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Antibiogram of bacterial isolates recovered from goats with nasal discharge

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Abstract
Respiratory tract ailments, characterized by nasal discharge, sneezing and coughing, are prevalent among goats in rural India where veterinary healthcare facilities are limited. Present study focuses on the critical role of small ruminant rearing in the rural parts of India where goats are raised for milk and meat purpose. In these rural areas, goat owners keep animals in close proximity with each other which leaves a possibility of rapid spread of infectious diseases. Lack of timely veterinary care by experts encourages owners to get their animals treated instantly by use of different antibiotics haphazardly which can lead to antibiotic resistance in animals. Hence, the present study was conducted to study antibiogram of bacterial isolates recovered from privately-owned goats having nasal discharge as a symptom irrespective of age, breed, gender and disease. The most consistent bacteria isolated from nasal swabs in goats with nasal discharge was *Staphylococcus* spp. (93.33%) followed by *Streptococcus* spp. (83.33%), *Escherichia coli* (73.33%), *Klebsiella* spp. (53.33%) and *Salmonella* spp. (23.33%). The mixed presence of two different isolates was observed highest for *Staphylococcus* spp. + *Escherichia coli* (56.67%) followed by *Staphylococcus* spp. + *Klebsiella* spp. (43.33%), *Streptococcus* spp. + *Escherichia coli* (16.67%), *Staphylococcus* spp. + *Salmonella* spp. (23.33%) and *Streptococcus* spp. + *Klebsiella* spp. (10.00%). The results of antibiotic sensitivity test (ABST) revealed a considerable variation in the sensitivity of these organisms to antimicrobial agents. Notably, some antibiotics, including Gentamicin, Ciprofloxacin, Streptomycin, Clotrimoxazole, and Chloramphenicol, showed variable efficacy against the isolates. In contrast, a smaller proportion of the isolates exhibited sensitivity to Tetracycline, Cefixime, Erythromycin, and the Ampicillin/Sulbactam combination. This suggests that these antibiotics had limited effectiveness against the majority of the isolates. Alarmingly, all of the isolates displayed resistance against Penicillin G, Ampicillin, and Cefotaxime, rendering these antibiotics ineffective for treating infections associated with nasal discharge in this study. These findings underscore the urgent need for responsible antibiotic use and the exploration of alternative treatment options for respiratory tract infections in small ruminants to combat the growing concern of antibiotic resistance.

Keywords: Antibiogram, nasal bacteria, goat, respiratory diseases

Introduction
The present era has witnessed significant advancements in the diagnostics and treatment of various diseases in livestock animals like cattle, buffalo, sheep, and goats. India, with its substantial population of these animals and significant production, particularly in terms of milk and meat, holds a prominent position in this regard. With this prominence comes a responsibility for stakeholders, researchers, and academics to assess the current gaps and deficiencies in the healthcare and management of these animals. This evaluation is essential to ensure the well-being and productivity of livestock and to meet the growing demands of the industry.
India boasts a significant presence in the realm of livestock production, including cattle, buffalo, sheep, and goats. According to the 20th Livestock Census in 2019, India's total livestock population stood at a remarkable 535.78 million, with an impressive milk production of 187.7 million tons in the same year [1]. Among these livestock animals, goat constituted a substantial portion, totalling 148.88 million in India [1]. Sheep and goats hold paramount importance for rural livestock owners in India, primarily sought after for their contributions to milk, meat, and hair production.

Notably, in rural settings, it's common to find livestock owners rearing a significant number of these small ruminants, ranging from a few to over 500 animals within a single farm or flock. Unfortunately, many rural areas lack proper standardized housing, management practices, and hygienic conditions for these animals. This situation often results in small ruminants being confined to smaller areas, increasing the risk of rapid disease transmission within the population. This emphasizes the need for improved livestock management practices, infrastructure, and disease control measures in rural areas to safeguard the health and productivity of these valuable animals.

Goats suffer from various systemic and non-systemic diseases, with respiratory tract diseases being common in veterinary clinics. Factors such as close proximity between animals, environmental factors, housing practices, availability of healthcare services, trained manpower, lack of knowledge, and inappropriate disease prevention strategies contribute to the spread of these diseases. Common respiratory tract diseases include rhinitis, laryngitis, tracheitis, bronchitis, tracheobronchitis, pneumonia, aspiration pneumonia, and nasal bots. Nasal openings act as entry points for infectious pathogens, such as bacteria, fungus, and viruses. Unattended cases of nasal discharge can lead to further infection spread to the lower respiratory tract which makes the treatment difficult in later stage irrespective of underlying etiology [2, 3]. Therefore, it is crucial to collect information on common nasal bacteria in goats with nasal discharge (Picture-1).

The present era is facing a difficult challenge of antimicrobial resistance (AMR) and it is perceived that rural goat owners with or without nomadic lifestyle are using antibiotics and other veterinary drugs without consulting veterinarians. Among all diseases, respiratory illness is one of the most common clinical entities with nasal discharge as clinical symptoms in most of the cases. It is learned that antibiotics such as Oxytetracycline, Tetracycline, Enrofloxacin, Penicillin G etc. are commonly used by goat owners without evaluating antibiogram of bacteria present in such cases with nasal discharge. Hence, such practices contribute to development of AMR in field making it crucial to check susceptibility and resistance of bacteria against common antibiotics. Considering such facts, the present study was carried out to investigate types of bacterial pathogens present in goats with nasal discharge and their antibiogram as a step to check current status of AMR in study population of goats.

Picture 1: Goats with nasal discharge

Materials and Methods

The study was carried out at the Postgraduate Institute of Veterinary Education & Research (PGIVER) and Veterinary Hospital of Kamdhenu University, Rajpur (Nava), Himmatnagar in collaboration with (a) Central Diagnostic Laboratory at Polytechnic in Animal Husbandry, Kamdhenu University, Rajpur (Nava), Himmatnagar, (b) Private farms/flocks of goat in villages nearby campus (*viz.*, Rajpur, Kesharpura Kampa and Khed of Himmatnagar taluka, Sabarkantha district). The study was carried out from September-2021 to March-2022. Total 60 animals were included following two categories where both categories had 30 animals of each (goats; irrespective of age, breed and sex). [I] Category-A (Clinically healthy animals; 30 goats) [II] Category-B (Animals with nasal discharge/respiratory symptoms; 30 goats). Nasal swabs were collected as per methods described by Markey *et al.* (2014) [4] from goats (30 healthy, 30 with nasal discharge) included under Categories (A) and (B) [Picture-2].

Picture 2: Collection of nasal swab from a goats

All the nasal swabs were subjected to bacteriological cultural isolation on general/non-specific media as per methods described by Koneman *et al.* (2012) [5], Thairu *et al.* (2014) [6], Tille (2017) [7], Markey *et al.* (2014) [4] and Mondal (2019) [8]. Bacterial colonies grown on general/non-specific media were collected and subjected to bacteriological cultural isolation on specific isolation media. Bacterial colonies grown on general and specific media were taken on clean microscopic slide using sterile platinum loop to perform staining procedures by use of Gram's stain and Ziehl-Neelsen (ZN) stain for bacterial identification as per methods described by Tille (2017) [7], Markey *et al.* (2014) [4] and Mondal (2019) [8]. The bacterial isolates recovered from nasal swabs were subjected to ABST as per methods described by Tille (2017) [7], Markey *et al.* (2014) [4] and Mondal (2019) [8] to observe antibiogram of bacteria in cases with nasal discharge as a clinical symptom. Data generated through the study requiring statistical analysis were subjected to suitable statistical methods described by Snedecor and Cochran (1990) [9].

Results and Discussion

Bacterial isolates recovered from nasal swabs of healthy goats (Category-A)

The most consistent bacteria isolated from nasal swabs of clinically healthy goats (n=30) was *Staphylococcus* spp. (23.33%; 07/30) followed by *Escherichia coli* (13.33%; 04/30) and *Streptococcus* spp. (06.67%; 02/30). *Klebsiella* spp. and *Salmonella* spp. could not be isolated from healthy goats. Mixed presence of *Staphylococcus* spp. + *Escherichia coli* and *Streptococcus* spp. + *Escherichia coli* was observed in similar numbers of healthy goats (06.67%; 02/30, each).

Bacterial isolates recovered from nasal swabs of goats having nasal discharge (Category-B)

The most consistent bacteria isolated from nasal swabs of goats having nasal discharge was *Staphylococcus* spp. (93.33%; 28/30) followed by *Streptococcus* spp. (83.33%; 25/30), *Escherichia coli* (73.33%; 22/30), *Klebsiella* spp. (53.33%; 16/30) and *Salmonella* spp. (23.33%; 07/30). Mixed presence of

two different isolates was observed highest for *Staphylococcus* spp. + *Escherichia coli* (56.67%; 17/30) followed by *Staphylococcus* spp. + *Klebsiella* spp. (43.33%; 13/30), *Streptococcus* spp. + *Escherichia coli* (16.67%; 05/30), *Staphylococcus* spp. + *Salmonella* spp. (23.33%; 07/30) and *Streptococcus* spp. + *Klebsiella* spp. (10.00%; 03/30). Presence of such bacterial pathogens has also been observed by various scientists in different parts of the world in past [10, 11, 12].

Table 1: Bacterial isolates recovered from healthy goat (n=30)

Bacteria	Goat	
	n=30	%
Staphylococcus spp.	7	23.33
Streptococcus spp.	2	6.67
Escherichia coli	4	13.33
Klebsiella spp.	0	0.00
Salmonella spp.	0	0.00
Staphylococcus spp. + *Escherichia coli*	2	6.67
Staphylococcus spp. + *Klebsiella* spp.	0	0.00
Streptococcus spp. + *Escherichia coli*	2	6.67
Streptococcus spp. + *Klebsiella* spp.	0	0.00

Table 2: Bacterial isolates recovered from goat having nasal discharge as a clinical symptom (n=30)

Bacteria	Goat	
	n=30	%
Staphylococcus spp.	28	93.33
Streptococcus spp.	25	83.33
Escherichia coli	22	73.33
Klebsiella spp.	16	53.33
Salmonella spp.	7	23.33
Staphylococcus spp. + *Escherichia* coli	17	56.67
Staphylococcus spp. + *Klebsiella* spp.	13	43.33
Streptococcus spp. + *Escherichia coli*	5	16.67
Streptococcus spp. + *Klebsiella* spp.	3	10.00
Staphylococcus spp. + *Salmonella* spp.	7	23.33

Antibiogram of bacterial isolates from goats having nasal discharge

In the present study, an antibiogram of bacterial isolates recovered from nasal swabs of goats having nasal discharge as a clinical symptom was observed by performing ABST using different antibiotics. The antibiogram was interpreted in terms of reducing sensitivity towards different antibiotics in percentage.

Antibiogram of bacterial isolates recovered from goats having nasal discharge

Amongst *Staphylococcus* spp. isolates (n=28), all isolates were found sensitive to high concentration of Gentamicin (120 mcg; 100.00%; 28/28) followed by low concentration of Gentamicin (10 mcg; 96.43%; 27/28); Ciprofloxacin (92.86%; 26/28); Streptomycin (71.43%; 20/28); Chloramphenicol and Cotrimoxazole (53.57%; 15/28, each); Cefixime (28.57%; 08/28); Tetracycline (10.71%; 03/28); Ampicillin/Sulbactam and Erythromycin (03.57%: 01/28, each). These isolates were resistant to Ampicillin, Cefotaxime and Penicillin G.

Amongst *Streptococcus* spp. isolates (n=25), all isolates were found sensitive to high concentration of Gentamicin (120 mcg; 100.00%; 25/25) followed by low concentration of Gentamicin (10 mcg; 96.00%; 24/25); Ciprofloxacin and Streptomycin (72.00%: 18/25, each); Chloramphenicol (44.00%; 11/25); Cotrimoxazole (40.00%; 10/25); Cefixime (12.00%; 03/25) and Tetracycline (04.00%; 01/25). These isolates were resistant against Ampicillin, Ampicillin/Sulbactam, Cefotaxime, Erythromycin and Penicillin G.

Amongst *Escherichia coli* isolates (n=22), all isolates were found sensitive to high concentration of Gentamicin (120 mcg; 100.00%; 22/22) followed by low concentration of Gentamicin (10 mcg; 86.36%; 19/22); Streptomycin (77.27%; 17/22); Chloramphenicol (13.64%; 03/22); Cefixime and Cotrimoxazole (09.09%; 02/22). These isolates were resistant against Ampicillin, Ampicillin/Sulbactam, Cefotaxime, Ciprofloxacin, Erythromycin, Penicillin G and Tetracycline.

Table 3: Antibiogram of bacterial isolates recovered from goat (n=30) showing sensitive results against commonly used antibiotic drugs

Antibiotic	*Staphylococcus* spp. (n=28)		*Streptococcus* spp. (n=25)		*Escherichia coli* (n=22)		*Klebsiella* spp. (n=16)		*Salmonella* spp. (n=7)	
	No.	%	No.	%	No.	%	No.	%	No.	%
Ampicillin (AMP; 25 mcg)	0	0.00	0	0.00	0	0.00	0	0.00	0	0.00
Ampicillin/Sulbactam (A/S; 10/10 mcg)	1	3.57	0	0.00	0	0.00	0	0.00	0	0.00
Cefixime (CFM; 5 mcg)	8	28.57	3	12.00	2	9.09	0	0.00	0	0.00
Cefotaxime (CTX; 30 mcg)	0	0.00	0	0.00	0	0.00	0	0.00	0	0.00
Chloramphenicol (C; 30 mcg)	15	53.57	11	44.00	3	13.64	0	0.00	0	0.00
Ciprofloxacin (CIP; 5 mcg)	26	92.86	18	72.00	0	0.00	14	87.50	1	14.29
Cotrimoxazole (COT; 25 mcg)	15	53.57	10	40.00	2	9.09	12	75.00	2	28.57
Erythromycin (E;15 mcg)	1	3.57	0	0.00	0	0.00	0	0.00	0	0.00
Gentamicin (GEN; 10 mcg)	27	96.43	24	96.00	19	86.36	15	93.75	6	85.71
Gentamicin (HLG; 120 mcg)	28	100.00	25	100.00	22	100.00	16	100.00	7	100.00
Penicillin G (P; 10 units)	0	0.00	0	0.00	0	0.00	0	0.00	0	0.00
Streptomycin (S; 10 mcg)	20	71.43	18	72.00	17	77.27	0	0.00	0	0.00
Tetracycline (TE; 30 mcg)	3	10.71	1	4.00	0	0.00	0	0.00	0	0.00

Conclusion

In the present study, bacteria such as *Staphylococcus spp.*, *Streptococcus spp.*, *Escherichia coli*, *Klebsiella spp.*, and *Salmonella spp.* could be isolated from nasal discharge of goats with clinical illness. The ABST revealed variable sensitivity to common antibiotics such as Gentamicin, Ciprofloxacin, Streptomycin, Clotrimoxazole, and Chloramphenicol. However, the bacteria showed complete resistance to Penicillin G, Ampicillin, and Cefotaxime. This suggests a concerning issue of antimicrobial resistance among free-living goat populations in the study area. To better understand the extent of this problem, further research is warranted in larger areas covering more animals in different geographical regions, to assess the current status of bacterial resistance against commonly used antibiotics.

Conflict of Interest

Authors declare no conflict of interest with regards to funding. All the cases were included in the study after obtaining owners'

consent. The project was approved and recommended by the advisory committee and Director of Research & Dean PG Studies.

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