

## Crosssectional Study of Asymptomatic Bacteriuria and to Establish Antimicrobial Drug Susceptibility Among Pregnant Women in Ateaching Hospital ,Telangana

Siva Santhi Atmakuri<sup>1</sup>, Afreen Iqbal<sup>2</sup>, Mallely Ravi Kumar<sup>3</sup>

<sup>1</sup>Assistant Professor, Department of Microbiology, Shadan Institute of Medical Sciences, Peerancheru, Hyderabad, Telangana, India

<sup>2</sup>Assistant Professor, Department of Microbiology, Shadan Institute Of Medical Sciences, Peerancheru, Hyderabad, Telangana, India

<sup>3</sup>General Medicine, Civil assistant surgeon, Community health centre, Rajendranagar, Budwel, Telangana, India

Received: 01-05-2021 / Revised: 19-06-2021 / Accepted: 15-07-2021

### Abstract

**Background :** Asymptomatic bacteriuria is defined as the presence of bacteria in urine without having signs and symptoms. Urinary tract infection is widespread among pregnant women and is accompanying adverse maternal, fetal, and neonatal outcomes. **Aim of the study :** To study the prevalence of asymptomatic bacteriuria and to establish antimicrobial drug susceptibility in pregnant women attending in teaching hospital; ,Telangana. **Materials and methods :** Cross sectional study done on 230 early morning midstream urine samples from pregnant women who came for regular antenatal checkup for the department of Gynaecology over a period of one year i.e, between August 2019 and July 2020. The samples were sent for further processing to the department of Microbiology, Shadan Institute Of Medical Sciences ,Peerancheru, Hyderabad, Telangana. **Results :** Prevalence of significant bacteriuria was 52.1%. Higher rate of infection was seen in 1st and 2nd trimester of pregnancies, 41.6% and 45.8%. On microscopy pus cells were seen in 79.1% of urine samples and epithelial cells were seen in 50% samples. rbc's 1.6%, 25% wbc's. The commonest isolated organism was E. coli in 69.1% patients followed by Klebsiella pneumonia 15%. Imepenam, Amikacin, Gentamycin and Nitrofurantoin were found to be highly effective drugs against most of isolates from urine of pregnancies. ceftriaxone, levofloxacin revealed moderately effective against those isolates. **Conclusion :** The prevalence of asymptomatic bacteriuria in pregnant women in our study was 52.1% and E. coli was the dominant organism isolated. Quantitative urine culture is the ideal test for detection of asymptomatic bacteriuria. Early detection and treatment are essential to protect the health of mother and fetus.

**Keywords:** Significant bacteriuria, E.coli, Klebsiella.

This is an Open Access article that uses a fund-ing model which does not charge readers or their institutions for access and distributed under the terms of the Creative Commons Attribution License (<http://creativecommons.org/licenses/by/4.0>) and the Budapest Open Access Initiative (<http://www.budapestopenaccessinitiative.org/read>), which permit unrestricted use, distribution, and reproduction in any medium, provided original work is properly credited.

### Introduction

Urinary tract infections represent the most common bacterial infection in pregnancy. Asymptomatic bacteriuria occurs in 2–10% of all pregnancies [1]. The prevalence of bacteriuria in pregnancy is closely related to socioeconomic status [1]. Other factors that have shown an association with bacteriuria include a history of recurrent urinary tract infections, diabetes and anatomical abnormalities of the urinary tract [2]. The effects of other host factors, such as race, sickle cell disease, age and parity on the prevalence of bacteriuria are less clear and there is controversy in the literature [3,4]. One of the biggest risk factors for symptomatic infection is asymptomatic bacteriuria. The presence of a considerable amount of bacteria in the urine in the absence of clinical signs or symptoms of a urinary tract infection is known as asymptomatic bacteriuria (ABU).

Asymptomatic bacteriuria is one of the most common risk factors for UTIs during pregnancy, accounting for around 70% of cases. It causes 40% cystitis and 30% pyelonephritis if left untreated, which can result in early or low-birth-weight babies, intrauterine growth retardation, preterm labour, intrauterine foetal death, and increased perinatal mortality and morbidity. Early detection and treatment of ABU during pregnancy can reduce the occurrence of maternal problems such as anaemia, preeclampsia,

renal failure, and septicemia. ABU screening is suggested in all pregnant women, according to the American College of Obstetricians and Gynecologists. Antibiotic therapy directed at the cultured organism should be given to pregnant women with ASB. As a result, ASB screening has been identified as a cost-effective strategy for enhancing maternal and newborn health. Despite this, inaccurate assays are employed to detect ABU, and antimicrobials are routinely utilised on a trial-and-error basis, particularly in poorer countries. The impact of antibiotic misuse on human pathogen antimicrobial susceptibility reduces the efficacy of current and future antimicrobial drugs, and the creation of resistant bacterial infections is on the rise.

The aetiologic agents associated with bacteriuria are similar in pregnant and non-pregnant women. The relatively short female urethra is frequently colonized with organisms from the gastrointestinal tract. Escherichia coli is the most common pathogen associated with both symptomatic and asymptomatic bacteriuria, representing 70–80% of isolates [5,6]. Other organisms include other gram negative bacteria and Group B streptococcus. The original criterion for diagnosing asymptomatic bacteriuria was  $> 10^5$  cfu/mL of a single uropathogen on two consecutive clean catch samples, with a 95% probability that the woman has true bacteriuria. The detection of  $> 10^5$  cfu/mL in a single voided midstream urine is accepted as a more practical and adequate alternative, although there is only an 80% probability the woman has true bacteriuria [7]. In non-pregnant symptomatic patients with an identified pathogen, specifically E. coli or Staphylococcus saprophyticus, a colony count of  $\geq 10^2$  –  $10^3$  cfu/mL may indicate infection, but this cut-off has not been

\*Correspondence

Dr. Afreen Iqbal

Assistant Professor, Department of Microbiology, Shadan Institute Of Medical Sciences, Peerancheru, Hyderabad, Telangana, India

E-mail: [drafreeniqbal999@gmail.com](mailto:drafreeniqbal999@gmail.com)

evaluated for symptomatic urinary tract infections in pregnancy [8]. Penicillin, Amoxicillin, ceftaxidime, norfloxacin, and cefoxitin are the most competent antibiotics for the treatment of the majority of the urinary tract pathogens [9]. The changes in the urinary tract during pregnancy and immune system increase the prevalence of bacteriuria, causing serious risks for both mother and fetus. Parity, Increasing age, sickle cell anemia, diabetes, urinary tract disorders and history of UTI, may increase the risk of urinary tract infection in pregnant women [10,11].

**Aim of the study:** To study the prevalence of asymptomatic bacteriuria and to establish antimicrobial drug susceptibility of the pathogens resulting asymptomatic UTI in pregnant women attending in teaching hospital; Telangana

#### Materials and Methods

The study was approved by the Institutional Ethics Committee . Written informed consent was obtained from the all the cases included in the study .

**Sample size:** A total of 230 midstream urine samples of pregnant women .

**Type of study:** Cross sectional study .

**Place of study:** Department of Microbiology at Shadan institute of medical sciences, Peerancheru, Hyderabad, Telangana.

**Duration of study:** One year between August 2019 and July 2020.

**Inclusion criteria :** Pregnant women with Age between 20 to 45 years ,Pregnant women without any symptoms of UTI such as lower abdominal pain, fever, burning micturition, frequency of micturition, dysuria.

**Exclusion criteria:** Age <20 years and > 45 years, Cases having symptomatic UTI, Diabetes, Past history of UTI, History of any antibiotic use .

The pregnant women included in this study were randomly selected We collected demographic data such as age, weeks of gestation , parity,height, weight, BMI,from the file records in the Gynaecology and Obstetrics department in our institute. Each pregnant women included in the study were asked to collect early morning clean catch mid-stream urine sample into a container which was sterile, screw capped, and labelled with sample number and patient details . and all were advised to clean the external genitalia properly. Samples were sent to microbiology laboratory within 30minutes of collection. Firstly Complete urine examination was done including physical ,chemical and microscopy examination .To limit the risk of contamination, urine specimens were obtained from each pregnant woman who had been instructed by midwives to collect mid-stream pee. In a sterile screw-capped, wide-mouth container, 10 to 15 mL of mid-stream urine samples were collected from each pregnant woman. It was subsequently transferred to the microbiology laboratory, where it was processed for analysis in 1–2 hours. For GBS and other bacteria, the cut-off values of traditional semi-quantitative evaluation (S-QBC) for substantial bacteriuria were determined to be 10<sup>3</sup> cfu/mL and 10<sup>5</sup> cfu/mL, respectively. For the diagnosis of GBS-induced UTI, a lower threshold of 10<sup>3</sup> S-QBC would serve. Colony features, gram-staining technique, and biochemical pattern following

standard protocols were used to identify isolates at the species level. The indole test, lysine decarboxylase, H<sub>2</sub>S and gas production in triple sugar agar, citrate utilisation, urease, and motility tests were used to identify the majority of Gram-negative bacteria. The Gram-positive bacteria were identified using catalase and coagulase tests, for the identification of GBS, pyrrolidonylarylamidase test (PYR), CAMP test and bacitracin and trimethoprim-sulfamethoxazole tests were used.

On physical examination we noted color and appearance and pH of urine sample. Microscopy examination was done for pus cells ,epithelial cells ,casts & crystals .The urine was cultured on blood agar, Mac conkey agar and CLED agar. A loopful of well mixed uncentrifuged urine was streaked on to the surface of culture plates. Incubation was done aerobically at 35 deg C for 18-24 hours. After 24 hours of inoculation, The urine culture plates were examined macroscopically to identify the color, appearance, morphology, and the colonies size. The isolated bacteria were diagnosed by using Gram stain and biochemical tests

#### Antimicrobial susceptibility

The isolates were identified and antimicrobial susceptibility done by the standardized Kirby-Bauer disc diffusion method on Muller Hinton agar plate. According to recommendations of Clinical Laboratory Standard Institute (CLSI) for antibiotic sensitivity testing and based on the size of inhibition zone around the disc three forms of Sensitive (S), Intermediate (I) and Resistant (R) pattern interpreted [12,13]

SPSS version 20 for Windows was used to enter and analyse the data. To find parameters linked to ASB, researchers used stepwise logistic regression analysis. A statistically significant P value of 0.05 was used.

#### Results

In the present study out of 230 urine samples among pregnant women, 52.1% (120/230) of urine samples revealed with significant bacteriuria considered as the asymptomatic UTI, while 47.8% (110/230) samples does not show bacterial growth . Age range from 20 years to 45 years .Among age distribution majority of age group were among 31-35 years ie, 37.5% ( 45/120) followed by 31.6% ( 38/120) among 25-30 years .20.5 % ( 25/120) among 20-25 years and 8.3%(10/120) among 36-40 years and 1.6% ( 02/120) among 41-45 years. In the present study on physical examination of urine. Yellow coloured urine was found in 70.8% (85/120) of cases, and 12.5 % (15/120) presented as deep yellow and 16.6% ( 20/120) were having urine as colourless .70.8% had acidic PH, 16.6% had alkaline PH and 12.5% had the neutral PH .Turbidly was seen in 75%(90/120) urine samples and 25% ( 30/120) showed clear appearance. On microscopy pus cells were seen in 79.1 % of urine samples And epithelial cells were seen in 50% samples ,rbcs 1.6%, 25% wbc. A higher rate of infection was seen in 1st and 2nd trimester of pregnancy ie, 41.65 (50/120) and 45.8% ( 55/120), followed by 12.55 (15/120) in third trimester .In the present study Significant bacteriuria was found more in primigravidae, 66.6% (80/120) than multigravida 33.3%(40/120).

**Table 1: Showing Isolated bacteria**

Isolated bacteria	No . of cases	%
Escherichia coli	83	69.1
Klebsiella pneumonia	18	15
Acenobacter	03	2.5
Pseudomonas aeruginosa	02	1.6
Proteus vulgaris	01	0.8
S.aureus	10	8.3
Enterococci	03	2.5
Total	120	99.9

The commonest isolated organism was E. coli in 69.1% (83/120) patients followed by Klebsiella pneumonia 15% (18/120) Acenobacter in 2.5% ( 03/120 ) .Pseudomonas aeruginosa in 1.6% (02/120) Proteus vulgaris in 0.8% (01/120) S. aureus in 8.3% (10/120) and Enterococci in 2.5% ( 03/120) .

Table 2: Antibiotic sensitivity pattern of gram negative and gram positive bacteria

Gram negative and gram positive bacteria	Amikacin	Gentamycin	Nitrofurantoin	Cefotriaxone	Levofloxacin	Ampicillin	vancomycin	Imipenem
E. Coli	37(44.5%)	47(56.6%)	76(91.5%)	45(54.2%)	26(31.3%)	12(14.4%)	-	80(96.3%)
K.pneumonia	08 (44.4%)	06(33.3%)	04(22.2%)	08(44.4%)	05(27.7%)	01(5.5%)	-	09(50%)
Acenobacter	01(33.3%)	01(33.3%)	01(33.3%)	01(33.3%)	02(66.6%)	0	-	03
P.aeruginosa	01(50%)	02(100%)	0	01(50%)	0	0	-	-
P. vulgaris	0	01(100%)	01	01(100%)	0	0	-	-
S.aureus	06(60%)	05(50%)	08(80%)	05(50%)	05(50%)	05(50%)	10(100%)	-
Enterococcus faecalis	0	01(33.3%)	03(100%)	0	01(33.3%)	0	02(66.6%)	-

In this study, Imepenam, Amikacin, Gentamycin and Nitrofurantoin were found to be highly effective drugs against most of isolates from urine of pregnancies. ceftriaxone, levofloxacin revealed moderately effective against those isolates.

#### Discussion

Antenatal care is provided free of charge in government-owned health institutions. During the follow-up phase, however, there is no systematic urine culture test for pregnant women to screen for ASB; instead, they are checked for urinalysis using urine strip testing, and treatment is empirical. Early identification of ASB during pregnancy is critical for avoiding negative outcomes. For service auditing, it's useful to know the amount of ASB and the isolates' current antimicrobial resistance profile.

**Comparative studies according to Prevalence:** In the Present study the prevalence of asymptomatic bacteriuria in pregnant women was 52.1% (120/230). In Laxmi Kumar Yadav et al study [14] 51.83% of urine samples revealed with significant bacteriuria considered as the asymptomatic UTI, hence correlating with our study. Whereas in Omer Mohammed Ali Ibrahim et al study [15] the prevalence of asymptomatic bacteriuria in pregnant women was 25/192 (13%). The vast range of ASB prevalence reported across studies is due to differences in related factors, sample size, regional variations, community social habits, and health education practise.

#### Comparative studies related to Age distribution

In the present study Age range from 20 years to 45 years. majority of age group were among 31-35 years ie, 37.5% (45/120) followed by 31.6% (38/120) among 25-30 years. In Aliasghar Farazi et al study [16] asymptomatic bacteriuria was found in the age group of 18-35 yrs (65.4%). In Anu Mary Bose et al study [17] Most common age group was 26-30 years followed by 21-25 years and 31-35 years. In Laxmi Kumari Yadav et al study [14] The subjects involved in this study had a mean age of 28.84 years. In Sudha Biradar Kerure et al study [18]. The highest number of culture positive cases among pregnant women were in the age group of 26-35 years (52%), In Omer Mohammed Ali Ibrahim et al study [15] asymptomatic bacteriuria was noted in 25% of pregnant women in the age group (12-20 years). This may be explained by the fact that early and intensive sexual intercourse which may cause minor urethral trauma and transfer bacteria from the perineum into the bladder. Similar finding was also reported in the previous studies

Table 3: showing Comparative studies related to bacterial isolates

Isolated bacteria	Sudha Biradar Kerure et al study [18]	In Aliasghar Farazi et al study [16]	Anne CC Lee et al study [19]	Laxmi Kumari Yadav et al study [14]	Omer Mohammed Ali Ibrahim et al study [15]	Present study
E. coli	77.7%	46.2%	38%	67.8%	32%	69.1
K.pneumonia	6.66%	15.4%	12%	21.6%	16%	15
Acenobacter	2.22%	-	-	5.2%	-	2.5
P. aeruginosa	-	-	-	1.5%	8%	1.6
P. vulgaris	-	-	-	0.3%	28%	0.8
S.aureus	11.1%	23.1%	23%	-	12%	8.3
Enterococci	-	7.7%	-	-	-	2.5
Streptococcus Group B	-	7.7%	5.3%	-	4%	-

#### Comparative studies related to CUE

In the present study Yellow coloured urine was found in 70.8% (85/120) of samples and 12.5% (15/120) presented as deep yellow and 16.6% (20/120) were having urine as colorless. whereas in Laxmi Kumari Yadav et al study [14] it was found that urine color was light yellow in 84% of cases while 12.3% have deep yellow and remaining 2.8% have colorless.

**Comparative studies related to PH:** In the present study 70.8% had acidic PH, 16.6% had alkaline PH and 12.5% had the neutral PH. Laxmi Kumari Yadav et al study [14]. 93.8% had acidic PH, 5.8% had alkaline PH and only 0.3% had the neutral PH.

#### Comparative studies related to trimester distribution

In the present study, a higher rate of infection was seen in 1st and 2nd trimester of pregnancy ie, 41.65 (50/120) and 45.8% (55/120) followed by 12.55 (15/120) in third trimester. In Anu Mary Bose et al study [17]. Maximum number of asymptomatic bacteriuria was detected in 5-10 weeks of gestation (50%) followed by 10-15 weeks (26.92%) and 15-20 weeks (19.23%). In Aliasghar Farazi et al study [16]. In this study, a higher rate of infection was detected in 1st and 2nd trimester of pregnancy (34.6%), followed by third trimester (30.8%). In Omer Mohammed Ali Ibrahim et al study [15] The study revealed that 25% of pregnant women in the first trimester were with asymptomatic bacteriuria, and lesser percentages were noted in women in the second and third trimesters 11.7% and 11.6%. This could be because ASB/UTI in pregnant women usually starts around week 6 and peaks around week 22-24 due to urethral dilatation, increased bladder volume, and decreased bladder tone, as well as decreased urethral tone, all of which favour bacterial growth in the urine. The gold standard for detecting ASB is said to be within the first 12-16 weeks of pregnancy. Another study came to a similar conclusion.

#### Comparative studies related to Parity

In the present study Significant bacteriuria was found more in primigravidae, 66.6% (80/120) than multigravida 33.3% (40/120). In Anu Mary Bose et al study [17] asymptomatic Significant bacteriuria ASB was more common among primigravida (53.85%) followed by gravida 2 (30.77%). In Sudha Biradar Kerure et al study [18] Significant bacteriuria was found more in primigravida (56%) than multigravida (44%). In Sreekumary Radha et al study [20], 82.45% were multigravida and 15.75% were primigravida.

**Antibiotic sensitivity pattern of gram negative and gram positive bacteriuria**

In the Present study imepenam.,Amikacin, Gentamycin and Nitrofurantoin were found to be highly effective drugs against most of isolates from urine of pregnancies. ceftriaxone, levofloxacin revealed moderately effective against those isolates. Omer Mohammed Ali Ibrahim et al study[15].The isolated bacteria showed high susceptibility to amikacin (84%), followed by ciprofloxacin (76%),and gentamycin (56%). And all isolates were resistant to cotrimoxazole.In Aliasghar Farazi et al study[16]The maximum antibiotics resistance was to three drugs cotrimoxazole, nalidixic acid and Amoxicillin/clavulanate and the least resistance was to nitrofurantoin, ceftriaxone, and norfloxacin. The majority of widely prescribed antimicrobials are freely available at local pharmacies, and people can buy and use them without a prescription, potentially contributing to drug resistance.

**Conclusion**

The prevalence of asymptomatic bacteriuria in pregnant women in our study was 52.1% and E. coli was the dominant organism isolated. Quantitative urine culture is the ideal test for detection of asymptomatic bacteriuria.Early detection and treatment are essential to protect the health of mother and fetus.As a result, antenatal care practises should include screening and treatment of pregnant women for ASB, particularly in the first and second trimesters, and there is a need for periodic surveillance of the type of bacterial pathogens and their updated antimicrobial resistance profile in the study .

**References**

- Whalley P. Bacteriuria of pregnancy. *Am J Obstet Gynecol.* 1967;97:723–38
- Golan A, Wexler S, Amit A, Gordon D, David MP. Asymptomatic bacteriuria in normal and high-risk pregnancy. *Eur J Obstet Gynecol Reprod Biol.* 1989;33:101–8
- Sharma P, Thapa L. Acute pyelonephritis in pregnancy: a retrospective study. *Aust N Z J Obstet Gynaecol.* 2007;47:313–5.
- Thurman AR, Steed LL, Hulsey T, Soper DE. Bacteriuria in pregnant women with sickle cell trait. *Am J Obstet Gynecol.* 2006; 194:1366–70.
- Gilstrap LC, Leveno KJ, Cunningham FG, Whalley PJ, Roark ML. Renal infection and pregnancy outcome. *Am J Obstet Gynecol.* 1981;141:709–16.
- Mc Fadyen IR, Eykyn SJ, Gardner NH, Vanier TM, Bennett AE, Mayo ME et al. Bacteriuria in pregnancy. *J Obstet Gynaecol Br Commonw.* 1973;80:385–405
- Kass EH. The role of asymptomatic bacteriuria in the pathogenesis of pyelonephritis. In: Quinn EL, Kass EH, editors. *Biology of Pyelonephritis.* Boston: Little, Brown and Co, 1960, 399–412p.
- Stamm WE, Counts GW, Running KR, Fihn S, Turck M, Holmes KK. Diagnosis of coliform infection in acutely dysuric women. *N Engl J Med.* 1982;307:463–8.
- Faidah H, Ashshi A, HadaA, Al-ghamdi G, Amr M. Urinary Tract Infections among pregnant women in Makkah, Saudi Arabia *Biomedical & Pharmacology Journal.* 2013; 6(1):01-07.
- Giraldo PC, Araújo ED, Junior JE, Amaral RLGD, Passos MRL, Gonçalves AK. The prevalence of urogenital infections in pregnant women experiencing preterm and full-term labor. *Infect Dis Obstetrics Gynecol.* 2012, 1–4 .
- Raza S, Pandey S, Bhatt CP. Microbiological analysis of isolates in Kathmandu medical college teaching hospital, Kathmandu, Nepal. *Kathmandu Univ Med J (KUMJ).* 2011; 9(36) :295-7.
- Performance Standards for Antimicrobial Disc Susceptibility Tests; Approved Standard. twelfth ed. Wayne, PA.USA: M02-A12. National Committee for Clinical Laboratory Standards, 2015, 32(1).
- National Committee for Clinical Laboratory Standards. Performance Standards for Antimicrobial Susceptibility Testing: Twelfth Information Supplement. NCCLS Document M100- S12 Wayne, PA: NCCLS, 2002.
- Laxmi Kumari Yadav, Ram LochanYadav. Asymptomatic UTI in pregnancy attending at tertiary care of Nepal international Journal of Research in Medical Sciences. *Int J Res Med Sci.* 2018; 6(4):1119-1128
- Omer Mohammed Ali Ibrahim, Mohammed EH Azoz, Mohammed Ibrahim Alsadig, Mahdi BAAIagab, Abdelmageed Elmugabil. Asymptomatic Bacteriuria in Pregnant Women at Kosti Teaching Hospital, Kosti-White Nile State (Sudan): *International Journal of Current Microbiology and Applied Sciences,* 2018, 7(06):09
- Aliasghar. Farazia. Asymptomatic bacteriuria in pregnancy in the central region of Iran: Frequency, risk factors, and causative organisms :*Clinical Epidemiology and Global Health.* 2019; 7:309-312
- Anu Mary Bose, Sreekumary PK, Sobha KurianPulikkottil. Microbiological profile of asymptomatic Bacteriuria in Pregnancy:*Critical Care Obstetrics and Gynecology.* 2016; 2(5):26
- Sudha Biradar Kerure,Sheela S Sagarad, Sneha. Asymptomatic bacteriuria among pregnant women: *International Journal of Reproduction, Contraception, Obstetrics and Gynecology.*Int J Reprod Contracept Obstet Gynecol. 2013;2(2):213-216
- Anne CC Lee, Luke C. Mullany, Alain K. Koffi, Lian V. Folger, Dipak K. Mitra, Alain Labrique, Parul Christian, Jamal Uddin, Parvez Ahmed, Mohammad A. Quaiyum, Samir K. Saha, Abdullah H. Baqui. Urinary tract infections in pregnancy in a rural population of Bangladesh: population based prevalence, risk factors, aetiology, and antibiotic resistance : *BMC Pregnancy and Childbirth.* 2020; 20:1
- Sreekumary Radha, Shahida Jamal. Prevalence and outcome of asymptomatic bacteriuria in early pregnancy *International Journal of Reproduction, Contraception, Obstetrics and Gynecology: Int J Reprod Contracept Obstet Gynecol.* 2017; 6 (1): 223-227

**Conflict of Interest: Nil**

**Source of support:Nil**