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ANTIBIOGRAM OF BACTERIA ISOLATED FROM CHRONIC CUTANEOUS WOUND FLUIDS IN CAPTIVE ASIAN ELEPHANTS *(Elephas maximus)*

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ABSTRACT

Asian elephants possess one of the thickest skins among animals but, injuries to skin are not uncommon. Almost all the wounds get contaminated, if unattended immediately. The chances of progressing of contamination into infection is more and when such infection gets established and developed further the wound becomes chronic. In such cases, wound management strategies need to be revised accordingly. From the clinical point of view, rather than identifying the organism responsible for infection, sensitivity of that organism towards available antibiotics (antibiogram) is the information needed. The study was conducted on captive Asian elephants (*Elephas maximus*) with cutaneous wounds within Kerala and outside over a period of three years. The sensitivity, intermediate sensitivity and resistance to commonly used antibiotics were recorded where resistance was predominant. From this study, it is recommended that an antibiogram should be a part of wound management protocol. Since captive animals share environment with humans, livestock, other animals etc., antimicrobial resistance is an emerging global concern which require further studies taking zoonosis and one health approach into consideration.

Keywords: Chronic wounds, captive elephant, wound fluid, antibiogram, anti-microbial resistance

INTRODUCTION

For all living animals, skin is the primary physical barrier which protects them from external violence (Low *et al.*, 2020) and hence highly prone to injuries. Wounds have existed since the dawn of time. Even though Asian elephants possess

one of the thickest skin among animals (Shoshani, 1982), injuries to skin are not uncommon. In captive animals, it can happen from sharp things (thorns, nails while walking or grazing), improper use of guide devices, harnesses, restraints or conspecific attacks (from own group or others) (Gogoi *et al.*, 2017). Almost all the wounds get contaminated, if unattended immediately. Even with treatment interventions, due to the peculiar habits of elephants like mud bathing (Senthilkumar *et al.,* 2014) and due to unscientific wound management by unprofessional practices, the chances of progressing of contamination into infection is more. When such infection gets established and developed further the wound becomes chronic. In such cases, wound management strategies need to be revised accordingly. From the clinical point of view, rather than identifying the organism responsible for infection, sensitivity of that organism towards available antibiotics (antibiogram) is the information needed. Antibiogram based treatments resulted in faster wound healings in elephants (Barman *et al.,* 2013). Since there was no data regarding the antibiogram of chronic cutaneous wound fluid in captive Asian elephants, a study was undertaken.

MATERIALS AND METHODS

The study was conducted on captive Asian elephants (*Elephas maximus*) with cutaneous wounds reported to Veterinary Hospitals of Kerala Veterinary and Animal Sciences University at Kokkalai and Mannuthy and Elephant camps within Kerala and outside over a period of three years from September 2019 to October 2022. Twelve cases of elephants those with the history of wound persistence beyond eight weeks were selected. For chronic wound fluid sample collection, Levin's technique of quantitative swab culture was performed by spinning the tip of a sterile cotton-tipped applicator stick for five seconds on a one square centimeter area of the wound (Levine *et al.*, 1976) after proper cleansing of the wound area (Gardner and Frantz, 2004). Disc diffusion method was performed on MHA or blood agar where pure culture was used as inoculum. Three to four similar colonies were selected and transferred into three millilitre of Brain heart infusion broth, incubated at 37°C for two to eight hours till light to moderate turbidity was developed and compared with McFarland standard. A sterile non- toxic cotton swab dipped in to the inoculum used to streak the entire agar surface of the plate and allowed to dry with the lid in place. The antibiotic discs were applied under aseptic conditions keeping a distance of at least 24 mm apart. The plates were incubated immediately at 37°C and examined after 14-18 h or later, if necessary. The clear zones were measured and the diameter of

the zones to the nearest millimeter was recorded. The zones were compared with the standard inhibition zone chart to find out sensitive and resistant antibiotics.

RESULTS AND DISCUSSION

During the study period, antibiogram of wound fluid samples from 12 chronic wounds obtained using 29 antibiotic discs. The sensitivity, intermediate sensitivity and resistance to commonly used antibiotics were shown in the figure-1. Mikota (2006) reported that disorders of the integument account for nearly one-third of all medical events reported in North America and in Sri Lanka, it was revealed a high prevalence of wounds (44%). Various levels of occurrence

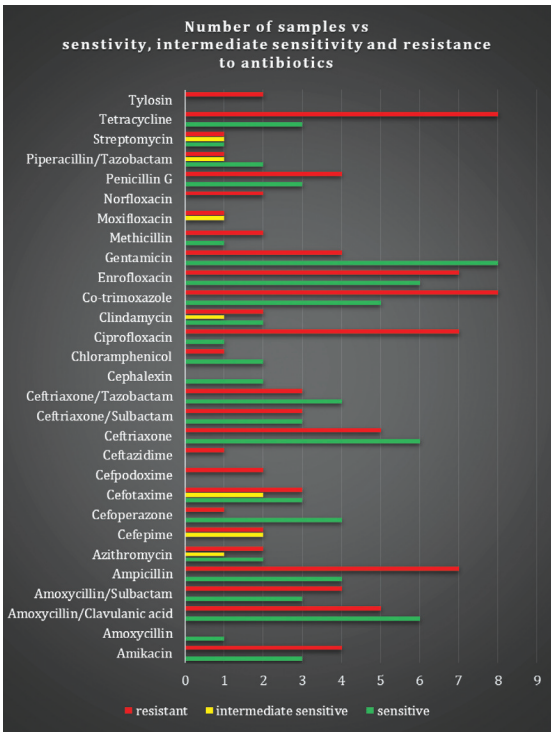

Figure.1

noticed in India also (Ramanathan and Mallapur, 2008; Sajesh, 2009; Narayanan, 2014). When these injuries got supervened by suppurative organisms, it resulted in infections (Gogoi *et al.* 2017) which were highly prevalent in captive Asian elephants (Buragohain *et al.,* 2019) in India. Chronic wounds require a prolonged time to heal (Wysocki, 1996) and infection is one major reason for chronicity (Guo and Dipietro, 2010).

When considering the microbial diversity, chronic wounds are considered as a diverse environment of polymicrobial nature with various levels of bacterial involvement and many of the bacteria live as biofilms which were extremely resistant to irrigation-based cleaning and antibiotic treatment (Atiyeh *et al.,* 2009; Cheong and Kalan, 2019; Schultz *et al.*, 2003). When a wound infection clearly evading or outpacing the body's defenses or when poses a risk to the animal's life, it should be termed as severe or at least serious (Orsini *et al.,* 2017).

Samples shown various levels of sensitivity and resistance but resistance was predominant. Tetracycline and co-trimoxazole shown resistance to the greatest number of samples (8 samples), followed by enrofloxacin, ciprofloxacin and ampicillin (7 samples) and amoxycillin-clavulanic acid and ceftriaxone (5 samples).

Antibiotics like tylosin, norfloxacin, cefpodoxime, cefepime shown resistance only. The antibiotic shown sensitivity most was Gentamicin (8 samples) followed by enrofloxacin, ceftriaxone and amoxycillin-clavulanic acid (6 samples). The challenge of increasingly common issue of antibiotic resistance complicated management of infection-related delay in chronic wounds (Dart *et al.*, 2017). As the antibiogram of this study revealed fairly good amount and wide variance of anti-microbial resistance in chronic cutaneous wounds in elephants, it is recommended that an antibiogram should be a part of wound management protocol. Similar recommendations were done previously also (Barman *et al.,* 2013). Antibiotics usage in cutaneous lesions prophylactically was not advised in the modern era of anti-microbial resistance and recommended only as a necessity when there was a risk of infection (Altoe *et al.,* 2019). It stressed the point that each wound should be considered individually (Nicks *et al.*, 2010).

CONCLUSION

Chronic wound fluids from captive Asian elephants were obtained and subjected to detailed study. Antibiogram of these samples revealed multiple antibiotic resistance which warranted individually customized treatment plans. Since captive animals share environment with humans, livestock, other animals etc. Antimicrobial resistance is an emerging global concern which require further studies taking zoonosis and one health approach also into consideration.

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