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Original

Genitourinary Microbial Isolates in Prelabour Rupture of Foetal Membranes in University of Maiduguri Teaching Hospital, Maiduguri North Eastern Nigeria.

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ABSTRACT:

Background: Prelabour rupture of membranes (PROM) is a common Obstetric problem associated with maternal and perinatal morbidity and mortality. PROM complicates 10.7% of all pregnancies worldwide. **Objective:** To determine the prevalence and genitourinary microbial pattern for PROM among antenatal patients at the University of Maiduguri Teaching Hospital, Maiduguri. **Methodology:** Socio-demographic and obstetric variables were obtained from consecutive patients with PROM. For each patient an endocervical swab, high vaginal swab and urine samples were taken for micro bacteriologic studies. The next patient without PROM was used as control. Data were analyzed using SPSS 20. **Results:** A total of 129 women with PROM and another 129 without PROM were analyzed. Genitourinary culture was positive in 102 (79.1%) and 7 (5.5%) of cases and control respectively (p-value 0.035) (OR 1.627, 95% CI: 1.281-2.273). The major micro-organism cultured from the genital tracts of women with PROM were *Candida albicans* (32 %), *Staphylococcus aureus* (16 %), *Streptococcus Spp (*15.5%), E.Coli (5.4%), *Klebsiella (*3.9 %), *Pseudomonas* aeruginosa (3.9%) and *Proteus mirabilis (2.3%).* While their urine yielded *Escherichia coli* (15.5%), *Staphylococcus aureus* (3.9%), *Candida albicans* (5.4%), *Pseudomonas aeruginosa (*5%), *Klebsiella Spp* (7%), *Proteus mirabilis* (4.7%), and *Streptococcus spp* (0.8%). **Conclusion:** Genitourinary tract infection was found to be significantly associated with the risk of developing PROM with candida albicans and staphylococcus aureus being the commonest microbial organisms cultured.

Keywords: Genitourinary Microbial Isolates, Northeastern Nigeria, PROM

INTRODUCTION

Spontaneous rupture of fetal membrane before the onset of labour is referred to as Prelabour rupture of membranes. Term PROM complicates 8-10% of pregnancies.[1] Preterm premature rupture of membranes complicates 2-4%[1,2,3] of singleton pregnancies, and 7-20%[1,2] of twin pregnancies.

PROM aetiology is multifactorial; however, PROM may be linked to underlying pathological processes, most likely due to inflammation and/or infection of the fetal membranes. Infections also can cause

complications in pregnant women that had Prelabour rupture of membranes leading to life threatening maternal and or neonatal infection. Microbial patterns are important in studies concerning prelabour rupture of membranes because genito- urinary infections is an established risk factor for PROM and also PROM is directly associated with chorioamnionitis,[4] and neonatal/maternal infections that can lead to adverse maternal and neonatal outcomes. The lower genital tract is a reservoir for bacteria that may ascend through the cervical canal and cause localized inflammation; as shown in previous studies that patients with PROM reportedly have a higher rate of abnormal microbial colonization of the genital tracts than patients without PROM and the prevalence of positive amniotic-fluid cultures in PROM is approximately 32%-35%.[5]

Maternal morbidities such as intra-amniotic infection occur in 13-60%[6] of women with PROM, also other morbidities, such as life-threatening maternal sepsis complicate 1% of cases.[7] In our environment maternal infection contributes 12% to maternal mortality and is the 4th leading cause of maternal death.[7]

The purpose of this study was to determine the prevalence of PROM together with the association and pattern of genitourinary microbial isolates associated with PROM at the University of Maiduguri Teaching Hospital, Northeastern Nigeria, Previous studies have documented the pattern of genital microbial isolates and have been documented in Northwestern,[8] Western and Eastern Nigeria. However, there may be variance in the distribution of microorganisms causing PROM in different regions of Nigeria. As documented in literature there are variation in the distribution of microorganism in different regions and countries.[9] The difference in variation could not be ascertained without conducting a study at the northerestern Nigeria. The result so obtained would be helpful in the management of PROM.

METHODOLOGY

Study Design

The study was conducted between 1st May 2016 to 28th February 2017. It was a hospital based cross sectional analytical study aimed at determining the genitourinary microbial isolates present in the endocervical samples of patients with PROM and matched patients without PROM, from 28 weeks to term presenting to the department of Obstetrics and Gynaecology at University of Maiduguri Teaching Hospital, Maiduguri, Borno-State, Nigeria. Participants with PROM who were randomly recruited using systematic sampling method until the desired sample size was obtained. The sample size was obtained using the formula for calculating sample size for comparison groups.[10]

$$N = \frac{2Z^2pq}{d^2}$$

Where:

N= Desired sample size for comparison groups.

P = Proportion of the target population estimated to have the particular characteristics (PROM), which is 1.94% extrapolated from a similar study at University of Calabar Teaching Hospital, Calabar,[11] Nigeria.

Z = Standard deviation usually set at 1.96 confidence level, which corresponds to 95% confidence level.

q = 1.0 ─ P.

d = Degree of accuracy desired, which is set at 2.5 % (0.025) since p is < 5 %.

Therefore,

$$N = \frac{2(1.96)^2 \times 0.0194 \times 0.09806}{(0.025)^2}$$

$$N = \frac{0.14616}{0.000625}$$

N= 233

To allow for attrition of upto 10%, the obtained value will be divided by 0.90; 233 ÷ 0.90 = 258 Therefore, 258 combined population size (129 pregnant women with PROM and another 129 patients who satisfy the inclusion criteria were recruited for each group), and all the consenting participants that have met the inclusion criteria were matched for age, parity and gestational age with participants without PROM were also recruited and served as control.

The purpose of the study was explained to the participants and a written consent was obtained from all consenting participants. The Socio-demographic variables and clinical characteristics such as age, parity, and gestational age were noted and recorded into a investigator administered questionnaire. Other information that was obtained included educational status, social class, gynecologic history and history of previous obstetric outcomes. Course of the pregnancy and lifestyle were also obtained. Gestational age was determined by the last menstrual period and the results of any ultrasonographic examinations performed before 24 weeks gestation. The diagnosis of PROM among those with complaints of drainage of fluid from the vagina was made by the

researchers. Membrane rupture was confirmed by visualizing amniotic fluid leaking from the cervical os on sterile speculum examination. Swabs from endocervical canal and high vagina for culture and Gram stain were taken at initial examination. Urine samples were also obtained for microbiological analysis. The samples were immediately inoculated to the appropriate culture media and were sent immediately to the department of microbiology for microscopy, culture analysis and sensitivity testing. Data obtained were recorded on a proforma and analyzed using the statistical package for social sciences, SPSS V 20.0.0(2010), IBM, and New York, USA.

Inclusion criteria

1. All consenting pregnant women from gestational age of 28 weeks and above who presented to the department of obstetrics and gynecology of UMTH with PROM served as cases.

Exclusion criteria

1. Women who declined consent.
2. Patients with vaginal bleeding.
3. Patients with iatrogenic rupture of fetal membranes e.g., artificial rupture of membranes.
4. Patients who were currently on antibiotics or history of having used antibiotics in last 2 weeks.
5. Patients with history of PROM for more than 24 hours.
6. Patients with PROM that had digital vaginal examination

A total subject of 258 (129 with PROM and another 129 without PROM) completed the study and their data were analyzed. For the purpose of the study PROM would be defined as rupture of fetal membranes with no palpable uterine contractions in 10 minutes with no cervical changes. Samples were taken from both cases and controls. A sterile speculum examination was carried out. This procedure was explained to the participant and she was informed that she may experience slight discomfort during the procedure. After obtaining consent from the participant, the patient was placed in lithotomy position. A face mask was worn and the specimen collection was done using a sterile procedure. Hand washing was done and sterile gloves were worn. For participants with PROM liquor drainage was observed while no liquor drainage was observed in participants without PROM. Two endocervical swabs were taken separately by introducing a sampling swab into the cervical canal and rotated firmly for 15-20 seconds. Sample for high vaginal swab was obtained from the posterior fornix of the vagina, one sample obtained from the endocervical canal and high vagina was immediately inoculated into the three principal cultures; chocolate agar, blood agar and MacConkey agar. The chocolate agar was placed in an anaerobic jar to which carbon dioxide gas packs were added. The cultures were incubated at 37^{o}C. The other endocervical swab was used for Chlamydia trachomatis testing using the Chlamydia antigen detection kit (Rapid detection method). Urine sample was also obtained by mid-stream urine. This was explained to the participants and the sample was collected with a sterile wide mouthed, plastic jar with tight fitting. The urine sample collected was immediately sent to the laboratory for processing within 2 hours of collection or was kept refrigerated at 4^{o}C, until delivery to the laboratory. The urine sample was processed no longer than 18 hours after collection.

Sample Processing

A wet mount was prepared by mixing the vaginal sample with saline on a glass slide after which a cover slip was used to cover the smear. The smear was prepared by gently rolling rather than smearing a swab over the glass slide. A diluted preparation is preferred to ensure separation of cells which may otherwise be clumped together. Examination at magnification of × 400 for the presence of Trichomonas vaginalis with typical movement, budding yeasts and clue cells for bacteria vaginosis was done.

A normal vagina contains predominantly lactobacilli and fewer than 5 leukocytes per field. Presence of clue cells; which are epithelial cells surrounded by bacteria giving a characteristic stippled appearance on examination was also searched but none was seen. The presence of clue cells is a sensitive and specific diagnostic test for bacterial vaginosis.[12] Presence of polymorphonuclear leukocytes was also searched. This is because presence of more than 10 polymorphonuclear leukocytes is a reasonably good indication of mucopurulent cervicitis, most often due to Neisseria gonorrhea and /or chlamydia

trachomatis. Gram stain was also done by preparing the smear by using a sterile bacteriological loop.

For the urine sample: using a sterile Pasteur pipette (one for each sample) a drop of well mixed, uncentrifuged urine was dropped on a glass slide. This was examined under oil-immersion lens (×600 or more) for the presence or absence of bacteria, polymorphonuclear leucocytes and squamous epithelial cells. One or more bacterial cells per oil immersion field imply that there are 10^5 or more bacteria per milliliter in the specimen. The presence of one or more leucocytes per oil immersion field is further indication of UTI.[12] For Culture -1ml of urine was inoculated into CLED culture medium using a calibrated loop technique. The plate was incubated over night at 35^{o}c and examined after 18-24 hours for growth. The presence of 10^4 colony forming unit in a clean catch urine specimen is clinically relevant for the diagnosis of urinary tract infection.[12]Gram staining was also done for urine sample microbial identification.

Principle of the Test

Microscopy analysis-Smear of specimen on glass slide and observation under the microscope,
×10 or × 40 magnifications was used.

Culture-Specimens were cultured on specific laboratory culture medium and gram staining was done for identification. Samples that developed bacteria isolates were subjected to antibiotics sensitivity analysis using the disk diffusion methods.[15]

Ethical Clearance

Institutional ethical clearance was obtained from the Ethical Committee of the hospital. Informed, written and signed consent was obtained from participants by way of a consent form, after the purpose and the procedure of the study had been explained. After participating in the study participants with PROM were then managed according to the departmental protocol.

RESULTS

Over the period of study 3698 deliveries were recorded and there were 387 cases of patients with PROM giving the prevalence of 10.4%.
Table 1- shows the socio-demographic and obstetric characteristics of the participants. The cases and controls were matched for age (p-value 0.000), parity (p-value 0.000) and gestational age (p-value 0.000). The mean age, gestational age and parity were 27 ± 6 years, 33±.3 weeks and 2 respectively. The study population consisted mainly of the parity group 1-4 (55%) of the participants. Term PROM (37-42 weeks) recorded the highest frequency

Table 1. Socio demographic and obstetrics characteristics of the participant

Sociodemographic Characteristic	Cases (%)	Control (%)
Age		
<19	6 (4.6)	5 (3.9)
20 – 24	38 (29.5)	39 (29.9)
25 – 29	34 (26.3)	35 (27.3)
30 – 34	31 (24.0)	30 (23.3)
35 or more	13 (15.6)	13 (15.6)
Total	129(100)	129(100)
	χ^2 = 0.17, P= 0.99	
Parity		
0	38 (29.4)	34 (26.4)
1-4	68 (52.7)	71 (55.0)
5 or more	23 (17.8)	24 (18.6)
Total	129(100)	129(100)
	χ^2= 0.31, P=0.86	
Social Class		
Low class	16 (12)	7 (5)
Middle class	54 (42)	39 (30)
High class	59 (46)	83 (65)
Total	129 (100)	129 (100)
	χ^2=9.99, P = 0.007	

(37.9%). Majority of the participants were married 127(98.4%) and 128 (99.2%) in cases and control respectively. The control group consisted predominantly of high socioeconomic status 83(65%) while the participants with PROM consist of predominantly middle and high socioeconomic status 54(42%) and 59(46%). About 16 (12%) of the participants with PROM were of low socio-economic status as compared to 7 (5 %) of participants without PROM.

Table 2: Outcome of genital culture in the study group

	Cases N (%)	Control N (%)	Total	OR 95% CI	P-Value
Positive culture	102(79.1)	7 (5.5)	109.	1.627 (1.281- 1.893)	0.035
Negative culture	27 (20.9)	122 (94.5)	149		

Table 2- Shows outcome of genital culture in participants, positive culture was seen in 102 (79.1%) and 7 (5.5%) of cases and control respectively (p-value 0.035) (OR 1.627, 95% CI: 1.281-2.273) and negative culture seen in 27 (20.9%) and 122 (94.5%) of cases and control respectively. The outcome of culture was between

the participants with PROM and those without PROM was statistically significant.

Figure 1a shows the percentage of various microbial organism isolated from the genital tract of participants with PROM. *Candida albicans* accounted for 32 %, *Staphylococcus aureas* 16 %, *Streptococcus Spp.* 15.5%, E. Coli for 5.4%, *Klebsiella* 3.9 %, *Pseudomonas* aurogenosa 3.9% and *Proteus mirabilis 2.3% of isolates*.

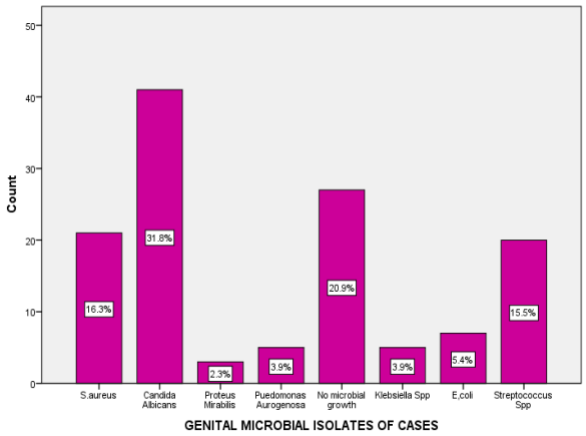

Figure 1a. Percentage of genital microorganisms isolated from participants with PROM

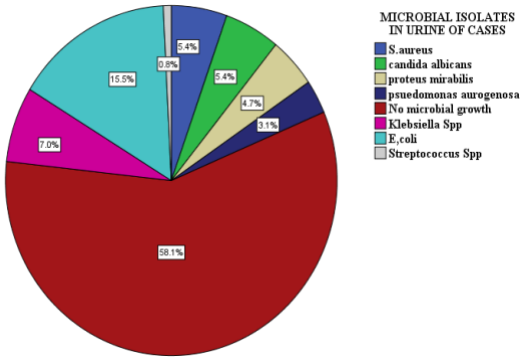

Figure 1b. Percentage of urine microorganisms isolated from participants with PROM.

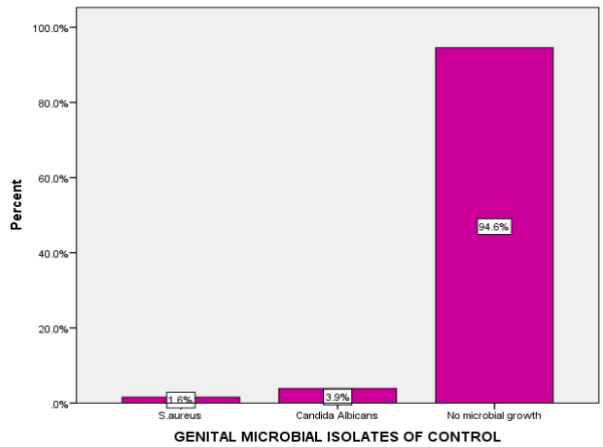

Figure 2a. Percentage of genital microorganisms isolated from participants without PROM (control)

Figure 1(b) shows the percentage of urine microbial isolates from participants with PROM; *Escherichia coli* accounts for 15.5%, *Staphylococcus aureus* accounts for 3.9%, *Candida albicans* accounts for 5.4%, *Pseudomonas aerogenosa* accounts for 5%, *Klebsiella* accounts for 7%, *Proteus mirabilis* accounts for 4.7%%, and *Streptococcus spp* accounts for 0.8% of the urinary isolates.

Figure 2a shows the percentage of genital microbial isolates in participants without PROM; *Candida albicans* accounted for 3.9% and *Staphylococcus aureus* for 1.6%.

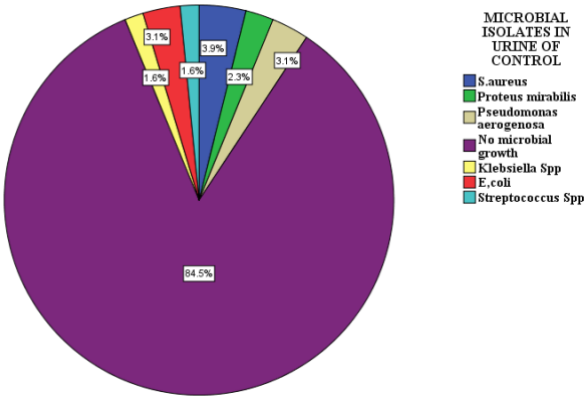

Figure 2 b. Percentage of urine microorganisms isolated from participants without PROM (control).

Figure 2b shows the percentage urinary microbial isolates in participants without PROM;
Staphylococcus aureus accounted for 3.9%, *Klebsiella* 1.6%, *Escherichia coli*, 3.1%, *Pseudomonas aurogenosa* 3.1%, *Proteus Mirabilis* 2.3% and *Streptococcus spp* for 1.6%.

DISCUSSION

Prelabour rupture of membranes from this study accounted for 10.4% of total deliveries. This was similar to reported incidence of 10.7% worldwide.[13]However, is higher than what was obtained in Southern Nigeria where incidence of 5.7%[14] was obtained. This difference may be attributed to the fact that their study only involved preterm Prelabour rupture of membranes.

This study tried to identify the genitourinary microbial isolates in subjects with PROM and in those without PROM. Evidence for bacteriological cause for PROM has been established.[15] *E. coli*, B-hemolytic *Streptococci, Chlamydia trachomatis*, *Neisseria gonorrhea* and

Gardnerella vaginalis are microorganism implicated in the aetiology of PROM. [15,16] Similarly in this study significant number of microorganisms were cultured in subjects with PROM (79.1%) compared to subjects without PROM (5.5%) (p-value of 0.035) and this was consistent with the findings of Aboyeji,[17] Salou,[18]Kennedy[19]and Eleje.[2] Normal vaginal flora however consisted of predominantly *Lactobacillus* and fewer than 5 leukocytes per power field.[12]

It was observed in this study that there was a significant risk of developing PROM in those with low socioeconomic status, this was consistent with studies by Choudhary[20]and spinello[21] where they quoted that maternal low socioeconomic status is a strong independent predictor of PROM due its association with bacterial vaginosis. Similarly, Ganjoel et al[22] analyzing for risk factor for bacterial vaginosis found that low socioeconomic status was a significant risk factor for bacterial vaginosis, and recent studies have reported the relation between PROM and bacterial vaginosis.[23]

Significant amount of candida albicans was isolated from the subjects with PROM (31.8%) compared to subjects without PROM (3.9%). This is consistent with the findings of Salou[18] and Aboyeji,[17] and also in a study in Kano where *Candida albicans* was isolated in 5.0% and 10.0% among the cases and control respectively.[8] However, this is not consistent with findings of Eleje[2] and Karat[24] that did not demonstrate the association of *Candida albicans* with PROM. Nakubulwa[25] did not also demonstrate an association between PROM and *Candida albicans,* and concluded that *Candida albicans* seems to have a protective effect on PROM. The possible association of candidiasis and PROM is not clear. However, evidence of release of inflammatory cytokines during infestation and possibly leading to rupture of membranes was postulated.[26] Roberts[27] demonstrated indirect evidence that candidiasis increased risk of membrane rupture in a study where use of antifungal medication reduced preterm labour and PROM. These reasons with the added reduced immunity of pregnancy and our tropical environment[8] could possibly be the reason for the finding of *Candida albicans* in significant amount in patients with PROM in this study. Recent studies have linked *candida albicans,* HSV-1 and *Trichomonas vaginalis* with PROM.[28]

Staphylococcus aureus (16.3%) was also found in the genital isolates of subjects with PROM as compared to subjects without PROM (1.6%) in this study, however it was not consistent with the findings of Salou[18](5.7%) and Eleje[2](29%) but similar to findings of Aboyeji[17] (18.7%). Some previous studies did not associate *Staphylococcus aureus* with PROM.[29]However, Karat[24] and colleagues found a significant association between *Staphylococcus aureus* and PROM. Vaitkiene[30]and colleagues also found isolation of *Staphylococcus aureus* and significant increased risk of PROM. *Staphylococcus aureus* is protease releasing and bacterial proteases can clearly impair the structural integrity of fetal membranes and predispose to rupture.[17]*Staphylococcus aureus* also elicits the production of proinflammatory cytokines, which could ultimately perturb maternal-fetal tolerance during pregnancy leading to PROM and chorioamnionitis.[31]

Streptococcus spp. was also isolated from genital tract of Subjects with PROM (15.5%) but not isolated from Subjects without PROM, the finding is not consistent with findings of Salou[18] (7.5%) and Eleje[2] (31.43%). Nakubulwa[25] also showed no association between *Streptococcus spp.* and risk of PROM. However, Laola[32] reported that *Streptococcus spp.* was isolated from most of culture obtained from patients with PROM in western countries. Other microbial isolates that were isolated from the genital tract of subjects with PROM included *Escherichia coli, Klebsiella spp, Pseudomonas aurogenosa* and *Proteus mirabilis*. These isolates are significant because none of them was isolated among the subjects without PROM. However, in a more recent Southeastern Nigerian study *Klebsiella spp* was the commonest organism isolated. [33]

Escherichia coli is the most implicated pathogen in urinary tract infection and is found to be the commonly isolated pathogen in PROM by Kennedy.[19]Celen[35] also found that *Escherichia coli* was the most common microorganism isolated in PPROM. Genitourinary infection is a risk for PROM and *Escherichia coli* is the most implicated pathogen in the etiological agent of urinary tract infection.[36,37]*Escherichia coli* was isolated from the urine sample of 20 (15.5%) subjects with PROM compared to 4 (3.1%) of subjects without PROM which was significant (p =0.035)

Klebsiella spp, Proteus mirabilis and occasionally *Pseudomonas aurogenosa* are gram negative urinary pathogens that are implicated in causing urinary tract infection[38] that could cause genitourinary infection which may eventually lead to PROM and the frequency of these isolates were found to be higher in the urine samples of the subjects with PROM than those without PROM.

CONCLUSION

Genitourinary tract infection was significantly associated with the risk of developing PROM with candida albicans and staphylococcus aureus being the commonest microbes cultured. Results of this study suggest that presence of genitourinary infection in patients identifies a group of pregnant women at increased risk of premature rupture of membranes.

Limitations

Some of the patients with PROM may have presented in established labour if they did not present early to the hospital and they may have been missed. Pathogens that are implicated in chorioamnionitis such as *Mycoplasma hominis* and *Ureaplasma urealiticum* could not be isolated in this study because of lack of facilities for their identification. Similarly, anaerobes could not be isolated from this study because of the lack of facilities needed for their culture. Anaerobic gas packs needed for anaerobiosis for the growth of anaerobic organisms implicated in PROM could not be obtained and this led to the inability to culture anaerobic organisms in this study.

Recommendation

This study identified some microorganisms implicated in PROM. However, further studies would be needed to identify other implicated pathogens in PROM that were not possible to isolate in this study. Also antimicrobial sensitivity pattern should be done in other to assist in choosing the most appropriate antibiotics in the management of PROM in this centre. *Candida albicans* was significantly isolated from the cases however; further study with a larger population needed be done to determine its association with PROM.

Conflicts of interest

I declare that there is no conflict of interest in this study.

REFERENCES

1. Omneya M.O, Mohammed E. Can vaginal washing fluid, creatinine and qualitative β-hcG diagnose suspected premature rupture of membranes? Open Journal of Obstetrics and Gynaecology.2014;(4) : 967-972.
2. Eleje GU, Ezebialu IU, Umeobika JC, Eke AC, Ezeama CO, Okechukwu ZC. Pre-labour rupture of membranes at term: a review of management in a health care institution. Afrimedic Journal 2010;1(2): 10-14
3. Kimberly BF, Chad AG, Carla ER, Rex CB, Liping F, Lan L et al. Bacteria localization and chorion thinning among preterm premature rupture of membranes. PLoS One .2014;9 (1):1-10.
4. Yeung SW, Sahota DS, Leung TY. Comparison of the effect of penicillin versus erythromycin in preventing neonatal group B streptococcus infection in active carriers following preterm premature rupture of membranes. Taiwan J Obstet Gynaecol 2014; 53:210-4.
5. Kenyon S, Boulvain M, Neilson JP. Antibiotics for preterm rupture of membranes. Cochrane Database syst Rev 2013; (12): CD001058.
6. Al-Riyami N, Al-RuhelliI, Al-Shezaw F and Al-Khabori M. Extreme preterm premature rupture of membranes: Risk factors and feto maternal outcomes. Oman Med J 2013 Mar;28 (2): 108-111.
7. Payam D, Xavier B, Francesc F, David M, Rob B, Marta C et al. Air pollution and preterm premature rupture of membranes: a spatiotemporal analysis. American Journal of Epidemiology October 14, 2013,179(2) 200-207
8. Habib RU, Rabiu A, Mohammed Z, Abiodun OO. Microbial isolates and antibiotic sensitivity pattern among women with early preterm spontaneous Prelabour rupture of fetal membranes in a Nigerian teaching hospital. Tropical Journal of Obstetrics and Gynecology; 2019; 36(5): 378-386.
9. Li-nan Zang, Ling –Li Zhang, Jing Shi, Li-Ling Gu, Wende Grogan, Mary M. Gargano, Chen-chen. The primary microbial pathogens associated with premature rupture of the membranes in China: a systemic review. Taiwanese Journal of Obstetrics and Gynecology 53(2014) 443-451
10. Araoye M O: Subject Selection in Research Methodology in Health and Social Sciences. Unipress, Nigeria 2002; p: 120.
11. Emechebe C. I , Njoku C.O , Anachuna K, Udofia U: Determinants and complications of Prelabour Rupture of membranes (PROM) at the University of Calabar Teaching Hospital (UCTH), Calabar, Nigeria. Scholars Journal of Applied Medical Sciences (SJAMS) sch. J. App. Med. Sci., 2015; 3(5B): 1912-1917.
12. Gina Crosley-Corcoran. Preterm premature fetal Membranes: http://crosley-corcoran .com/writing-samples/preterm-premature–rupture-of-membranes-a-2015-literature-review/assessed 6/1/2016.
13. Jun S, Keisuke I, Toshiko K, Akiko Y, Shusaku H, Nobuaki. Three- year outcome of very low birth weight infants in Conservatively managed premature rupture. Open Journal of Obstetrics and Gynaecology; 2015;5: 70-78.
14. Therpe N. Post Pregnancy genital tract and wound infection: Midwifery Women Health 2008; 53(3): 236-246.
15. Vandapitte J and Verhaegen J – Urine and sexually transmitted diseases. Basic procedures in clinical bacteriology. World Health Organization, Geneva 2003.
16. Ashley S. R. Late Pregnancy complications. In: Current Diagnosis and Treatment Obstetrics and Gynaecology.11thEd. New York (NY) McGraw Hill Companies Inc.; 2013: 250-266.
17. Aboyeji AP, Abdul MA, Ijaiya Ma, Nwabuisi C and Ologe. The bacteriology of pre-labour rupture of membranes in a Nigerian teaching hospital. J Obs and Gynaecol, 25:8,761-764.
18. Salou M and Dossim S. Premature rupture Of the Membranes at the Sylvanus Olympio University Hospital of Lome, Togo: Microbial findings. American Journal of Infectious Diseases and Microbiology, 2015;3(6)152-156.
19. Kennedy O.O, Current microbial pattern of patients presenting with Prelabour rupture of membranes at labour ward in Kenyatta National Hospital. MMED. Thesis, UON .2009. Unpublished.
20. Choudry M, Rathore S B, Chowdry J , Garg S. Pre and Post Conception risk factors in POM. Int J Res Med Sci 2015 Oct; 3(10): 2594-2598.
21. Spinello A, Nicola S, Pia IK. Epidemiological correlates of preterm premature rupture of membranes. Int J Gynae Obstet.1994: 47:7-11
22. Ganjoei TA. Risk factors for bacterial vaginosis in Women attending a hospital in Kerman, Islamic republic of Iran. La Revue de Sante de la Mediterrance Orientale 2005; 11(3):410-5.
23. Rani S, Mehra R, Gupta V, Huria A, Chander J. Vaginal flora in preterm premature rupture of membranes and their sensitivity to commonly used antibiotics. Asian Journal of Medical sciences. 2014;5(4): 58-60
24. Karat C, Madhivanan P, Knapp K, Poornina S, Jayanthi NV, Suguma JS et al; the clinical and microbial correlates of premature rupture of membranes. Indian J med microbial 2006;24: 283-285.
25. Nakubulwa S, Kaye D, Bwanga F, Tumwesigye N.M, Mirembe F.M. Genital infections and risk of premature rupture of membranes in Mulago Hospital, Uganda: a case control study. BMC Research Notes 2015 8: 573.
26. Roberts CL, Richard K, Kotsiou G, Morris JM. Treatment of asymptomatic vaginal candidiasis in pregnancy to prevent preterm birth: an open-label pilot randomized control trial. Pregnancy childbirth. 2011:11(1):518-27.
27. McGregor JA, French JI, Parker R, Draper D , Patterson E, Jones W et al . Prevention of premature birth by screening and treatment for common genital tract infections, results of a prospective controlled evaluation. American Journal of Obstetrics and Gynaecology.173:157-167.
28. Dava V, Staffen B, Vida M. Antenatal risk factors associated with preterm prelabour rupture of membranes: Acta Medica Lituanica.2002;(9) 3: 203-209.
29. Gahwagi MM, Busarira MD, Atia M. Premature rupture of membranes characteristics, determinants and outcome in Benghazi, Libya. Open Journal of Obstetrics and Gynaecology,2015;5:494-504.
30. Vaitkiene D, Bergtrom S. Management of amniocentesis in women with oligohydramnios due to membrane rupture: evaluation of cervical adaptor. Gynecol Obstet Invest. 1995;40 (1):28-31.
31. Rani S, Mehra R, Gupta V, Huria A, Chander J. Vaginal flora in preterm premature rupture of membranes and their sensitivity to commonly used antibiotics. Asian Journal of Medical sciences. 2014;5(4): 58-60
32. N'tesse L.C. Premature rupture of membranes: A retrospective study at department of Obstetrics and

Gynaecology .Kinshasa,2002, Available on line at http://www.memoireonline.com/07/08/1436/m. Accessed on 29/3/2017.
33. Eleje G.U, Adinma J.I, Ugwuanyi D.C, Ikechebelu J.I, Okafor C.I, Ezeama C.O et al. Genital tract microbial isolate in women with prelabour rupture of membranes in resource constrained community setting. Journal of Obstetrics and Gynaecology, July 2015; 35:465-468.
34. Calen S, Tamizhan RO, Eyi E, Seckin T, Saygan S, Ozkan M. Comparison of the cervical culture results in preterm labour and premature rupture of membranes. Erciyes Medical Journal 2012. 34:174-17.
35. Beazley D. Special circumstances affecting labour. In Whitfield CR, Editor. Dewhurst's Textbook of Obstetrics and Gynaecology for postgraduates. London: Blackwell. Pp. 327- 332.
36. Delzell HR, Lefevre ML. Urinary tract infections during pregnancy. Am Fam Physician 2000; 10:1-10.
37. Margrethe M, Borch K, Thomsen A.C, Kristen D, Zdravkovic M. Rupture of fetal membranes and premature delivery associated with group B streptococci in urine of pregnant women. The lancet.1984.324(8394):69-70.
38. Kallirhoe K, Dimitrios D, Michail K, Apostolos Athanasiadis and Loannis K Urinary tract infection during pregnancy: Current problem on a multifaceted Problem, Journal of Obstetrics and Gynaecology.2018, 38:4,448-45