

## Bacterial Infection, Antibiogram and Wound Treatment in Domesticated Asian Elephants

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### Introduction

In India domesticated Asian elephants (*Elephas maximus*) are mostly used for heavy work like moving logs, lifting vehicles and demolishing structures. Skin wounds are one of the most commonly encountered pathological conditions in domesticated elephants (Sukklad *et al.* 2006). Mechanical injury from sharp objects like the ankus used by mahouts or pieces of wood, and cut injuries from chains are the most common causes of skin wounds. Foot ailments are also common with a recorded 62% incidence due to split nails, 25% abscesses and 12.5% cracked soles (Singh *et al.* 2010). The large corpus of an elephant is a major predisposing factor to surface injury. The lack of sebaceous glands makes the skin dry and in natural habitats, elephants keep their skin moist and protected from UV rays by covering with mud and dust (Sukklad *et al.* 2006). When the skin is very dry, surface pathogens can easily multiply in skin breaches and aggravate wounds. Wallowing of elephants in mud and dust bathing may facilitate the growth of soil pathogens in damaged skin.

The present study was undertaken to identify organisms associated with skin wounds of elephants and assess their antibiotic sensitivity.

### Materials and methods

A total of 20 exudates and 12 swabs from abscesses and wounds were collected from domesticated elephants engaged in forest logging in Assam, India. Samples were placed in Brain Heart Infusion broth, transported to the laboratory on ice and processed immediately.

Primary isolation was done on 10% oxoid blood agar and MacConkey's lactose agar. After incubation at 37°C for 24-48 hours, the colonies were sub-cultured on to selective media like Mannitol salt agar for *Staphylococcus* and Pseudomonas agar supplemented with Cephalothin, Fucidin and Cetrimide for *Pseudomonas* to obtain pure cultures. Positive cultures were identified on the basis of morphology, staining reactions, cultural characteristics and biological/biochemical tests as per standard methods (Cruickshank *et al.* 1975).

Antibiotic sensitivity of the isolates was performed by the standard disc diffusion method in Mueller Hinton agar plates using a panel of antibiotics. The concentration of antibiotics per disc was: amoxicillin (30 µg), ampicillin (10 µg), chloramphenicol (30 µg), ciprofloxacin (5 µg), enrofloxacin (5 µg), gentamicin (10 µg), cefotaxim (30 µg), norfloxacin (10 µg), neomycin (30 µg), cloxacillin (30 µg), streptomycin (25 µg), amikacin (30 µg), tetracycline (30 µg), oxytetracycline (30 µg) and co-trimoxazole (25 µg). The zone of inhibition was measured, recorded and interpreted according to Clinical and Laboratory Standards Institute criteria (CLIS, 2006).

For management of wounds well trained mahouts were engaged as they were familiar with the respective animals and treatment was done under the guidance of the local veterinary doctor. For wounds located near the vertebral column or legs and/or very deep, the elephant was tranquillized for treatment. A combination of 15 mg medetomidine and 200 mg ketamine was used as a single dose for tranquilization, using

a dart gun. A supplementary dose was given if needed as in draining and dressing of wounds i.e. after 2 hours. Deep seated wounds were opened surgically to facilitate proper drainage of the exudate and removal of decaying tissue. The affected area was drenched daily with 0.1% potassium permanganate solution for 5-7 days and painted with povidone iodine solution. The wound was externally smeared with 'Topicure' antiseptic and fly repellent ointment daily. Based on antibiotic sensitivity test results, all affected elephants were injected with 90 ml of enrofloxacin per 3.5-4.0 ton body weight intramuscularly on 7-10 occasions daily. Tetanus toxoid was injected subcutaneously. Wounds were dressed regularly up to 15-20 days.

### Results and discussion

In the present study a total of 20 domesticated elephants with various categories of septic wounds were assessed. Wounds were mostly located either on the leg (Fig. 1) or on the back of the elephant (Fig. 2) or both. Out of 29 wounds 21 were on the back and 7 were on legs and 1 on the tail. Wounds on back were mostly due to friction with a saddle and on legs due to the use of chain/wire to restrain elephants. They showed different degrees of sepsis ranging from moderate to severe as evidenced by pus formation.

There was a predominance of *Staphylococcus* infections, being isolated from 91% of samples. *Pseudomonas* was isolated from a single tail



**Figure 1.** Deep seated cut wounds on hind leg and foreleg from chains.



**Figure 2.** Coagulated exudate draining from a back wound.

wound sample. All *Staphylococcus* isolates were positive to catalase and grew in high salt concentrations. Twenty seven out of 29 *Staphylococcus* isolates were coagulase positive, while two were negative. Of the *Staphylococcus* isolates 86 % were sensitive to enrofloxacin, followed by gentamicin 69%, and norfloxacin and cefataxim 52%. The *Pseudomonas* isolate also showed a similar trend of antibiotic sensitivity to enrofloxacin, gentamicin, norfloxacin followed by cefataxim. Both *Staphylococcus* and *Pseudomonas* isolates were resistant to cotrimoxazole, ampicillin, cloxacillin, tetracycline, oxytetracycline, neomycin and amikacin. Use of enrofloxacin cleared exudates within 10 days and complete healing of the wounds in 15-20 days.

Coagulase positive *Staphylococcus* was isolated from most wounds assessed. Coagulase positive *Staphylococcus aureus* is considered pathogenic and is associated with a variety of septic conditions. In humans, the skin is colonized by indigenous microbial flora consisting of *Staphylococci*, *Corynebacteria*, *Propionibacteria*, and yeast (Diekema *et al.* 2001). The vast majority of skin and soft tissue infections in humans are caused by *Staphylococcus aureus* (Diekema *et al.* 2001) and  $\beta$  haemolytic *Streptococci* (Di Nubile & Lipsky 2004; Stevens *et al.* 2005). In humans, localized pus-producing lesions such as boils, abscesses, carbuncles and localized wound sepsis are usually due to *Staphylococcus* infection, while rapidly spreading infections such as lymphangitis or cellulitis are usually caused by beta hemolytic *Streptococci* (Dryden 2010). However, in elephants information on

characterization of skin microflora and septic agents is not available.

In the present study all the isolates were resistant to co-trimoxazole, ampicillin, cloxacillin, tetracycline, oxytetracycline, neomycin and amikacin. Resistance to commonly used  $\beta$ -lactam antibiotics (such as penicillin derivatives and cephalosporins) in human and veterinary medicine practices is not uncommon (Akindele *et al.* 2010; Mamza *et al.* 2010). The widespread and inappropriate use of antibiotics is the most common factor responsible for bacterial resistance. Further, transmission of resistant organisms between animal and animal handlers could occur. Methicillin resistant *Staphylococcus aureus* (MRSA) has drawn significant interest in human skin and soft tissue infections. In elephants, MRSA has been reported from a skin infection in an elephant calf at the San Diego Zoo (Janssen *et al.* 2009). Antibiotic resistance of MRSA is not limited to methicillin alone, but other antibiotics such as chloramphenicol, clindamycin, erythromycin and fluoroquinolones as well. *Staphylococcus* resistance to glycopeptides (such as vancomycin, teicoplanin, telavancin, bleomycin, ramoplanin, and decaplanin) remains rare (Awad *et al.* 2007). However, the rising 'Minimum Inhibitory Concentration' of glycopeptides may restrict the efficacy of such agents (Moise-Broder *et al.* 2004).

Management of skin and soft tissue infections normally involves a combination of surgical intervention and empirical antibiotic therapy (Dryden 2010). The main choice of antibiotic depends on the clinical presentation and resistance pattern of organisms involved. In probable gram positive infection, where MRSA is not suspected, penicillins, cephalosporins, clindamycin, and co-trimoxazole are used for treating the wound (Stevens *et al.* 2005). In the present investigation the use of specific antibiotics that the organisms were sensitive to and regular cleaning reduced the oozing of exudate from the first week onwards. Necrotic tissue was replaced by healthy fibroblastic cells and the wound gap minimized by the second week. An antibiogram is therefore an essential tool in the management of skin wounds in elephants.

In conclusion, this study demonstrates that coagulase positive *Staphylococcus aureus* is the most commonly associated pathogen of skin abscesses and wound in domestic working elephants. Enrofloxacin was found to be the most sensitive antibiotic and used for effective wound treatment. Antibiotic sensitivity study and judicious use of suitable drugs is likely to prevent development of resistant organisms. Therefore it should be adopted as a regular part of skin-wound management protocol in elephants.

## References

- Akindele AA, Adewuyi IK, Adefioye OA, Adedokun SA & Olaolu AO (2010) Antibiogram and beta-lactamase production of *Staphylococcus aureus* isolates from different human clinical specimens in a tertiary health institution in Ile-ife, Nigeria. *American-Eurasian Journal of Scientific Research* **5**: 230-233.
- Awad SS, Elhabash SI, Lee L, Farrow B & Berger DH (2007) Increasing incidence of methicillin-resistant *Staphylococcus aureus* skin and soft-tissue infections: reconsideration of empiric antimicrobial therapy. *American Journal of Surgery* **194**: 606-610.
- Cruickshank R, Duguid JP, Marmion BP & Swain RH (1975) *Medical Microbiology: The Practice of Medical Microbiology. Vol. 2.* Churchill Livingstone, Edinburgh.
- Di Nubile MJ & Lipsky BA (2004) Complicated infections of the skin and skin structures: when the infection is more than skin deep. *Journal of Antimicrobial Chemotherapy* **53 (S2)**: 37-50.
- Diekema DJ, Pfaller MA, Schmitz FJ, Smayevsky J, Bell J, Jones RN, & Beach M & the SENTRY Participants Group (2001) Survey of infections due to *Staphylococcus* species: frequency of occurrence and antimicrobial susceptibility of isolates collected in the United States, Canada, Latin America, Europe, and the Western Pacific region for the SENTRY Antimicrobial Surveillance Program, 1997-1999. *Clinical Infectious Diseases* **32 (S2)**: 114-132.

Dryden MS (2010) Complicated skin and soft tissue infection. *Journal of Antimicrobial Chemotherapy* **65** (S3): 35-44.

Janssen D, Lamberski N, Dunne G, Ginsberg M, Roach C, Tweeten S, Gorwitz R, Waterman S, Bensyl D & Sugerman D (2009) Methicillin-resistant *Staphylococcus aureus* skin infections from an elephant calf - San Diego, California, 2008. *Morbidity and Mortality Weekly Report* **58**: 194-198.

Mamza SA, Egwu GO & Mshelia GD (2010) Beta-lactamase *Escherichia coli* and *Staphylococcus aureus* isolated from chickens from Nigeria. *Veterinaria Italiana* **46**: 155-165.

Moise-Broder PA, Sakoulas G, Eliopoulos GM, Schentag JJ, Forrest A & Moellering Jr. RC (2004) Accessory gene regulator group II polymorphism in methicillin-resistant *Staphylococcus aureus*

is predictive of failure of vancomycin therapy. *Clinical Infectious Diseases* **38**: 1700-1705.

Singh JL, Das AK, Patel M & Thathoo AK (2010) Management of foot affections in Asian elephants. *Indian Veterinary Journal* **87**: 201.

Stevens DL, Bisno AL, Chambers HF, Everett ED, Dellinger P, Goldstein EJC, Gorbach SL, Hirschmann JV, Kaplan EL, Montoya JG & Wade JC (2005) Practice guidelines for the diagnosis and management of skin and soft-tissue infections. *Clinical Infectious Diseases* **41**: 1373-1406.

Sukklad S, Sommanustweechai A & Pattanarangsarn R (2006) A retrospective study of elephant wound, wound management from Thai Veterinarians. In: *Proceedings of AZWMP*. Chulalongkorn University, Faculty of Veterinary Science, Bangkok, Thailand. pp 16.



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