Evaluation of Chlorhexidine in the Detection of Bacteriuria in Pregnancy

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Abstract
Context: Urine culture is the gold standard for diagnosing bacteriuria in pregnancy, which is a very serious condition with a lot of consequences to both mother and foetus. Several chemical tests can equally be used to detect presence of bacteria in urine.
Objective: The aim of this study was to determine the accuracy of Chlorhexidine solution in detecting significant bacteriuria in pregnancy
Methods:This was a cross- sectional study in which patients with urinary tract infection had their urine samples tested with Chlorhexidine and bacteriological method. The predictive value method was used to compare both methods.
Results: A total of seventy-five (75) patients had clinically diagnosed urinary tract infection. Of these, only twelve (16%) were confirmed by bacteriology. Chlorhexidine detected all of these patients with laboratory- confirmed urinary tract infection and also detected other patents with abnormal components in their urine. Sensitivity was 100%, accuracy 40% and a specificity of 28.5% . The low specificity was due to the fact that Chlorhexidine solution cross reacts with bacteria and other major components of urine when present in significant amount.
Conclusion: The Chlorhexidine reaction is of little value in detecting bacteriuria per se, but it is useful for detecting other anomalies such as the presence of epithelial cells, crystals and pus cells. The test is however recommended for use as a screening tool for urinary tract infection in pregnancy because of its high sensitivity, cost effectiveness and its ease of performance.

Key Words: Evaluation, Chlorhexidine, Bacteriuria, Pregnancy.

Introduction

Pregnancy causes anatomical and physiological changes in the urinary system [1] These changes include amongst others, increased circulating hormone levels and pressure effects of the pregnant uterus on the collecting system, resulting in dilatation of the ureters, renal pelvis and calyces [2,3,4] The altered secretions of steroid sex hormones and the pressure exerted by the gravid uterus upon the bladder, cause hypotonia and congestion predisposing to uretero vesical reflux and urinary stasis [5]. These changes make the urinary tract vulnerable to infection during pregnancy
The presence of urinary tract infection in pregnancy results in fetal and maternal morbidity. These include pre-term labour, intra uterine growth restriction and chronic pyelonephritis, a major cause of death in older women [5]. The importance of bacteruria and urinary tract infection cannot be overemphasized in pregnancy. Effective antibiotic treatment must necessarily be based on accurate diagnostic method because treatment of bacteruria in pregnancy significantly reduces the incidence of acute pyelonephritis [6].

There is the need to have accurate laboratory methods of diagnosing urinary tract infection in pregnancy to ensure its prompt treatment. Urine microscopy and culture which is the gold standard for diagnosing urinary tract infection has several limitations particularly in areas where there is no electricity supply and facilities for microscopy and culture do not exist. Therefore, there is a need to develop cheap and result oriented tests for accurately detecting urinary tract infection in resource poor settings.

This study compared the accuracy of Chlorhexidine 1:1000 solution with bacteriological method in detecting bacteriuria in pregnancy. The above concept was based on a chance observation 20 years ago, by one of the authors, when addition of Chlorhexidine to contaminated urine yielded a cloudy and particulate solution. A pilot study carried out confirmed the observation.

Addition of Chlorhexidine to suspensions of bacterial cells produces an immediate adsorption of Chlorhexidine molecules on the bacterial surface [7]. The events following this adsorption would depend on the extent of adsorption. At low concentration (approximately 0.01%) there is a rapid and irreversible loss of cytoplasmic constitutions [8,9]. This leads to precipitation of the cellular protein and nucleic acid [7]. The action of Chlorhexidine on pus cells or other cellular components in the urine is not known but Chlorhexidine is known to be adsorbed on pus cells, serum and red blood cells [7]. Addition of Chlorhexidine to salts such as phosphates, silicates and carbonates would produce precipitation of insoluble salts [7] These salts are occasionally found in urine.

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This study intended to evaluate the accuracy of Chlorhexidine in detecting bacteriuria in pregnancy.

Materials and Methods

This was a cross sectional study conducted at the Department of Obstetrics and Gynecology in the University of Benin Teaching Hospital. All patients who presented with symptoms of urinary tract infection in pregnancy were recruited into this study. Their mid stream urine samples were shared in two parts, one part for the chemical test with Chlorhexidine 1:1000 solution in water. The other part of the urine was sent for microscopy, culture and sensitivity. Results from both tests were then compared. Patients on antibiotics and patients with known renal disease and hypertensive disease were excluded from the study population.

Using a laboratory dropper, 10drops of urine were placed in a clean test tube the colour and appearance of the urine was noted. 5 drops of Chlorhexidine were added to the test tube and as this was being done, the urine in the test tube was observed for any changes. E.g cloudiness, precipitate or particulate matter. Lastly the entire contents of the test tube are shaken and observed after one minute. A cloudy colour indicates a positive result. Half the volume of Chlorhexidine (5drops) was titrated against the full volume of urine (10drops) because this had been found in an earlier pilot study to be the critical point were the colour changes were maximal. This was not surprising because increase in concentration of Chlorhexidine produces increase in the amount adsorded but a decrease in the rate of loss of cytoplasmic constituent[7].

For the purpose of this study significant Pyuria was defined as greater than three pus cells per high power field (hpf.) [10]. While microscopic haematuria was defined as greater than two red cells per high power field (hpf) [10]. Epithelial cell count greater than three per high power field (hpf) was taken as significant [10]. A Database was created using Microsoft Excel 97 for ease of data management and analysis. The statistical method for describing the performance of a diagnostic test using the predictive value method was employed [11].

Table 1:
Comparison of culture results against the Chlorhexidine Reaction

Culture.	Chlorhexidine Reaction		Total
	Positive (%)	Negative (%)	
Positive	12 (16)	0(0)	12(16)
Negative	45 (60)	18 (24)	63 (84)
Total	57 (76)	18 (24)	75 (100)

Sensitivity = 100%
Specificity = 28.5%
False negative Rate =0%
Predictive value positive = 21.1%
Predictive value Negative = 100%
false positive Rate = 71.5%

Results

A total of 615 women attended the antenatal clinic during the study period of these 75 mid-stream urine sample were analysed for this study. Twelve samples (16%) yielded a bacterial growth on culture. Of the positive cultures, 6 (8%) yielded growth of Staphycoccus aureus, 3 (4%) yielded growth of Escherichia Coil and the remaining 3 (4%) yielded growth of Klebsiella specie. The incidence of urinary tract infection during the period of the study was therefore 1.9%.

Of the 75 mid stream urine samples 24 (32%) had more than 3 pus cells (pyuria) while 51 (68%) has less than 3 pus cells per high power field. There was no case of microscopic haematuria or urinary casts present in the urine. Thirty (40%) of the total number of samples had significant number of Epithelial cells in their urine. 18 (24%) patients has crystals in their urine 15 (20%) patients had calcium oxalate crystals while 3 (4%) has cystine crystals.

Evaluation of the urine specimen using the Chlorhexidine reaction showed that 57 (76%) patients has a positive result. Eighteen (24%) patients had a negative Chlorhexidine reaction. All the (12) patients with positive cultures (laboratory confirmed urinary tract infection) has a positive Chlorhexidine reaction. Of the 63 (84%) patients with negative cultures 45 (60%) had a positive Chlorhexidine reaction. None of the 18 patients who had a negative Chlorhexidine reaction had a positive culture or pyuria. They neither had microscopic haematuria, significant number of epithelial cells nor crystals in their urine. No other observations in respect to colour changes or precipitate were made. The evaluation of the urine using Chlorhexidine viz bacteriology showed that the Chlorhexidine reaction was 100% sensitive in detecting positive urine cultures but had low specificity at 28.5%. The false positive rate was 71.5%.

There were no false negatives and the accuracy was 40% Table 1. The Chlorhexidine reaction was also 100% sensitive in detecting pyuria but it had a specificity of only 35.3%. There however were no false negatives. The accuracy was 56% Table 2.

Table 2:
Relationship between the chlorhexidine Reaction and miscroscopy in detecting pyuria

Microscopy.	Chlorhexidine Reaction		Total
Pus Cells	Positive (%)	Negative (%)	
> 3 cells	24 (32)	0(0)	24 (32)
< 3 cells	33 (44)	18 (24)	51 (68)
TOTAL	57 (76)	18 (24)	75 (100)

Sensitivity = 100%
Specificity = 35.3%
False Negative Rate =0%
Accuracy = 56%
Predictive value positive = 42.1%
Predictive value Negative = 100%
False Positive Rate = 64.7%

Discussion

Urine culture is the Gold standard for detecting bacteriuria and urinary tract infection. All the positive cultures were detected by the Chlorhexidine reaction. This could be explained on the basis that Chlorhexidine acts on bacteria causing a rapid and irreversible loss of cytoplasmic constituents [8,9]. This leads to precipitation of cellular protein and nucleic acid resulting in the cloudy colour change which is observed. [7] The Chlorhexidine reaction when compared to other chemical tests, especially the Greiss test has the same sensitivity of 100% [12] However the specificity and predictive value of the Chlorhexidine reaction compared to the Greiss test is much lower, while the Chlorhexidine reaction has specificity and predictive value of 28.5% and 21.1% respectively, the Greiss test has 99.5% and 90.5% respectively as its specificity and positive predictive value [13]. This may be attributed to the fact that the Chlorhexidine reaction has a high detection rate for major components of urine, like cells and crystals. This ability to recongnise true positives is what confers on the Chlorhexidine reaction its sensitivity of 100%. The Greiss test may have false negatives in patients on ascorbic acid or if frequent voiding of dilute urine does not allow sufficient time for nitrites to be produced [13]. The Chlorhexidine reaction has no false negatives, thus making it a more appropriate screening test.

A total of 24 patients had pyuria, with only 12(50%) of them having positive cultures. This confirms previous ascertion by Lang and Levin [14] of poor correlation between pyuria and urine culture. It must be recognized that pyuria may be due to conditions like renal tuberculosis, analgesic nephropathy and renal stones [15]. Cystine crystals were identified in 3 (4%) patients, and the presence of Cystine is indicative of renal stones. [14, 15] These patients contributed to the number of patients with sterile pyuria. The Chlorhexidine reaction was however very sensitive to pus cells with a sensitivity of 100%. This is not surprising because Chlorhexidine is known to be adsorbed in the surface of Pus cells [7] and may act on them like it does on bacterial cells causing loss of cytoplasmic constituents. The specificity of the Chlorhexidine reaction was very low at 35.3% and the ability to predict a positive result was 42.1%. These figure are very low and may again be attributable to the high sensitivity of Chlorhexidine to other major components of urine. The false negative rate and negative predictive value of the Chlorhexidine reaction is 0% ad 100% respectively, which gives it a high pick up rate.

In conclusion the Chlorhexidine reaction is very sensitive in detecting bacteriuria and pyuria in pregnancy with sensitivity of 100%. Unfortunately it is also 100% sensitive to other components of urine which are not necessarily pathological e.g Epithelial cells, crystals.The net result is a low specificity and accuracy.The test itself is of little value in detecting bacteriuria per se, but is useful for detecting other anomalies such as the presence of epithelial cells, pus cells and crystals. It is therefore recommended that this test be only considered as a screening test in pregnancy as it would identify all patients with bacteruria and pyuria .The test has the advantage of being very cheap, cost being about forty kobo (less than one cent US) per test, compared with other chemical tests (e.g Greiss test). It is a simple test to conduct and can easily be done by low-grade medical personnel.It does not need the use of a microscope or power supply of any sort and so would be of advantage in the rural areas. The limitations of the study include the fact that the study population was small and Chlorhexidine reaction would need further research and more evaluation in order to improve its specificity.

References

1. Cardozo ID. Urinary tract disorder in pregnancy In: Whitfield CR (ed), *Dewhurst's Textbook of Obstetrics and Gynaecology for Postgraduates*, 5th Edition, Blackwell Publication, 1995; 251-255.
2. Cietak KA and Newton JR Serial Quantitative Maternal Nephrososnography in Pregnancy. *Br J Radiol* 1985 58; 405 13.
3. Dunlop W and Davidson JM. Renal haemodynamics and tubular function in human pregnancy. *Bacilhire's Chi Obstet Gynaecol* 1987; 1: 769-87.
4. Lindheimer MDD and Katz AL The kidney in pregnancy. In Brenner BM, Rector FC Jr (eds). *The kidney*. Philadephia : WB Saunders, 1986; Pp 1253-95.
5. Benson RC. Medical and surgical complications during pregnancy. In: *Current obstetrics and gynecologic diagnosis and treatment*. Lange Medical Publications, 1984; 907-908.
6. Gratacos E, Torres PJ, Vila J Alonso PL, Cararach V. Screening and treatment of Asymptomatic Bacteriuria in pregnancy prevent Pyelonephritis. *J Infect Dis* 1994; 169: 1390-1392.
7. Lognworth AR. Chlorhexidine. In: Hugo WB (ed), *Inhibition and destruction of the microbial cell*. London: Academic Press, 1971; 95-105.
8. Hugo WB and Wiseman DJ. *Pharm Pharmac* 1964; 16, 655-662.
9. Rye RM and Wiseman DJ. *Pharm Pharmac* 1964; 16, 516-21
10. Sternheimer R and Malbin B Clinical. Recognition of Pyelonephritis. *Am J Med* 1951; 11: 312.
11. Knapp RG, Miller MC. *Clinical Epidermiology and Biostatistics*. Pennsylvania: Harwal Publishing Company, 1992.
12. Javier O.I.F, Gayon V.E, Arredondo G.J.L Usefulness of two test for rapid diagnosis of urinary infections in pregnancy. *Ginecol Obstet Mex* 1993; 61: 290-4.
13. Mathews J.E, George S; Mathews P; Mathai E, Brahmadathan K.N, Seshadri. L The Griess test: an inexpensive test for asymptomatic bacteriuria in pregnancy. *Aust NZ J Obstet Gynaecol* 1998; 38 (4): 407-10.
14. Lang G.R and Levin S. Diagnosis and treatment of urinary tract infection. *Med Clin North Am* 1971; 55: 1439.
15. Sweny P and Varghese Z: *Intermediate test in clinical test; renal disease*. Wolfe Medical Publications Ltd 1988; 19-26.