Microbiological Profile of PROM in a Tertiary Care Centre in Mangalore, Karnataka State

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ABSTRACT

Background: Premature rupture of the fetal membranes (PROM) is associated with 30-40% of premature births and is an important cause of perinatal morbidity and mortality. Limited data are available for India, but studies from other parts of the world have shown the etiology of the condition to be multifactorial. Infection has been cited as a major cause of membrane damage.

Aim: To determine the infective etiology and its association with PROM.

Materials & Methods: A total of 104 cases of PROM that presented after 37 completed weeks' gestation were screened for infections by wet mount, Gram stain & routine culture of high vaginal swabs. Results: The most common pathogen encountered was Candida spp. Other organisms isolated were Klebsiella pneumoniae, Staphylococcus aureus, Group B Streptococcus, Acinetobacter and a rare case of Streptococcus pneumonia all of which were significantly associated with PROM.

Conclusion: It is evident from this study that diagnostic accuracy is the key to successful management and improved perinatal outcome in cases of PROM. Newer techniques and the search for difficult to culture pathogens need to be analyzed to widen the microbiological scope of describing PROM.

Key Words: Premature rupture of membranes, Premature births, Perinatal morbidity.

INTRODUCTION

Premature rupture of membranes (PROM) refers to a patient who is beyond 37 weeks' gestation and has presented with rupture of membranes (ROM) prior to the onset of labor. [¹] A whole host of cultivable and non-cultivable microorganism plays an important role in PROM. Apart from infective causes there are also other associated factors like hydramnios, incompetent cervix, placental abruption and amniocentesis. [²] Very few studies were
available from this part of the country regarding association of bacterial infections in PROM. Hence we made an attempt to study the relation between bacterial infections and PROM in our institute.

**MATERIALS & METHODS**

A hospital-based prospective case study of 104 pregnant women presenting with complaints of leakage of liquor, greater than 37 completed weeks of gestation. The period of study was from February 2012 to July 2014 in collaboration with the Department of Microbiology.

**Inclusion criteria:** Gestational age > 37 weeks; diagnosis of PROM confirmed by clinical finding of posterior vaginal pool, vaginal pH, cervical dilation of < 3 cms; singleton pregnancy; and clear liquor.

**Exclusion criteria:** Woman in labor; mal-presentations; multiple gestation; medical disorders; and a history of cervical encirclage.

After ethical clearance, data were collected from all eligible participants who gave informed consent. A structured questionnaire collected data on socio-demographic and other relevant variables. Collected biological specimens included liquor from the posterior fornix of the vagina, peripheral part of the amniotic surface of the placental membrane. The samples obtained were collected in sterile containers labeled and transported to the laboratory without delay. A wet mount preparation was done for bacteria, fungi and parasites like Trichomonas. Gram stain smears were assessed by Nugent’s criteria for bacterial vaginosis. Culture and identification were carried out as per the standard recommended procedures for aerobes, anaerobes and fungi. The primary culture media employed for bacteria was Chocolate and MacConkey’s agar. Samples were also inoculated into Robertson cooked media (RCM) for 48 hrs for anaerobes. Potential anaerobes were subcultured onto appropriate media and grown anaerobically in the Gas Pak system and later identified by Gram staining characteristics and standard recommended identification procedures (Hemolysis on blood agar, catalase, RCM meat pieces color change, Penicillin 2 units disc sensitivity, Indole test and growth in 20% bile). Sabouraud’s dextrose agar culture and identification was done if any fungal elements were detected by wet mount or Gram stain. Reliance was placed on solid media so that only those organisms present in appreciable numbers would be recorded. Coagulase negative staphylococci, diphtheroids, lactobacilli and scanty growth of faecal Streptococci were ignored for the purpose of this study. Data obtained was entered in MS Excel software and analyzed statistically.

**RESULTS**

Majority of the cases were in the age group of 18-30yrs (Table 1). 10 (9.6%) cases of PROM positively correlated with women residing in rural areas and coming from a lower socio-economic background. Our study had 87 (83.6%) primi and 17 (16.3 %) multigravida cases. 74 (71.1 %) cases were normal delivery and 30 (28.8 %) were posted for cesarean section. A previous history preterm labor was seen in 6 (5.76 %) cases with and 9 (8.6 %) cases of urinary tract infections. 6 women with PROM reported a previous history of PROM in earlier pregnancies. Good maternal outcome was seen in 96 (92.3 %) cases. 56 (53.8 %) cases had pus cells in their vaginal fluid indicating presence of genital tract infection but were treated promptly and discharged on a healthy note. The duration of PROM was < 24 hrs in 84 (80.7%) cases and > 24 hrs in the remaining 20 (19.2%). The pathogens detected are specified in
(Table 2) were significantly associated with PROM and a rare isolate of Streptococcus pneumoniae was identified.

Table 1: Age group distribution of the PROM cases studied.

<table>
<thead>
<tr>
<th>Age Group (In yrs)</th>
<th>Total number of cases (n=104)</th>
</tr>
</thead>
<tbody>
<tr>
<td>18 - 30</td>
<td>87</td>
</tr>
<tr>
<td>31 - 40</td>
<td>17</td>
</tr>
</tbody>
</table>

Table 2: Pathogens isolated from the cases of PROM.

<table>
<thead>
<tr>
<th>Pathogen isolated</th>
<th>Total number (n=28)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Candida species</td>
<td>15</td>
</tr>
<tr>
<td>Klebsiella pneumoniae</td>
<td>4</td>
</tr>
<tr>
<td>Staphylococcus aureus</td>
<td>4</td>
</tr>
<tr>
<td>Acinetobacter species</td>
<td>2</td>
</tr>
<tr>
<td>Group B Streptococcus</td>
<td>2</td>
</tr>
<tr>
<td>Streptococcus pneumoniae</td>
<td>1</td>
</tr>
</tbody>
</table>

DISCUSSION

PROM is characterized by rupture of membranes before onset of labor. PROM occurs in 10% of all deliveries and results in the loss of normal protection of fetus and intrauterine contents from bacterial invasion. A number of factors have been associated with PROM including infections, anatomic, and pregnancy-related factors. However in certain cases the cause remains idiopathic. PROM tends to recur in subsequent pregnancies hence offers an opportunity for prevention. Infections commonly seen in these cases involve Trichomonas, Gonococcus, Chlamydia and Group B streptococci. Staphylococcus aureus, Trichomonas vaginalis and microorganisms that cause bacterial vaginosis secrete proteases that degrade collagen and weaken the fetal membranes, leading to PROM. Other conditions like bacterial vaginosis (BV), urinary tract infection (UTI), cervical incompetence and procedures by unskilled professionals like amniocentesis pose a risk for PROM.

Women from rural background and low socioeconomic status and malnutrition show a greater preponderance for PROM as was evident in our study. Studies have shown that malnourished women have a decreased level of host defense factors regularly present in amniotic fluid so infectious agents such as E. coli and S. aureus can play a larger role. PROM being a multifactorial entity, findings from our study revealed the role of exogenous risk factors such as UTI, previous preterm delivery and bacteria significantly associated with the condition which was similar to the studies by Karat et al. It still not clear if these above mentioned conditions directly cause PROM. There is no documented evidence to suggest differences in PROM in primi and multigravid cases. However studies by Mikam et al suggest that the number of organisms detected in the vagina provides sensitive and specific prediction of intrauterine infection with aerobic or anaerobic organisms in pregnancies with PROM. In our study 16 pathogens were isolated. Maternal outcome was good in 96 (92.3 %) cases.

The time interval between the rupture of membrane and delivery and antibiotics given to mother of PROM does not always totally protect neonates from infection, as was evident in our study. Our prophylaxis protocol of 500 mg of Amoxycillin I.V thrice daily was given to those diagnosed with PROM but the antibiotic cover was not enough to contain a safe neonatal outcome. Ofloxacin and Azithromycin were found to be effective against all the isolated pathogens for the treatment aspect. After the culture & sensitivity report, treatment was continued or changed based on sensitivity reports. The concept of heavy colonization of the genital flora can be a risk factor or considered as a predictor of PROM. Anaerobes are considered potentially pathogenic, as they have been frequently isolated in patients with genital infections.

Infections of genital tract are one of the possible causes for PROM. One possible
mechanism is ascending infection results in replication of bacteria in placenta, decidua and membranes. Others like *Group B Streptococci*, *Staphylococcus aureus*, *Trichomonas vaginalis* and microorganisms that cause bacterial vaginosis secrete proteases that degrade collagen and weaken the fetal membranes, leading to PROM. [11] Candidiasis was a common condition among our cases and other predominant isolates like *Klebsiella*, *Staphylococcus*, *Acinetobacter* are proven pathogens in PROM. *Group B Streptococcus* isolated in our case could have been a colonizer not directly associated with PROM in our case. We had a rare isolate of *Streptococcus pneumonia* that is not a part of the resident vaginal flora. However, in some women it can be a transient part of the vaginal flora, and pelvic infection can occur if a predisposing condition exists (e.g., use of an intrauterine contraceptive device, a recent birth, or gynecologic surgery). However the mother responded well to antibiotics and the neonate had a good outcome. [12]

In 10% of our cases we were not able to isolate organisms present as an infective etiology evident in our direct smears of the vaginal swabs. This may be due to infections with *Chlamydia*, *Mycoplasma* or *Ureaplasma*, which are noncultivable by routine methods. Gram negative cocci were not seen in any of our smears and there were no cases of bacterial vaginosis as was a similar finding in a study by Ziaei S et al who found no significant association between BV and PROM. [13] It’s true that a wide variation in the results from various studies might be due to differences in diagnostic methods and in study populations. [11] Certain limitations in this study could have shown a relatively low incidence as sample size was less and pathogens were isolated only by routine culture, but not by antigen detection and molecular methods or special tests due to financial constraints.

**CONCLUSION**

A significant association was observed between the PROM cases and infections encountered, with *Candida species* being the most common isolate. BV was not associated in these PROM pregnancies. Diagnostic accuracy is the key to successful management and improved perinatal outcome in cases of PROM. Newer techniques and the search for difficult to culture pathogens need to be employed to widen the microbiological scope of describing PROM.

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