

Preliminary investigation of microbial diversity and antibiotic susceptibility profiling of *Staphylococcus* spp. isolated from tick salivary glands infesting domestic animals in Kandy

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Introduction and Objectives: Ticks pose a hazard to both humans and animals by feeding on blood and transmitting diseases, facilitated by their saliva which plays a crucial role in pathogen transmission. Heavy tick infestations in animals often lead to skin infections necessitating antimicrobial treatment. *Staphylococcus aureus* is known for causing skin and soft tissue infections. This study aimed to identify aerobic bacteria in tick saliva and assess the antimicrobial susceptibility profiles of *Staphylococcus* species.

Methods: Ten ticks (6 from dogs, 3 from cattle, and 1 from a horse) were aseptically dissected to collect salivary glands. Each salivary gland was enriched in buffered peptone water at 37 °C for 2 hours and cultured on blood agar at 37 °C for 18-24 hours under sterile conditions. Morphologically distinct bacterial colonies were subjected to biochemical tests for species identification. Gram-positive isolates were tested for catalase, slide and tube coagulase and cultured on MSA agar. Identified *Staphylococcus* spp. underwent disc diffusion assays with the following antibiotics: erythromycin (15 µg), cefoxitin (30 µg), ampicillin (10 µg), ciprofloxacin (5 µg), trimethoprim-sulfamethoxazole (25 µg), gentamicin (10 µg), and clindamycin (2 µg), following CLSI guidelines. Parallel to our experiments, ATCC *Staphylococcus aureus* 25923 was tested and zone diameters obtained were evaluated using CLSI values for QC strains, which was used as a positive control.

Results: The collected ticks represented *Boophilus* (3), *Ixodes* (2), *Rhipicephalus* (2), *Ornithodoros* (1), *Haemaphysalis* (1), and *Aponomma* (1) species. Eighteen bacterial colonies were isolated from tick saliva, consisting of 8 Gram-negative rods and 10 Gram-positive cocci. Biochemical assays identified the Gram-negative isolates as *Citrobacter* species and 5 of the Gram-positive cocci isolates as coagulase-positive *Staphylococcus* (*Staphylococcus aureus* and *Staphylococcus pseudintermedius*). All coagulase-positive *Staphylococcus* isolates were susceptible to all tested antimicrobials except clindamycin, which showed intermediate susceptibility (80%) or resistance (20%).

Conclusions: Besides medically and veterinary significant protozoans and viruses, ticks also transmit a variety of bacteria. All tested ticks carry aerobic bacteria in saliva, with *Staphylococcus* species being the most prevalent and showing susceptibility to the tested antimicrobials. A more comprehensive investigation involving a broader range of tick species from diverse hosts and geographical areas is essential for accurately identifying the risk posed to hosts.

Keywords: Ticks, saliva, bacteria, antimicrobial, susceptibility

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