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A survey of veterinary hospitals in Nigeria for the presence of some bacterial organisms of nosocomial and zoonotic potential

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Summary

This study was undertaken to determine the type and estimate the prevalence of bacterial organisms on contact surfaces of five close-to-patient facilities in three veterinary health care settings within the Sokoto metropolis of north-western Nigeria. A total of 30 samples (10 from each setting) were collected and analysed using culture, microscopy and biochemical testing. Bacterial species isolated from samples in this study included the following: *Bacillus* sp. (27.3%), *Staphylococcus aureus* (15.9%), *Listeria* sp. (13.6%), *Streptococcus* sp. (11.4%), *Salmonella* sp. (6.8%), *Escherichia coli* (4.5%), *Staphylococcus epidermidis* (4.5%), *Citrobacter* sp. (2.3%), *Klebsiella* sp. (2.3%), *Lactobacillus* sp. (2.3%), *Micrococcus* sp. (2.3%), *Pasteurella* sp. (2.3%), *Proteus* sp. (2.3%), and *Yersinia* sp. (2.3%). A higher percentage (64.3%) of the total bacterial isolates were zoonotic in nature and hence of public health significance. Some pathogens have the potential of nosocomial spread. In this study, we seek to establish the first evidence of bacterial presence in the major veterinary health care settings in the Sokoto region of north-western Nigeria. Of particular interest is the hypothesis, which has not previously been formally tested, that nosocomial infections are especially likely to be implicated in both animals and occupational diseases in Nigeria. It was suggested that some of these isolates were associated with the risk of nosocomial and zoonotic infections and hence draws attention to the need to rigorously employ standard veterinary precautions as part of the hospital's infection control programme in an attempt to protect both patients and staff from infections.

Keywords

Bacterium, Hospital, Nigeria, Nosocomial, Veterinary, Zoonosis.

Un'indagine negli ospedali veterinari in Nigeria sulla presenza di alcuni organismi batterici con potenziale nosocomiale e zoonotico

Riassunto

Scopo di questo studio era determinare il tipo, stimandone la prevalenza, di organismi batterici sulle superfici di contatto di cinque sale per pazienti in tre cliniche veterinarie della metropoli di Sokoto, nella Nigeria nord-occidentale. Sono stati raccolti in totale 30 campioni (10 per ciascuna clinica) e sottoposti ad analisi delle colture, esami

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microscopici e biochimici. Le specie batteriche isolate dai campioni sono le seguenti: Bacillus *sp. (27,3%),* Staphylococcus aureus *(15,9%),* Listeria *sp. (13,6%),* Streptococcus *sp. (11,4%),* Salmonella *sp. (6,8%),* Escherichia coli *(4,5%),* Staphylococcus epidermidis *(4,5%),* Citrobacter *sp. (2,3%),* Klebsiella *sp. (2,3%),* Lactobacillus *sp. (2,3%),* Micrococcus *sp. (2,3%),* Pasteurella *sp. (2,3%),* Proteus *sp. (2,3%) e* Yersinia *sp. (2,3%). Una percentuale più elevata (64,3%) degli isolati batterici totali era di natura zoonotica e pertanto significativa per la salute pubblica. Alcuni patogeni presentano potenziale di diffusione nosocomiale. In questo studio gli autori hanno cercato di stabilire le prime evidenze di presenza batterica nelle principali cliniche veterinarie della regione di Sokoto, nella Nigeria nord-occidentale. Particolarmente interessante è l'ipotesi, non formalmente verificata in precedenza, della probabilità che in Nigeria le infezioni nosocomiali siano coinvolte nelle malattie sia animali che occupazionali. È stato proposto che alcuni di questi isolati siono associati al rischio di infezioni nosocomiali e zoonotiche, il che sottolinea la necessità di applicare rigorosamente norme precauzionali veterinarie nell'ambito del programma ospedaliero di controllo delle infezioni allo scopo di proteggere sia i pazienti che il personale.*

Parole chiave

Batteri, Nigeria, Nosocomiale, Occupazionale, Ospedale, Veterinario, Zoonosi.

Introduction

Field and laboratory veterinary work has been associated with a wide variety of hazards for many years (3). Nigerian veterinarians are increasingly exposed to zoonotic diseases through occupational exposure and contact with animals, and only a few veterinary health care settings employ the recommended precautions as standard tools in infection control.

Health is a high priority for any society and infections remain a leading cause of disease globally. Infections that occur among hospitalised patients and become manifest only after 48 h of stay are called 'nosocomial' (17). Such nosocomial or hospital-acquired infections lead to significant additional morbidity, mortality and economic burdens than should be expected from the patient's underlying disease alone.

Veterinary practices are unique environments that bring humans into close contact with many different species of patients. In the practice environment, whether in a building or in the field, veterinary personnel are frequently exposed to recognised and unrecognised infectious pathogens, many of which are zoonotic (11). Some of the documented zoonotic infections that have occurred in veterinary personnel include: methicillin-resistant *Salmonella* Typhimurium, cryptosporidiosis, cat-associated plague, cat-associated sporotrichosis, methicillin-resistant *Staphylococcus aureus* and dermatoplytosis. Such zoonotic diseases that occur in veterinary personnel as a result of being in contact with infected animals are referred to as infectious occupational hazards. Occupational hazards are mainly caused by agents of zoonotic infections such as *Brucella* sp., *Mycobacterium* sp., *Salmonella* sp., *Bacillus* sp., *Neisseria* sp., *Yersinia* sp., *Listeria* sp., etc. (10, 11).

During the career of a veterinarian, the majority suffer an animal-related injury resulting in hospitalisation and/or significant loss of work time (4, 6, 7, 8, 12). Most clinically infected wounds from dog and cat bites are mixed infections of aerobic and anaerobic bacteria (16).

The aim and objective of this survey was to establish preliminary evidence of the presence of common aerobic or aero-anaerobic bacteria on five close-to-patient hospital facilities and to determine their prevalence, placing emphasis on those with possible nosocomial and zoonotic potential, to enable us to develop a tailor-made infection control programme for each setting and to recommend possible considerations on the need for further study. Other sources of contamination that are equally or more dangerous, such as anaerobic bacteria, viruses and parasites have not been demonstrated in this study.

Materials and methods

Study area

This study covered three major veterinary hospitals in the city of Sokoto, the capital of Sokoto State, located in the north-western zone of Nigeria. The state is located between longitudes 4°05′E and 6°40′E and latitudes 11°30′N and 14°00′N and shares common borders with Niger to the north, Kebbi State to the south-west and Zamfara State to the east.

Materials used

All the glassware used were first washed and sterilised in a hot air oven at a temperature of 160°C for 2 h and later stored in aseptic conditions until required for use.

Sample collection

Samples were collected from three metropolitan veterinary premises, namely:

- the State Veterinary Clinic located at the Sokoto North Local Government Area
- the Large Animal Clinic of the Usmanu Danfodiyo University Veterinary Teaching Hospital (UDUVTH)
- the Small Animal Clinic of the UDUVTH.

All three hospitals are located in the state capital and receive cases from different areas through visits from patients or through ambulatory services. The State Veterinary Clinic is owned by government and treats ovine, caprine and avian cases. Most of the canine and complicated cases in all species are referred to the UDUVTH, due to the fact that it is an institutionally based hospital with improved facilities and better expertise. The UDUVTH was divided into large animal, small animal and avian clinics with each receiving species-specific cases that are handled by specialists in each respective area. All three hospitals perform bacterial disease diagnosis on live animals admitted to the hospital. Diagnoses of dead animals are performed in post-mortem rooms available on the premises of each hospital. The Veterinary Microbiology and Public Health Laboratories are owned by the institution and receive clinical veterinary samples from across the state.

The prepared sterile swabs were used to swab the various contact surfaces that are considered to be the most proximal to patients. Those surfaces include surgical tables, surgical packs, examination tables, equipment trays and surgical drapes/towels. The original sample was collected in 5 ml of transport medium after swabbing an area of 100 cm^2 of each surface. The samples were collected in pairs, i.e. one in peptone water and the other in selenite-F broth media and transported to the laboratory. All surfaces and equipment from which samples were taken were assumed to be cleaned and disinfected.

Inoculation and isolation

A total of 1 ml per sample collected was inoculated onto the respective media after a serial (10-fold) dilution, using a dilution factor of 10^{-5}. All samples in peptone water were inoculated in two Petri-dishes, with blood and MacConkey agar, respectively. Samples in selenite-F broth were inoculated in Petri-dishes containing deoxycholate citrate agar (DCA). All inoculated Petri-dishes were incubated at 37°C for 24 h, after which the colonies were observed and broth cultures were then sub-cultured in the corresponding media plates, incubated at 37°C for 24 h, and observed for characteristic colonies.

Each isolated colony was preserved in a nutrient agar slant followed by smearing of each isolate on microscopic glass slide for staining.

A portion of each isolated colony was smeared on a glass slide and stained with Gram staining reagents and observed under a light microscope using an oil immersion objective (×100). Both the morphology and the Gram reaction of the organisms were observed and noted.

Biochemical tests

Biochemical tests were performed to confirm the genus and/or species of the bacteria associated with each sample. The catalase, coagulase, triple sugar ion and indole tests were performed in accordance with the methods described in the *Cowan and Steel Manual* (2).

Results

In general, bacteria of different genera/species were isolated with varying frequencies among all the sites sampled/examined. The occurrence of common bacterial isolates from different premises and sampling sites are presented in Tables I and II, respectively.

Duplicate plates were used. Supposing the dilution on a plate was 10^{-5}, the average counts were obtained using (12+10)/2 and the product multiplied by the dilution factor. The estimated number of organisms in 5 ml was found to be 5.5×10^{-6} colony-forming units (cfu) and the total count per cm^2 was found to be 5.5×10^{-4} cfu/cm^2. Bacterial counts exceeding 150 cfu/cm^2 are an indication of inadequate sanitation and call for improved sanitation. The study was conducted to establish evidence of presence of those bacterial isolates in premises but the possible origin of pathogens recorded in staff and in animals was not determined.

Discussion

The results of this study are shown in Tables I and II. The study revealed that facilities in veterinary clinics in Sokoto are highly contaminated with both pathogenic and non-pathogenic bacteria. Although the aim of this survey was not to protect veterinarians as they have to touch animals that are more contaminated than the equipment, but rather the staff who were not involved in caring for the animals (those involved in maintenance and cleaning activities), possibly the owners of the animals and the healthy animals that were examined on contaminated surfaces.

In this study, *Bacillus* sp. had the highest prevalence (27.3%), with equipment trays being the major source of infection. This may be due to the ubiquitous nature of the bacillus spores that are known to resist heat (high temperatures), the organism germinates and multiplies rapidly at room temperature (1). Other reasons that may be responsible for the high prevalence of *Bacillus* sp. may be the ability of the organism to survive on dry surfaces and the hygienic conditions under which the veterinary clinics are being managed.

S. aureus was the second most prevalent organism in this study and constituted 15.9% of the total isolates. Examination tables, equipment trays and surgical drapes/towels were the major source of infection by this

Table I
Prevalence of bacterial isolates from different premises

Bacterial isolates	State veterinary clinic	Large animal clinic, UDUVTH	Small animal clinic, UDUVTH	Total
Bacillus sp.	5	3	4	12 (27.3%)
Staphylococcus aureus	3	4	0	7 (15.9%)
Listeria sp.	1	2	3	6 (13.6%)
Streptococcus sp.	2	3	0	5 (11.4%)
Salmonella sp.	3	0	0	3 (6.8%)
Escherichia coli	0	2	0	2 (4.5%)
Staphylococcus epidermidis	1	0	1	2 (4.5%)
Citrobacter sp.	1	0	0	1 (2.3%)
Klebsiella sp.	0	1	0	1 (2.3%)
Lactobacillus sp.	0	1	0	1 (2.3%)
Micrococcus sp.	1	0	0	1 (2.3%)
Pasteurella sp.	0	0	1	1 (2.3%)
Proteus sp.	0	1	0	1 (2.3%)
Yersinia sp.	1	0	0	1 (2.3%)
Total	18 (40.9%)	17 (38.6%)	9 (20.5%)	44 (100%)

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Table II
Prevalence of bacterial isolates from sampling sites

Bacterial isolates	Surgical tables	Surgical packs	Examination tables	Equipment trays	Surgical drapes/towels	Total
Bacillus sp.	1	3	2	5	1	12 (27.3%)
Staphylococcus aureus	1	0	2	2	2	7 (15.9%)
Listeria sp.	2	2	1	1	0	6 (13.6%)
Streptococcus sp.	0	2	1	2	0	5 (11.4%)
Salmonella sp.	1	1	1	0	0	3 (6.8%)
Escherichia coli	1	0	0	1	0	2 (4.5%)
Staphylococcus epidermidis	0	1	0	0	1	2 (4.5%)
Citrobacter sp.	0	0	1	0	0	1 (2.3%)
Klebsiella sp.	0	0	0	0	1	1 (2.3%)
Lactobacillus sp.	0	1	0	0	0	1 (2.3%)
Micrococcus sp.	1	0	0	0	0	1 (2.3%)
Pasteurella sp.	1	0	0	0	0	1 (2.3%)
Proteus sp.	0	1	0	0	0	1 (2.3%)
Yersinia sp.	0	1	0	0	0	1 (2.3%)
Total	8 (18.2%)	12 (27.3%)	8 (18.2%)	11 (25.0%)	5 (11.4%)	44 (100%)

organism. This prevalence observed might be due to the fact that the organism is comparatively stable in the environment and in certain conditions, such as improper cleaning and disinfection of veterinary facilities. The organism occurs as a commensal on the skin and mucous membrane of animals and is the causative agent of staphylococcal gastroenteritis in animals and humans through the production of enterotoxins. Worldwide, *S. aureus* is the leading cause of gastroenteritis resulting from the consumption of contaminated food (9). The organisms can be spread on the hands and clothes of veterinary personnel during clinical procedures that involve animal handling for medical examination or surgery, or cleaning of hospital premises by workers.

Listeria sp. was responsible for approximately 13.6% of the bacterial isolates and was most often isolated from surgical tables and packs. The organisms can be recovered from animal faeces and can grow and replicate in the environment at a wide range of temperatures (from 4°C to 45°C). This contributes to the strong presence of the organism in our hospital premises. The organism is the causative agent of listeriosis, a disease characterised by abortion, septicaemia and endopthalmitis (15).

Streptococcus sp. constituted 11.4% of isolates, with a higher prevalence on surgical packs and equipment trays. Streptococci are often commensals of mucous membranes, are known to cause pyogenic infection in many species of animals and in humans and are associated with abscess formation, suppurative conditions and septicaemia (15).

Salmonella sp. constituted 6.8% of isolates and was frequently identified on surgical tables, surgical packs and examination tables. *Salmonella* sp. belongs to the family *Enterobacteriacae* which inhabits the gastro-intestinal tract of humans and animals. It is of public health importance in that 1 cfu/g of its isolates has been incriminated in food poisoning (1). Improper cleaning and disinfection of contact surfaces in veterinary clinics contribute to the proliferation of the organism, even at high levels, because it can grow and multiply at a wide range of environmental temperatures. Infection is characterised by an incubation period that varies between 6 h to 7 days (1).

Escherichia coli constituted 4.5% of the bacterial isolates and its highest prevalence was observed on surgical tables and equipment trays. *E. coli* also belongs to the family *Enterobacteriacae*. It is a normal component of the flora in the large intestine of warm-blooded animals including humans. This organism occurs in appreciable numbers of veterinary clinics of which its source is the faecal material of animals being presented for one treatment or another, coupled with the unhygienic condition of the environment.

Other bacterial isolates incriminated in this study include: *Staphylococcus epidermidis* (4.5%), *Citrobacter* sp. (2.3%), *Klebsiella* sp. (2.3%), *Lactobacillus* sp. (2.3%), *Micrococcus* sp. (2.3%), *Pasteurella* sp. (2.3%), *Proteus* sp. (2.3%) and *Yersinia* sp. (2.3%). These are also very important organisms that survive in the practice environment as a result of improper hygienic practices.

This study also revealed that the degree of contamination varies between the premises sampled (Table I). The highest prevalence of bacterial isolates (40.9%) was observed in the State Veterinary Clinic; this is followed by the Large Animal Clinic of the UDUVTH which had a prevalence of 38.6%. The Small Animal Clinic of the UDUVTH was observed to have the lowest prevalence (20.5%), which may be due to the fact that in the Small Animal Clinic, more sterilisation/disinfection measures are employed in comparison with the Large Animal Clinic and the State Veterinary Clinic.

The frequency of infectious agents in the facilities was also recorded (Table II). It was observed that surgical packs and equipment trays, with a prevalence of 27.3% and 25%, respectively, are the two major sources of infection to both patients and personnel. This is then followed by surgical tables and examination tables, each of which had a prevalence rate of 18.2%. Surgical drapes/towels revealed the lowest prevalence (11.4%).

To minimise and control the risk of contracting and disseminating zoonotic diseases to practitioners and nosocomial infections to visiting animal patients, we recommended the following:

- all clinicians should strictly adhere to the standard veterinary precautions at all times
- each setting should formulate an infection control plan tailored for its practice
- adequate hospital disinfectants, necessary safety equipment and a clean water supply should be standardised in all hospitals
- staff education and training, especially on the appropriate use of hospital disinfectants and cleaning procedures, should be implemented
- public health veterinarians should be encouraged to implement client education programmes that will contribute to the protection of clients and their children from contracting zoonoses
- since both patients and veterinarians are at increased risk of contracting nosocomial and zoonotic infections, respectively, specific guidelines, both for animals and veterinarians, infection control and general guidelines with protocols for reporting accidents should be instituted in every health care setting
- further studies and the use of improved methodologies for bacterial speciation, antibiograms and molecular characterisation of isolates should be conducted to lead to a better understanding of their biology which may help in developing more effective infection control measure(s).

Conclusion

In conclusion, samples collected and analysed in this study showed that the veterinary hospitals in Sokoto are heavily contaminated with bacteria, among which most are zoonotic and hence are of serious public health significance and can be considered nosocomial hazards.

Further studies on the acceptable contamination thresholds per ingestion for each of the different pathogens isolated, together with their antibiogram to determine possible resistance, are recommended. The latter is required due to the fact that the presence of resistant pathogens in hospital premises has

implications for animal husbandry and the transfer of antibiotic-resistant bacteria to humans (13) and other visiting patients. The evidence of the presence and prevalence of other non-bacterial pathogens should be studied, especially in practices involved in the handling of pets as they offer significant benefits to our society and ownership of pets is associated with health hazards due to close companionship (5, 14).

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