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ISOLATION AND IDENTIFICATION OF AEROBIC BACTERIA FROM SHEEP WITH RHINITIS

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Abstract: Sheep are important in the livestock economy by their adaptability to adverse environmental conditions as they are good sources of protein and income for the rural poor. Studies conducted on the bacterial flora of the respiratory tract in sheep focused on the pneumonic lungs with rhinitis. The nasal swab from each sheep was analyzed using standard methods. Thirty eight samples were obtained and all samples contain bacterial isolates. The most frequently isolated species was *Staphylococcus aureus* (42.1%), while *Staphylococcus saprophyticus* (18.4%) and *Escherichia coli* (13.1%) were the second dominant bacteria. Other species were isolated at relatively lower rates. The isolation of *Staphylococcus sp.,* and *E.coli* from the nasal cavity of sheep in this study reflects their possible role in most common respiratory diseases encountered in small ruminants. This study shows the relationship between misuse or unrestricted use of antibiotics and drug resistance. Therefore, there is a need for practitioners and researchers to be informed of the appropriate antibiotics to be used in respiratory infections and during control programs.

Keywords: Sheep, Rhinitis, Microbial flora.

Introduction

Respiratory diseases of various etiologies have been described in different domestic animals. However, the problem is more common in sheep due to the fact that the ratio of the alveolar surface to metabolic weight is very low in sheep compared to other species [1].

Although a single agent may be the primary invader, most respiratory infections are complicated by the presence of secondary or opportunistic invaders. When the local resistance of respiratory mucus is lowered, bacteria growing in the nose and throat extend down wards, usually producing multiple bacterial infections [2].

Many specific primary pathogens have been implicated sheep pneumonia. The most common bacterial causes of pneumonia in sheep include *pasteurella spp., Mannheimia haemolytica*, *Actinomyces pyogenes* and several *mycoplasma species*. *P. multocida* and *m.haemolytica* are important contributory pathogens in enzootic or primary pneumonia in sheep, although their pathogenic effects are enhanced when sheep are infected with viruses [3].

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Respiratory disease can lead to severe financial losses and welfare implications in sheep flocks. Individual animals may be affected or outbreaks can occur, with losses due to mortality, reduced production – poor or delayed growth in fattening lambs with a greater feed consumption for finishing, and ill economy and poor milk production in adult ewes – and treatment costs. Because of the clinical economic importance of the disease in sheep, it was a topic of interest of many researchers in the field of small ruminant practice.

Besides, most of the infectious agents that cause respiratory disease are abundant in nature and are normal inhabitants of the nasopharynx of normal animals. This often creates difficulty with the interpretation of findings in outbreaks of respiratory diseases [4]. Therefore, the objectives of this study were to determine the type of normal bacterial inhabitants in nasopharyngeal passageways of apparently healthy and clinically sick sheep.

The extent of normal bacterial inhabitants of nasal cavity that serves as a standard in the diagnosis and treatment of respiratory disease out breaks and identifies potential bacterial species that may be considered for future research work in etiological identification [5].

Materials and Methods

Sample Collection

The nasal samples were collected by inserting sterile cotton-tipped applicator sticks or swab into the nasal passage after proper cleaning and disinfection of the external wares using 70% alcohol. Each nasal swab was carefully cut and put into a labeled bottle containing 2 mL brain heart infusion broth. The swabs were transported in a cool box to the laboratory for bacterial culture. In the laboratory, samples were immediately incubated aerobically at 37°C for 24 hours [6].

Bacteriological examination

Each nasal swab was removed from the bottle and streaked over the plates containing blood agar base supplemented with 7% sheep blood and McConkey agar. The streaking was further spread with inoculating loop to aid colony isolation. The plates were labeled and incubated aerobically at 37°C for 24-48 h [7]. After taking note of cultural growth characteristics, positive cultures were subjected to Gram's staining properties and cellular morphology observed with a light microscope (x100). Mixed colonies and Gram negative bacteria were subcultured on both blood and McConkey agars and further incubated aerobically for 24 h. Pure culture of single colony type from both blood and McConkey agars were transferred onto nutrient agar slants for a series of biochemical tests including catalase, oxidase and fermentative/oxidative tests for final identification following standard procedures [8].

Results

From a total of 38 swab samples collected from the nasal passage of clinically sick sheep, all of them contained bacteria. The gram staining revealed 23 (60 %) and 15 (40 %) were G +ve and G-ve bacteria respectively.

Table 1: Bacterial species isolated from clinically infected sheep (n = 38)

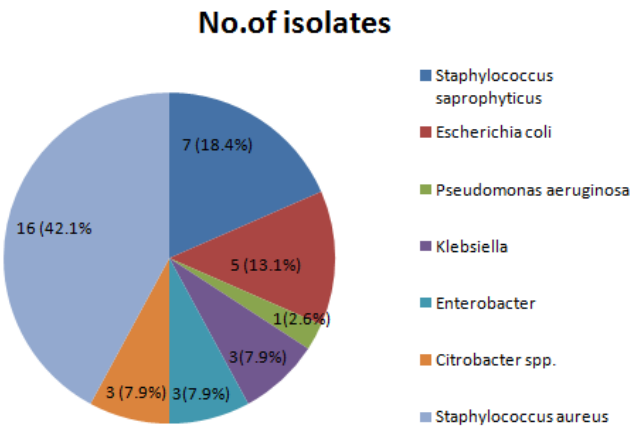
Sl. no.	Bacterial isolate	No. of isolates	Percentage (%)
1	*Staphylococcus saprophyticus*	7	18.4%
2	*Escherichia coli*	5	13.1%
3	*Pseudomonas aeruginosa*	1	2.6%
4	*Klebsiella*	3	7.9%
5	*Enterobacter*	3	7.9%
6	*Citrobacter spp.*	3	7.9%
7	*Staphylococcus aureus*	16	42.1%

The most frequently isolated species from animals were:

1. *Staphylococcus saprophyticus* (18.4%)
2. *Escherichia coli* (13.1%)
3. *Pseudomonas aeruginosa* (2.6%)
4. *Klebsiella, Enterobacter, and Citrobacter spp.* (23.7%)
5. *Staphylococcus aureus* (42.1%)

Pseudomonas aeruginosa were the least encountered bacterial species among the isolates. On the other hand, the predominant species among the isolates recovered from the nasal cavity of clinically sick sheep were *Staphylococcus aureus* (42.1%).

The majority of isolates colonize the nasal cavity of the examined animals with the exception of *Staphylococcus aureus* 42.1% and *Klebsiella, Enterobacter, and Citrobacter spp.23.7%.* Gram positive bacteria were dominant over Gram negative (60% Vs 40%) in clinically sick sheep in this study.



Bacterial species isolated from clinically infected sheep (n = 38)

Discussion

The study has showed a wide variety of bacterial species colonized the nasal passage of clinically sick sheep. Several workers isolated similar bacteria from pneumonic caprine lungs [9, 10]; apparently healthy respiratory tract and nasal cavity of goats **[11]** and with fewer reports from apparently healthy sheep [12].

The invariable isolation of these organisms from the nasal cavity of clinically sick sheep in this study reflects their possible role in respiratory syndrome. The normal bacteria of healthy individual animal can be altered by several factors such as the nutritional and immunological status of the animal or the environment.

The suppression of the normal bacteria frequently allows the development of potential pathogens, leading to the presentation of a variety of pathologies [2]. The pathogenic bacteria isolate, *Staphylococcus aureus*, *Klebsiella, Enterobacter and Citrobacter spp* were isolated in higher proportion from the nasal passage of clinically sick sheep.

Other previous papers [11] reported high incidence rate (55.46%) from pneumonic lungs. *Mannhaemia haemolytica* which is normal flora of upper respiratory tract may play a secondary role after the primary initiating agent suppress the host defense mechanism and favors the multiplication of Pasteurella species leading to bronchopneumonia.

In the current study, *Pasteurella multocida* was not recovered from the nasal passage of clinically sick sheep. *Pasteurella multocida* frequently inhabited tonsil and nasal cavity. This is inconsistent with previous reports from nasal cavity of clinically sick sheep [13].

This finding conforms well to the previous study similarly isolated relative proportion rate from the nasal cavity of clinically sick sheep [10].

Therefore, there is a need for practitioners and researchers to be aware of the aerobic nasal bacterial flora of the sheep and of their antibiotic sensitivities so as to be informed of the appropriate antibiotics to be used in the course of respiratory infections and control programs.

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