



Microbial Prevalence and Antimicrobial Sensitivity in Dairy Animals Suffering From Purulent Vaginal Discharge

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ABSTRACT

Uterine infection can be diagnosed by taking a swab from fornix vagina or directly from cervix and can be used for cytology and microbiological evaluation. In this study, vaginal swabs for culture and sensitivity test were obtained from 20 buffaloes and two cows which were shedding purulent pus discharge from their reproductive tract. The most isolated microorganism was *Escherichia coli* which were detected in 17 animals, followed by *Staphylococcus spp.* and *Streptococcus spp.* which was detected in 15 and 14 animals, respectively. *Klebsiella spp.* and *Bacillus spp.* were diagnosed in two animals whereas; *Pseudomonas spp.* and *Candida spp.* were also diagnosed in one animal. Regarding sensitivity tests, the most efficient antibiotics were Gentamicin 54.5% (12/22) followed by Ciprofloxacin 50% (11/22), Enrofloxacin 45% (10/22), Neomycin 45% (10/22), Amikacin 45% (10/22), Cefoperazone 36% (8/22), Levofloxacin 31% (7/22), Norfloxacin 27% (6/22), Chloramphenicol 27% (6/22) and Cephalexin 22.7% (5/22). Likewise, the most resistant antibiotics in descending order were Streptomycin 95.4% (21/22), Penicillin and Cloxacillin 90.9% (20/22), Amoxicilin, Moxifloxacin and Ampicillin 86% (19/22), Oxytetracycline and Ceftriaxone 81.8% (18/22), Cephalexin 72.7% (16/22), Levofloxacin 68% (15/22), Carbenicillin, Chloramphenicol, Kanamycin and Cefoperazone 63.6% (14/22), Tobramycin, Neomycin and Enrofloxacin 54.5% (12/22), Amikacin and Ciprofloxacin 50% (11/22), Cephotaxime 45.5 (10/22), Norfloxacin and Gentamycin 40.9% (9/22), Clindamycin and Septran 22.7% (5/22), Amoxiclav and Erythromycin 18 % (4/22). The present study concluded that the most prevalent microorganisms isolated from uterine discharge, were found to be resistant to beta-lactam antimicrobials as well, the study highlighted the significance of performing microbiological analyses as well as sensitivity tests prior to applying an antimicrobial therapy.

Key words: Bovine, uterine infection; microbiological study; culture sensitivity test.

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INTRODUCTION

Bacterial contamination of uterus is the most commonly observed after parturition with an incidence 80 to 100% during the first 2 weeks of puerperium period (Foldi *et al.*, 2006). A large number of bacteria are eliminated during the first 15 days postpartum, whereas the remaining bacteria cause uterine infections. Common bacteria isolated from cows with uterine infections are *Escherichia coli*, *Prevotella* spp., (Sheldon *et al.*, 2006) *Staphylococcus* spp., *Streptococcus* spp. or non-coliform aerobic gram-negative rods (Kaczmarowski *et al.*, 2004; Jadon *et al.*, 2005). For selection of an appropriate antimicrobial for treatment of postpartum uterine infection, it is necessary to know the susceptibility of the microbes to antibiotics. Antibiotic resistance has emerged as big problem in veterinary practice and indifferent use of antimicrobial agent is considered the most important factor for the emergence, selection and dissemination of antimicrobial agent-resistant bacteria (Takamtha *et al.*, 2013). Postpartum uterine infections can delay the regeneration of endometrium and disrupts the resumption of cyclic ovarian function which leads to the postponement of first insemination (AI), increase in number of inseminations per conception and thus calving interval is prolonged (Foldi *et al.*, 2006). Economic losses are also increased due to decrease in milk production, herd replacement, costs of drugs, veterinary services and labour cost (LeBlanc *et al.*, 2006; Getahun *et al.*, 2008; Momtaz *et al.*, 2010). The information related to the pathogenic organism and sensitivity test in buffalo is still not fully explored and validated. Therefore, the present study was conducted to find out causative organism and suitable antimicrobial agents that were highly sensitive in postpartum uterine infection of bovines.

MATERIALS AND METHODS

The present study was conducted on 22 post parturient animals that were referred to the Veterinary Clinical Complex, College of Veterinary Sciences, LUVAS, Hisar for diagnosis and treatment of uterine infections. A total of 22 animals (2 cows and 20 buffaloes) with age between 3 and 12 years old were included in the study. This is a clinical study performed in bovine for research purposes from the animals referred to the Veterinary clinics by the field veterinarian. All the animals had presence of abnormal vaginal discharge. Since all manipulations were clinically needed, permission from the Ethical Committee was not mandatory. A signed consent from owners to use data was obtained. Uterine discharge was collected from in front of cervix using sterile cotton swab. The samples were

collected and sent to the Laboratory for culture sensitivity test. Identification of colonies/microbial agent and sensitivity tests were performed in this laboratory.

The antibiotic discs of Enrofloxacin, Cephalexin, Clindamycin, Penicillin G, Carbinicillin, Streptomycin, Amoxicillin, Tobramycin, Septran, Ciprofloxacin, Oxytetracycline, Erythromycin, Norfloxacin, Chloramphenicol, Kanamycin, Ceftriaxone, Moxifloxacin, Levofloxacin, Ampicillin, Gentamycin, Cephotaxime, Neomycin, Amikacin, Cloxacillin and Cefaperazone were tested. The percentage of sensitive and resistant antimicrobials was analyzed as well as percentage of animals having particular microorganisms was also analyzed.

RESULTS AND DISCUSSION

The most isolated microorganism was *Escherichia coli* which detected in 17 animals, followed by *Staphylococcus* spp. which was observed in 15 animals and *Streptococcus* spp. was detected in 14 animals. *Klebsiella* spp and *Bacillus* spp. was diagnosed in two animals. *Psuedomonas* spp. and *Candida* spp was diagnosed in one animal (Table 1).

Table 1: Microbial combinations diagnosed in affected animals

Sr. No.	Combination of microorganisms diagnosed	Number of animals
1	<i>Streptococci</i>	1
2	<i>Staphylococci</i> and <i>Streptococci</i>	3
3	<i>Streptococci</i> and <i>E. coli</i>	3
4	<i>Staphylococci</i> , <i>Streptococci</i> and <i>E. coli</i>	6
5	<i>Staphylococci</i> and <i>E. coli</i>	3
6	<i>E. coli</i> and <i>Klebsiella</i>	1
7	<i>Staphylococci</i> , <i>E. coli</i> and <i>Klebsiella</i>	1
8	<i>Staphylococci</i> , <i>Streptococci</i> , <i>E. coli</i> , <i>Psuedomonas</i> and <i>Candida</i>	1
9	<i>E. coli</i> and <i>Bacillus</i>	1
10	<i>E. coli</i>	1
11	<i>Staphylococci</i> and <i>Bacillus</i>	1

All these microorganisms were diagnosed in combination in all animals. Regarding sensitivity tests, the most efficient antibiotics were Gentamicin 54.5% (12/22) followed by Ciprofloxacin 50% (11/22), Enrofloxacin 45% (10/22), Neomycin 45% (10/22), Amikacin 45% (10/22), Cefoperazone 36% (8/22), Levofloxacin 31% (7/22), Norfloxacin 27% (6/22), Chloramphenicol 27% (6/22) and Cephalexin 22.7% (5/22), whereas Ceftriaxone 18% (4/22), Moxifloxacin 13.6% (3/22), Cephotexime 13.6% (3/22), Cloxacillin 9% (2/22) have medium sensitivity and Clindamycin 9% (2/22), Penicillin 9% (2/22), Carbenicillin 4.5% (1/22), Streptomycin 4.5% (1/22), Septran 4.5%

(1/22), Kenamycin 4.5% (1/22), amoxiclav 4.5% (1/22) were the least effective sensitivity (Table 2). Likewise the most resistant antibiotics in descending order are given as Streptomycin 95.4% (21/22), Penicillin and Cloxacillin 90.9% (20/22), Amoxicillin, Moxifloxacin and Ampicillin 86% (19/22), Oxytetracycline and Ceftriaxone 81.8% (18/22), Cephalexin 72.7% (16/22), Levofloxacin 68% (15/22), Carbenicillin, Chloramphenicol, Kanamycin and Cefoperazone 63.6% (14/22), Tobramycin, Neomycin and Enrofloxacin 54.5% (12/22), Amikacin and Ciprofloxacin 50% (11/22), Cephotaxime 45.5 (10/22), Norfloxacin and Gentamycin 40.9% (9/22), Clindamycin and Septran 22.7% (5/22), Amoxiclav and Erythromycin 18 % (4/22) as per Table 2. In our study, Beta-lactam antibiotics are not efficient as a treatment for uterine diseases.

Table 2: Sensitivity and Resistance pattern of antibiotics observed in affected animals

Sr. No.	Antibiotic	Sensitive (n) number of animals	Resistant (n) number of animals
1	Enrofloxacin	10	12
2	Cephalexin	5	16
3	Clindamycin	2	5
4	Penicillin	2	20
5	Carbenicillin	1	14
6	Streptomycin	1	21
7	Amoxiclav	1	4
8	Amoxicillin	3	19
9	Tobramycin	3	12
10	Septran	1	5
11	Ciprofloxacin	11	11
12	Oxytetracycline	2	18
13	Erythromycin	3	4
14	Norfloxacin	6	9
15	Chloramphenicol	6	14
16	Kanamycin	1	14
17	Ceftriaxone	4	18
18	Moxifloxacin	3	19
19	Levofloxacin	7	15
20	Ampicillin	3	19
21	Gentamicin	12	9
22	Cephotaxime	3	10
23	Neomycin	10	12
24	Amikacin	10	11
25	Cloxacillin	2	20
26	Cefoperazone	8	14

Buffaloes and cows are highly susceptible to uterine infections after parturition because of housing conditions

and production demands, these animals are under stress which results in diseases like sub-clinical endometritis, clinical endometritis, metritis and pyometra are associated with sub-fertility and infertility and can result in longer intervals from calving to first conception or ultimately lead to involuntary culling of animals failing to conceive (Sheldon *et al.*, 2008). Normally the animals which do not have uterine infection should become pregnant within 90 days, in case of uterine infection, number of infection per conception and calving to conception interval increased.

In our study, Gentamicin (12) followed by Ciprofloxacin (11), Enrofloxacin (10), of antibacterial agents were found highly sensitive to bacteria found in uterine sample. These results were in agreement with some other observations where percent sensitivity to Enrofloxacin and Ciprofloxacin has been reported as 92 and 96%, respectively (Patel *et al.*, 2009; Mshelia *et al.*, 2014). Whereas Carbenicillin (1), Streptomycin (1), Septran (1), Kenamycin (1), were the least effective in our study. In contrary, Streptomycin (21), Penicillin and Cloxacillin (20) were found highly resistant. Sadig (2010) also reported that Gram positive bacteria were resistant to Cloxacillin. Generally, indiscriminate use of antibiotics leads to development of resistant strains due to bacterial mutation. In the field those antibiotics are used routinely and indiscriminately that are found highly resistant e.g. Oxytetracycline (18) whereas Amoxiclav and Erythromycin were found least resistant. Oxytetracycline was routinely used for uterine infection, especially after calving, via intrauterine infusion or intramuscular administration. This may be due to bacterial resistance to this antimicrobial agent and its requirement in high concentration to inhibit bacterial growth when tested in vitro (Risco *et al.*, 2007).

Brooks *et al.*, (2001) also found that *Staphylococcus aureus* was resistant to Penicillin which agrees with our findings. Gentamicin was among the highly sensitive antimicrobial agents against Gram negative bacteria (Sadig, 2010). In an investigation, conducted by Takamtha *et al.* (2013) reported that Gentamicin was highly sensitive (93%) to gram aerobic bacteria, while oxytetracycline (42%), amoxicillin (26%) and streptomycin (25%) were resistant. The results of our study were also similar to the investigation of Takamtha *et al.* (2013).

In our study, Gentamicin was the most sensitive and streptomycin was the most resistant for both gram-positive and gram-negative pathogens. The treatment of bacterial infections is increasingly complicated by the ability of bacteria to develop resistance to antimicrobial agents. Chandrakar *et al.*, (2002) has reported the resistance to penicillin by Gram positive bacteria. Due to intrauterine bacterial contamination and repair of the endometrium

following parturition, uterine inflammation is a normal and necessary component of the postpartum uterine involution process (Ahmadi *et al.*, 2006). However, in some of the postpartum cows, the inflammation exceeds the normal threshold and leads to uterine infection (Le Blanc, 2014).

Uterine infections have been associated with an increase in days open and lower conception rate in cows (Sheldon and Dobson, 2004). This may be due to suboptimal conditions for sperm transportation and storage, oocyte maturation and ovulation, zygote development, implantation and embryonic and fetal growth (Gilbert, 2011).

CONCLUSIONS

In buffaloes and cows, infection of uterus is common disorder which affect reproduction and production that causes huge economic losses to the farmer by increasing inter-calving period and infertility. Therefore, the need to look into the mitigating factors is high lightened and thereby minimizing losses. In our study, the most efficient therapeutic approach of the postpartum uterine infections was exposed. Uterine infections can be limited by systemic administration of Gentamicin antibiotic during postpartum infection. The *E. coli* was found to be most prevalent micro-organism in postpartum uterine infection. This information is useful for therapeutic treatment planning and antibiotic control.

CONFLICT OF INTEREST

The authors declare no conflict of interest among themselves.

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