random urine samples in this study were obtained during normal daylight activity and before bedtime, except the first voided morning specimen because protein excretion is lower during the period of recumbency than during the active daytime hours⁽⁹⁾.

Clinician must know the approximate prevalence of the condition of interest in the population being tested before the interpretation of any diagnostic test because positive and negative predictive values depend on the prevalence. So very good test would have poor predictive value positive when applied to low-prevalence populations⁽²⁴⁾. For patients with a high pretest probability of disease and a negative random urinary protein-to-creatinine ratio, repeating the test or proceeding with collection of a 24-hour urine is a reasonable option.

The present study showed that the random urinary protein-to-creatinine ratio is a rapid and reliable test, so this test was suggested for the purpose of screening, assessment or follow-up of proteinuria in the hypertensive pregnant women instead of relying on semiquantitative dipstick determination.

The present data supported the use of random urinary protein-to-creatinine ratio in hospitalized pregnant patients to predict the 24-hour urine protein excretion. In order to apply this test to use in ambulatory patients, the future study should perform in outpatient basis. Furthermore, the future effort should be focused on the evaluation of clinical outcome when using this test for follow-up of

proteinuria in the hypertensive pregnant women. In addition, the accuracy of the test in combination of random urinary protein-to-creatinine ratio and dipstick urinalysis as the screening test for prediction of preeclampsia in hypertensive pregnancy should be prospectively evaluated because this approach results in higher sensitivity than would arise with either test used alone.

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ความแม่นยำของการใช้อัตราส่วนโปรตีนต่อครีอาตินีนในปัสสาวะที่เก็บแบบสุ่มเพื่อทำนายปริมาณโปรตีนในปัสสาวะในสตรีตั้งครรภ์ที่มีความดันโลหิตสูงชนิดครรภ์เป็นพิษ

ลินดา เมธาวรณพงศ์, อีระ ศิวดุลย์*

วัตถุประสงค์ : เพื่อศึกษาความแม่นยำในการทำนายปริมาณโปรตีนในปัสสาวะเพื่อวินิจฉัยภาวะครรภ์เป็นพิษในสตรีตั้งครรภ์ที่มีความดันโลหิตสูงโดยใช้อัตราส่วนโปรตีนต่อครีอาตินีนในปัสสาวะที่เก็บแบบสุ่ม เปรียบเทียบกับการวัดปริมาณโปรตีนในปัสสาวะที่เก็บ 24 ชั่วโมงซึ่งเป็นวิธีมาตรฐาน

วัสดุและวิธีการ : สตรีตั้งครรภ์ที่มีความดันโลหิตสูงหลังอายุครรภ์ 20 สัปดาห์ที่เข้ารับการรักษาในหอผู้ป่วยสูติกรรม โรงพยาบาลชลบุรี เนื่องจากสงสัย ภาวะครรภ์เป็นพิษ ได้รับการคัดเลือกเข้าศึกษา (โดยไม่ได้คำนึงถึงผลการตรวจโปรตีนในปัสสาวะด้วยวิธี dipstick) โดยให้ผู้ป่วยเก็บปัสสาวะแบบสุ่มเพื่อนำไปประเมินอัตราส่วนโปรตีนต่อครีอาตินีน และเก็บปัสสาวะ 24 ชั่วโมงเพื่อตรวจวัดปริมาณโปรตีน ซึ่งการวินิจฉัยภาวะครรภ์เป็นพิษถือตามปริมาณโปรตีนอย่างน้อย 300 มิลลิกรัมในปัสสาวะ 24 ชั่วโมงเป็นมาตรฐาน จากนั้นจึงวิเคราะห์หาความไว ความจำเพาะและความถูกต้องของการใช้อัตราส่วนโปรตีนต่อครีอาตินีนในปัสสาวะที่เก็บแบบสุ่มที่มากกว่าหรือเท่ากับ 0.25 และหาอัตราส่วนโปรตีนต่อครีอาตินีนที่เหมาะสมในการทำนายปริมาณโปรตีนในปัสสาวะในสตรีตั้งครรภ์ที่มีความดันโลหิตสูงชนิดครรภ์เป็นพิษ

ผลการวิจัย : ผู้ป่วยจำนวน 67 รายได้รับการคัดเลือกและอยู่จนจบการศึกษา โดยผู้ป่วยร้อยละ 45 ได้รับการวินิจฉัยว่ามีภาวะครรภ์เป็นพิษ อัตราส่วนโปรตีนต่อครีอาตินีนในปัสสาวะที่เก็บแบบสุ่มที่เหมาะสมในการทำนายภาวะครรภ์เป็นพิษคือมากกว่าหรือเท่ากับ 0.25 โดยมีความไวร้อยละ 90, ความจำเพาะร้อยละ 75.9 และค่าความแม่นยำร้อยละ 85.1 เมื่อใช้จุดตัดนี้ ผู้ป่วย 3 รายที่มีผลลบลงได้รับการวินิจฉัยหลังจากเก็บปัสสาวะครบ 24 ชั่วโมงว่าเป็นครรภ์เป็นพิษชนิดที่ไม่รุนแรง และผู้ป่วย 7 รายที่มีผลบวกลง มีปริมาณโปรตีนในปัสสาวะที่เก็บ 24 ชั่วโมงอยู่ระหว่าง 134 มิลลิกรัมถึง 288.2 มิลลิกรัม

สรุป : อัตราส่วนโปรตีนต่อครีอาตินีนในปัสสาวะที่เก็บแบบสุ่มที่มากกว่าหรือเท่ากับ 0.25 สามารถทำนายปริมาณโปรตีนในปัสสาวะเพื่อวินิจฉัยภาวะครรภ์เป็นพิษได้อย่างแม่นยำในสตรีตั้งครรภ์ที่มีความดันโลหิตสูงที่เข้ารับกษาตัวในโรงพยาบาล และเป็นอีกทางเลือกหนึ่งในการตรวจคัดกรอง ประเมินและติดตามปริมาณโปรตีนในปัสสาวะในสตรีตั้งครรภ์ที่มีความดันโลหิตสูงและหลีกเลี่ยงปัญหาที่เกิดจากการเก็บปัสสาวะ 24 ชั่วโมง

คำสำคัญ : อัตราส่วนโปรตีนต่อครีอาตินีนในปัสสาวะ, โปรตีนในปัสสาวะ, ภาวะครรภ์เป็นพิษ
