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Antibiogram study of *Pasteurella multocida* strains isolated from pneumonic sheep and goats in Warangal region of Telangana

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Abstract
Pasteurella multocida causing pneumonic pasteurellosis is one of the most economically important infectious diseases of small ruminants having a high prevalence rate and worldwide in distribution. A total of 16 isolates were obtained from the 191 pneumonic sheep and goat. Antibiotic susceptibility testing was performed using 12 antibiotics by the disc diffusion method. In this study, we found that the *P. multocida* isolates were found 100% sensitive to ceftriaxone, 93.75% for cotrimoxazole and enrofloxicin, 87.5% for amoxicillin-clavulanic acid, cefotaxime and tetracycline, 81.25% for amoxicillin/sulbactum, 75% for chloramphenicol and oxacillin, 68% for gentamicin and Co-trimaxazole, 62.5% for erythromycin and 60% for Sulphadiazine. Emergence of drug resistance in the isolates warrants the judicious use of antibiotics after performing antimicrobial susceptibility testing, wherever possible.

Keywords: Antibiotic sensitivity, *Pasteurella multocida*, Sheep and goats, Telangana

1. Introduction
Pasteurella multocida is a Gram-negative bacterium belonging to the family *Pasteurellaceae*. Different capsular types are associated with wide variety of diseases in animals and birds [1]. *P. multocida* type A is known to cause pneumonic pasteurellosis in small ruminants. However, other capsular types have been frequently isolated from cases of pneumonia in sheep and goat [2] posing major problem in disease control and prevention. Although antibiotics were originally used to treat pasteurellosis, their repeated and indiscriminate usage has resulted in the development of resistance in a number of strains. Currently, antimicrobial resistance is a global concern, therapeutic alternatives might be difficult, and understanding antimicrobial susceptibility is crucial when applying rationale efficient therapy [3]. Due to scarcity of antimicrobial resistance data on *P. multocida*, the present study was undertaken to study the antimicrobial susceptibility of *P. multocida* isolated from sheep and goat in the Warangal region of Telangana state.

2. Materials and Methods
A total of 16 *P. multocida* isolates recovered from 191 samples collected from pneumonic sheep and goat and previously confirmed as *P. multocida* by cultural, biochemical and molecular testing were included in the study [4]. Bauer-Kirby [5] used a modified disc diffusion method to conduct an antibiotic susceptibility test using antibiotic test discs. Test culture of *P. multocida* enriched in BHI broth and incubated at 37 ℃ for 7-8 h. The turbidity of the culture was adjusted to McFarland standard tube 0.5 before charging the sheep blood agar plate. Inoculum was spread with the help of sterile cotton swab evenly over entire surface of the sheep blood agar plates by swabbing back and forth across the agar. Antibiotic discs were kept aseptically by using sterile forceps. The plates were then incubated at 37 ℃ for 16-24 h. The zones of growth inhibition around the antibiotic discs were measured to the nearest millimetre. The zone diameter of each drug were interpreted using the criteria chart provided [6].

Based on disc diffusion diameters results were qualitatively interpreted as susceptible, intermediate and resistant. The antibiotics Amplicillin (10 µg), Amoxicillin (10 µg), Amoxicillin-Clavulanic acid (30 µg), Erythromycin (10 µg), Chloramphenicol (30 µg), Ceftriaxone (30 µg), Enrofloxicin (10 µg), Gentamicin (10 µg), Cotrimoxazole (30 µg), Oxacillin (30 µg), Ceftriaxone (10 µg), Tetracycline (30 µg), Co-trimaxazole (25 µg) and Sulphadiazine (100 µg) were used in the present study.

3. Results

In the present study, all the isolates (16) of *P. multocida* were tested for antibiotic sensitivity pattern by disc diffusion method against 14 commonly used antibiotics. The percentage sensitivity of the isolates against various antibiotics tested is in the order of Ceftriaxone (100%), Cotrimoxazole and Enrofloxicin (93.75%), Cefotaxime, Tetracycline, Amoxicilin/clavulinic acid (87.5%), Amoxicillin/Sulbactum (81.25%), Chloromphenicol, Amoxicillin and Oxacillin (75%), gentamicin (68%), Co-trimaxazole (68%), Erythromycin (62.5%), and Sulphadiazine (60%) (Fig 1). The antibiotics Amoxicillin, Chloramphenicol and Enrofloxacin were found to be intermediately sensitive for the isolates in the order 12.5%, 12.5% and 6.25% respectively. The antibiogram profile of the isolates are presented in Table 1 and depicted in Fig 1.

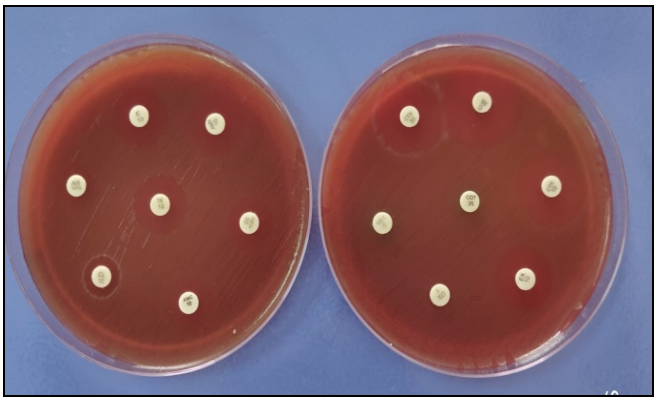

Fig 1: Showing antibiotic sensitivity testing of *P. multocida* isolate on sheep blood agar

Table 1: Antibiotic sensitivity / resistance pattern observed among 16 *P. multocida* isolates.

S. No	Antibiotic	Sensitive (%)	Intermediate (%)	Resistance (%)
1	Amoxicillin/Sulbactum	81.25	0	18.75
2	Amoxicillin	75	12.5	12.5
3	Amoxicillin-Clavulanic acid	87.5	0	12.5
4	Chloramphenicol	75	12.5	12.5
5	Ceftriaxone	100	0	0
6	Cefotaxime	87.5	0	12.5
7	Cotrimoxazole	93.75	0	6.25
8	Enrofloxacin	93.75	6.25	0
9	Erythromycin	62.5	0	37.5
10	Gentamicin	68	0	32
11	Oxacillin	75	0	25
12	Tetracycline.	87.5	0	12.5
13	Co-trimaxazole	68	0	32
14	Sulphadiazine	60	0	40

4. Discussion

Antibiotic sensitivity testing of the isolates: The early initiation of antibiotic treatment is very important to prevent treatment failures. The indiscriminate and extensive use of antibiotics both for prophylaxis and growth promotion in animals, resulted in developing resistance to antibiotics and even multidrug resistant strains of *P. multocida* have emerged [7, 8]. In the present study, *in vitro* antibiotic sensitivity testing was carried out by 12 commonly used antibiotics. The study revealed that the isolates were sensitive to ceftriaxone (100%) followed by Cotrimoxazole and Enrofloxacin (93.7%), Cefotaxime, Amoxicillin- Clavulanic acid and Tetracycline (87.5%). Similar antibiotic sensitivity and resistance profiles were reported by several other workers [9-14]. The study also observed that the resistance to sulphate group of drugs has been observed at 32% for Co-trimaxazole and 40% for sulphadiazine, the drugs of choice for pasteurellosis in animals. This warrants judicious use of antibiotics in treating the cases of pneumonic pasteurellosis in sheep and goats and wherever possible, antibiotic therapy should be initiated after testing the isolates for antibiotic sensitivity to prevent the development of emergence of drug resistance in *P. Multocida*.

5. Conclusion

In the present study, although many commonly available antibiotics are found to be sensitive to P. multocida isolates, the development of multi drug resistance strains and emergence of resistance to sulphate group of drugs, which were considered to be drugs of choice for treating pasteurellosis in animals warrants for judicious use and selection of appropriate antibiotic to prevent the treatment failures in animals.

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