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ANTIBIOGRAM OF PUS SAMPLE COLLECTED FROM UTERINE HERNIA OF A DOE WITH ABORTION VIS-A-VIS EMERGING HUMAN INCIDENCES : A CASE STUDY

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ABSTRACT

Aim: The present article reports on the bacteriological examination and antibiotic sensitivity test of pus sample collected from uterus of a doe suffering from uterine hernia.

Materials and Methods: The patient's sample was subjected to microbiological analysis and antibiotic sensitivity tests.

Results: The infecting bacterium was susceptible to the MIC of broad-spectrum antibiotics.

Discussion: Methicillin-resistant Staphylococcus aureus (MRSA) and vancomycin-resistant enterococci (VRE) are of particular concern as the common causes of nosocomial systemic and other infections in hospitalized patients. In the United States, approximately 60% of staphylococcal infections in the intensive care unit are now caused by MRSA, and percentages continue to rise. Outbreaks of hospital-acquired MRSA (HA-MRSA) are typically the result of clonal spread by MRSA being transferred from patient to patient, frequently using healthcare personnel as intermediaries. HA-MRSA strains are generally multidrug resistant. Vancomycin is the standard treatment for serious MRSA infections, but a few cases of vancomycin-resistant S. aureus (VRSA) have recently emerged in the United States. Community-acquired MRSA (CA-MRSA) is also increasing. Soft tissue infections are the most frequent presentations of CA-MRSA, but life-threatening invasive infections occur as well, including necrotizing pneumonia.

Conclusion: The present study indicated that the *Staphylococcus* spp. isolated from pus sample was sensitive to broad-spectrum antibiotics which were recommended for application in mixed preparations in divided doses.

Keywords: Bacteriological examination, pus, antibiotic sensitivity

INTRODUCTION

The purulent exudate 'pus' remains surrounded by a limiting membrane the pyogenic membrane (Tyagi and Singh, 2012; Sahoo and Ganguly, 2015). During pus formation, there occurs by a breach of surface of the skin or mucous membrane leading to the entrance of pyogenic microorganisms (Tyagi and Singh, 2012). Usually solitary pus containing external outgrowths are common in cattle and buffaloes (Thorat *et al.,* 2008). Hernia uterine inguinale is a rare condition often presenting within the first few years of life as an asymptomatic palpable mass in the inguinal/groin area. This type of hernia contains uterine tissue and may contain oviducts, ovaries, and rarely the bladder. Hernia uterine inguinale is a rare condition and an even more uncommon cause of pelvic pain, instead presenting as an asymptomatic palpable groin mass early in life. This has been reported most commonly in the literature as both persistent müllerian duct syndrome and male pseudo-hermaphroditism. It is most often seen in a phenotypically normal male infant having both testes and uterine tissue present. Few cases have been documented to occur in the female sex, the adult patient, or as a cause of pelvic pain. Abdominal and pelvic imaging is useful in the diagnosis of this condition because it may aid in identifying patients with coexisting mullerian malformations. This subset

may be at higher risk for hernia uterine inguinale, and, if presenting with complaints of pain or inguinal mass, it should likewise be considered in the differential diagnosis (Mandel, Beste & Hope, 2010)

Carbapenems were the most active drugs tested against most of the bacterial species. E. coli and P. mirabilis remained susceptible to most of the drugs tested. Mean rates of resistance to 9 of the 12 drugs tested increased with *Acinetobacter spp*. Rates of resistance to ciprofloxacin increased over the study period for most species. Ceftazidime was the only agent to which a number of species (*Acinetobacter. spp., C. freundii., E. aerogenes., K. pneumoniae., P. aeruginosa* and *S. marcescens*) became more susceptible. The prevalence of multidrug resistance, defined as resistance to at least one extended-spectrum cephalosporin, one aminoglycoside, and ciprofloxacin, increased substantially among ICU isolates of *Acinetobacter spp., P. aeruginosa, K. pneumoniae* and *E. Cloacae* (Lockhart *et al.,* 2007)

The prevalence and antibiotic susceptibilities patterns of bacterial isolates from pus samples collected from patients in a tertiary care hospital of Punjab, India were determined by Trojan *et al.*, (2016). *E. coli was the most prevalent pathogen (51.2%) followed by Staphylococcus aureus (21%), Klebsiellapneumoniae (11.6%),Pseudomonas aeruginosa (5.8%), Citrobacter spp. (3.5%), Acinetobacter baumannii(2.3%), Proteus mirabilis (2.3%), and Streptococcus spp. (2.3%).E.coli, K.pneumoniae, A.baumannii, and Citrobacter* isolates were resistant to multiple antibiotics including higher generation cephalosporins. S.aureus and Streptococcus isolates were sensitive to cloxacillin and vancomycin (Trojan *et al.,* 2016).

Zhang *et al.,* (2014) examined a total of 41 patients with severe intra-abdominal infection (SIAI) induced by abdominal trauma from which 123 abdominal pus samples were obtained. A total of 297 strains were isolated in which Gram-negative bacteria, Gram-positive bacteria and fungi accounted for 53.5 (159/297), 44.1 (131/297) and 0.7% (2/297), respectively. Anaerobic bacteria accounted for 1.7%. The five predominant bacteria were *E. coli, S. aureus, K. pneumoniae, Enterococcus faecalis* and *P. aeruginosa*. *E. coli* was highly susceptible to cefoperazone (91%) and imipenem (98%), while Gram-positive cocci were highly susceptible to teicoplanin (100%) and linezolid (100%). *S. aureus* was 100% susceptible to vancomycin and *K. pneumoniae* was highly susceptible to imipenem (100%) and amikacin (79%). *P. aeruginosa* was the most susceptible to ciprofloxacin (90%).

Gram-positive bacteria are common causes of bloodstream and other infections in hospitalized patients in the US and the percentage of nosocomial bloodstream infections caused by antibiotic-resistant gram-positive bacteria is increasing. The mechanisms of methicillin resistance are the same for CA-MRSA and HA-MRSA, but susceptibilities to non–â-lactam antibiotics often differ. CA-MRSA exhibits broader antibiotic susceptibility than does HA-MRSA (Rice, 2006).

Catarino *et al.,* (2015) investigated on a total of 130 women performed two consecutive self-HPV samples. Randomization determined which of the two tests was performed first: self-HPV using dry swabs (s-DRY) or vaginal specimen collection using a cytobrush applied to an FTA cartridge (s-FTA). After self-HPV, a physician collected a cervical sample using liquid-based medium (Dr-WET). HPV types were identified by real-time PCR.

The present study was conducted to identify the aetiology and the antibiotics/ antibacterial drugs which show sensitivity against the various pathogenic agents involved in the pus formation form the case of uterine hernia.

MATERIALS AND METHODS

The pus sample was collected by draining from auterine hernia case of a miscarried doe with macerated foetus presented for clinical examination at the Teaching Veterinary Clinical Complex (T.V.C.C.) of Arawali Veterinary College, Sikar during May 2017. The collected pus sample was then brought to the Department of Veterinary Microbiology of Arawali Veterinary College, Sikar for bacteriological examination and reporting.

The specimen was incubated overnight in nutrient broth medium. The pus sample was examined (Buxton and Fraser, 1977) by bacterial culturing on nutrient agar plate followed by staining by Gram's Method.

Antibiotic sensitivity test was carried out by Kirby-Bauer antibiotic disc diffusion assay method (Sinha, 2006) on Mueller-Hinton agar with certain modifications (Ganguly *et al.,* 2015) using antibiotic discs (Titan Biotech Ltd., Bhiwadi, Rajasthan, India) available at the department. The concentration of antibiotic in each filter paper disc was as per the specification of the manufacturer required for laboratory purpose. Then spread plate method of bacterial culture was done from the pus sample followed by its incubation at 37^0C for 24 h in a B.O.D. incubator installed at the department.

RESULTS

The pus sample was subjected to spread plate culture on Nutrient agar media plates. Grams' method of staining with the isolated pure colony revealed Gram positive cocci arranged in clusters or clumps when examined under the high power magnification of the compound microscope. To obtain pure bacterial colonies it was subcultured on Nutrient agar plates. It revealed the presence of circular, convex, glistening colonies with full regular edges after incubation. The bacteria were determined to be grouped under *Staphylococcuss*. (Cruickshank *et al.,* 1975; Finegold and Martin, 1982; Ananthanarayan and Paniker, 2009).

Antibiotic assay revealed the bacterial isolates to be highly sensitive to the minimum inhibitory concentration (MIC) of antibiotics, amikacin (30 mcg), tetracycline (30 mcg) and ceftriaxone (30 mcg) respectively. The degree of sensitivity was determined on the basis of zone of inhibition produced by the isolated bacteria after exposure to the particular antibiotics and after comparison with the minimum inhibitory concentration of the respective antibiotic.

DISCUSSION

The outcomes of the present study were in concurrence with these findings of Ayub *et al.,* (2015) who reported for the most common pathogens isolated from wound and sepsis were *S. aureus* and coagulase negative *Staph*. The potential microorganisms isolated were gram positive cooci (Beta haemolytic streptococci, *Erthrococci, Staphylococci*), gram negative aerobic rods (*Enterobacter spp., Escherchia coli, Klebsiella spp.*), anaerobes (*Bacteroides, Clostridium*) fungi (Yeasts, *Aspergillus*). Most of the pathogens are susceptible to vancomycin and ciprofloxacin that is 36.3% and 33.40% respectively while the most resistant drug was ceftriaxone. The culture sensitivity tests showed that numerous and multi drug resistant microorganisms are involved in wounds infection and sepsis. By determining a coefficient approach to the microbiological management of wound complications, meaningful savings in cost and time (i.e. nursing, medical, and microbiological) may be captured while allowing prompt and suitable treatment for the patient.

The present study revealed the findings similar to that of Trojan *et al.,* (2016) determined the prevalence and antibiotic susceptibilities patterns of bacterial isolates from pus samples collected from patients in a tertiary care hospital of Punjab, India. *E. coli* was the most prevalent pathogen (51.2%) followed by *Staphylococcus aureus* (21%), *Klebsiella pneumoniae* (11.6%), *Pseudomonas aeruginosa* (5.8%), *Citrobacter spp.* (3.5%), *Acinetobacter baumannii* (2.3%), *Proteus mirabilis* (2.3%), and Streptococcus spp. (2.3%). *E. coli, K. pneumoniae*, *A. baumannii and Citrobacter isolates was resistant* to multiple antibiotics including higher generation cephalosporins. *S. aureus* and *Streptococcus* isolates were sensitive to cloxacillin and vancomycin. However, *P. aeruginosa, P. mirabilis*, and *Streptococcus* isolates were found to be less resistant to the spectrum of antibiotics tested.

Ruiz *et al.*, (2016) reported *Staphylococcus aureus* is a major cause of nosocomial bacteraemia worldwide, and it has been associated with a high morbidity and mortality rate. Since its discovery in 1880 this microorganism has been able to develop resistance to different antibiotics to which it has been exposed. However, the main events in the evolution of *S. aureus* have been the emergence of resistance to methicillin and the progressive increase of vancomycin MICs, which has been described as 'MIC creep'. The present research findings were in accordance to the findings of Ruiz *et al*., (2016).

On the contrary, Seifoleslami, Safari, & Khameneie, (2015) determined from a total of 350 high vaginal swab specimens collected from fertile and infertile females. Out of the 350 collected samples, eleven were positive for *M. hominis* (3.14%), fifteen were positive for *U. urealyticum* (4.28%) and five were positive for both of them (1.42%). Prevalence of

U. urealyticum and M. hominis in the high vaginal parts of infertile females was higher than fertile females ($P<0.05$). The results of traditional method were also confirmed, using the PCR amplification of urease gene of *U. urealyticum* and 16SrRNA gene of the *M. hominis*. *Ureaplasma urealyticum* and *M. hominis* had a higher prevalence in the high vaginal samples collected during the summer season. Madico *et al.*, (1998) developed a PCR test using vaginal swab samples for the detection of *T. vaginalis* was developed to add *T. vaginalis* infection to the growing list of STDs that can be detected by DNA amplification techniques.

Krech *et al.*, (2009) used a dual collection device containing flocked and wrapped rayon swabs were used to collect vaginal and cervical samples from 494 women. The swabs were separated into individual tubes and sent to the laboratory in a dry state, where they were hydrated and tested for high risk HPV DNA [Digene-Qiagen hybrid capture 2] and *Chlamydia trachomatis* using in-house real-time PCR. The flocked swabs identified more high risk HPV and *C. trachomatis* infections from both sampling sites. However, the present study was carried out on animal subjects and did not reveal any similarity with the findings of Krech *et al.,* (2009) and Seifoleslami, Safari, & Khameneie (2015).

The overall lab investigation results obtained on cultural properties of the bacteria and its antibiotic disc diffusion assay revealed in the present study were supported by the findings of Zhang *et al.,* (2014), Ganguly *et al.*, (2015) and Ganguly (2016, 2017) respectively who found out that *Staphylococcus aureus visine* of the most predominant bacteria involved in the infection leading to pus formation due to infection in various cases and that the infecting etiology showed antibacterial drug(s).

CONCLUSION

The present study revealed the presence of *Staphylococcusspp.* in the pus sample collected from the case of cow abscess. The bacterial strain was found to be sensitive to broad spectrum antibiotics which was reported and recommended to the T.V.C.C. for administration in mixed preparations in divided doses at alternate daily intervals for seven days followed by daily monitoring.

Of late, various effective antibacterial drugs in combined doses are used due to the coexistence of anaerobic and aerobic bacteria in severe or complicated intra-abdominal infections. These pathogenic bacteria may become highly resistant to common antibiotics, triggering refractory or secondary infections. With reference to the findings of the previous studies, local bacteriology and susceptibility results provides clinical guidance for dealing with drug-resistant bacteria worldwide.

The initial empirical antibiotic therapy should be modified based on susceptibility analysis results. In addition, the patients suffering from infection should be immediately administered with the most potent antibiotics. Finally, it is critical to remove the sources of infection and to prevent intraoperative and postoperative bacterial contaminations in order to improve the therapeutic effects of effective antibiotics.

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