BACTERIOLOGICAL AND CYTOLOGICAL STUDIES OF ENDOMETRITIS IN BUFFALOES

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ABSTRACT
Endometritis is most common problem in buffaloes affecting productivity and fertility. For the present study, thirty two endometrial discharge samples were collected from infertile buffaloes after thorough rectal examination and in absence of palpable uterine abnormalities. Uterine aspirate was collected aseptically and carefully by sterile uterine catheter for bacteriological and cytological examination. Bacteriological examination of uterine aspirate samples yielded majorily Salmonella (34.37%) followed by Staphylococcus (28.12%), E.coli (21.87%) and Pseudomonas (15.62%). Endometrial cytological smears in nine cases revealed more number of neutrophils along with epithelial cells indicating acute endometritis. Lymphocytes and few polymorphonuclear cells were observed in addition to epithelial cells in eleven samples indicating sub acute endometritis cases. Twelve endometrial cytology cases revealed lymphocytes, few plasma cells in addition to epithelial cells and mucin strands indicating chronic endometritis.

Key Words: Endometritis, Endometrial Cytology, Infertility in Buffaloes

INTRODUCTION
Incidence of endometritis in Andhra Pradesh is the second most commonly encountered reproductive disorder. Altered uterine environment in sub clinical or clinical endometritis contribute to failure of fertilization or early embryonic death (Singh et al., 1983; Hussain and Daniel, 1990). Generally, uterine infections are classified as nonspecific infections (Bekana et al., 1994; Bonnett and Martin, 1995; Leblanc et al., 2002) and are considered to be main cause of endometritis (Elliot et al., 1968; Dholakia et al., 1987) and repeated conception failure (Sharma et al., 1988; Singia et al., 1993; Singh et al., 1996). Pathogenic microorganisms in uterus cause inflammation, histological lesions of endometrium (Bonnett et al., 1991; Sheldon et al., 2003; Azawi, 2008) affects productivity and fertility (Bondurant, 1999).

The indiscriminate use of antibiotics resulted into non-recovery of animals due to development of antimicrobial resistance. Therefore isolation and identification of infectious agent is prerequisite for adopting suitable therapeutic strategies (Prajapati et al., 2006). Combination of bacterial culture and endometrial cytology were one of the commonly used practical methods for diagnosing endometritis (Neilsen, 2005).

MATERIALS AND METHODS
A total of 32 infertile buffaloes with history of conception failure or abnormal discharges were identified and utilized for uterine aspirate collection after thorough rectal examination of reproductive tract and in absence of palpable uterine abnormalities. After thorough clinical examination at estrus phase, a fluid (2-3 ml approximately) was aspirated from uterine lumen using sterile uterine catheter and transferred into sterile tubes, then transported to laboratory aseptically and care fully at 4°C. The fluid was then smeared and stained by Papanicolaou and Giemsa staining methods (Humason, 1972). Swabs from the uterine discharge samples were collected in sterile conditions and kept in nutrient broth. After 24 hrs, the growth
was streaked on different suitable Medias for isolation of bacterial organisms as described by Noel et al., (1984).

**RESULTS AND DISCUSSION**

The results of bacteriological examination of 32 uterine biopsies from infertile buffaloes are presented in Table.1. Bacteriological examination of uterine aspirate samples from infertile buffaloes revealed different single type of bacteria in 14 cases (43.75%) and mixed bacterial isolates in18 cases(56.25%) of samples. The over all frequencies of bacteria isolated are *Salmonella* (34.37%) followed by *Staphylococcus* (28.12%), *E.coli* (21.87%) and *Pseudomonas* (15.62%). Prajapati et al., (2006) isolated *Staphylococcus* (36.17%), *Bacillus* (14.8%), *E.coli* (12.76%) *Proteus* (10.65%), *Streptococci* (10.6%), *Pseudomonas* (8.5%) and *Salmonella* (6.38%). Bacteria obtained from acute endometritis cases were *Salmonella E.coli, Staphylococci and Klebsiella*. From Sub acute endometritis cases *Salmonella E.coli, Pseudomonas, Pasteurella and Staphylococci* were isolated. Bacteria obtained from chronic endometritis were *Salmonella, E.coli, Pseudomonas, Staphylococci and Streptococci*.

**Table 1: Isolation of bacteria in endometrial biopsies of infertile buffaloes**

<table>
<thead>
<tr>
<th>Condition (Infertile animals)</th>
<th>No. of samples</th>
<th>Positive for bacterial isolates</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Acute endometritis</td>
<td>9</td>
<td>Single (43.75%)</td>
<td>5</td>
</tr>
<tr>
<td>Subacute endometritis</td>
<td>11</td>
<td>5</td>
<td>6</td>
</tr>
<tr>
<td>Chronic endometritis</td>
<td>12</td>
<td>05</td>
<td>7</td>
</tr>
</tbody>
</table>

Figure 1: Acute endometritis : More number of neutrophils in endometrial cytological smears Papanicolaou x 700

Figure 2: Acute endometritis : Endometrial cytology showing more number of neutrophils H & E x 700

Figure 3: Sub acute endometritis : Note few neutrophils and lymphocytes in endometrial cytology smear H & E x 700

Figure 4: Chronic endometritis : Plasma cells and lymphocytes in endometrial cytology smears Giemsa x 700
Javed and Khan (1991) isolated corynebacterium, Bacillus and Proteus in addition to E.coli, Staphylococcus and Streptococcus in mild endometritis. Messier et al., (1984), Singla et al., (1991) and Azawi et al., (2008) isolated Escherichia coli as the predominant bacterial isolate in majority of endometritis cases. E.coli was the predominant organism isolated in cases of sub acute endometritis. Staphylococcus was found to be predominant organism isolated in repeat breeder buffaloes according to (Ahmed and Bhattachryya, 2005; Prajapati et al., 2006; Gani et al., 2008). Uterine samples from chronic endometritis revealed Staphylococci as predominant organism. Shukla and Sharma (2005) isolated E.coli (18.75), Corynebacterium (15.64%) Bacilli (14.06%), Staphylococci (12.05%), Pseudomonas (12.50%) and Micrococcus (3.12%) in repeat breeding cows.

Cytological examination of nine endometritis cases revealed more number of neutrophils in addition to epithelial cells (Figure 1&2). Similar observations were made by Solmon Raju et al., (2006) and Virmani et al., (2007). Cytological examination of eleven endometritis cases revealed more number of lymphocytes and neutrophils indicating subacute endometritis (Figure 3). Similar observations were reported by Chapwanya et al., (2009). Endometrial cytology of twelve endometritis cases revealed lymphocytes and few plasma cells indicating chronic endometritis (Figure 4). Similar observations were made by Virmani et al., (2007).

REFERENCES
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