



# Antibiogram of *Escherichia coli* isolates from faecal samples of neonatal calf diarrhoea in Wayanad district\*

M. Xavier<sup>1</sup>, A. Janus<sup>2</sup>, P. M. Deepa<sup>3</sup>, K. C. Bipin<sup>4</sup>, B. P. Habeeb<sup>5</sup>, K. Vijayakumar<sup>6</sup>

Department of Veterinary Epidemiology and Preventive Medicine, College of Veterinary and Animal Sciences, Pookode, Kerala Veterinary and Animal Sciences University.

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## Abstract

Thirty faecal samples were collected from diarrhoeic neonatal calves reared in organized farms and households of Wayanad district. The collected rectal swabs were subjected to bacterial isolation and biochemical tests. Based on their cultural and biochemical characteristics isolates were confirmed as *Escherichia coli*. Based on clinical signs and bacterial isolation calves were diagnosed as affected with colibacillosis. Antibiotic sensitivity patterns of the isolates were evaluated using standard disc diffusion method in Muller Hinton Agar. Antibiogram revealed susceptibility to Chloramphenicol (70.0 per cent), Trimethoprim/Sulphamethoxazole (60.0 per cent), Ceftriaxone (33.3 per cent), Ampicillin (33.3 per cent) and Tetracycline (3.3 per cent). The sensitivity patterns of the isolates to the three antibiotics viz. Trimethoprim/Sulphamethoxazole, Ceftriaxone and Ampicillin correlated well with clinical response.

**Keywords:** *Escherichia coli*, Antibiogram, Wayanad,

Among the economically important diseases of bovine calves, neonatal diarrhoea caused by *Escherichia coli*, is very important and is associated with morbidity, retarded growth and mortality. Incidence of colibacillosis were reported from different states of India including Kerala. Amritha *et al.*, (2018) detected pathogenic *Escherichia coli* strains from diarrheic calves of Thrissur district.

Control of this disease requires antibiotic therapy along with supportive therapy. Selection of antibiotics for colibacillosis is very important as antibiotic resistance to this pathogen has been reported by many scientists (Malik *et al.*, 2013; Moreno *et al.*, 2006). Hence this study was undertaken with the objective of analysis of antibiogram and detection of suitable antibiotics against *Escherichia coli* isolates from neonatal calves.

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1. MVSc scholar and corresponding author (Email : drxaviervet@gmail.com)

2,3,4. Assistant Professors

5. Assistant Professor, Department of Clinical Medicine

6. Professor and Head, Department of Veterinary Epidemiology and Preventive Medicine, CVAS, Mannuthy

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## Materials and methods

Thirty calves with neonatal diarrhoea from various parts of Wayanad district were selected for the study.

Faecal samples were collected directly from the rectum of diarrhoeic calves in a sterile collection bottle for isolation and identification of *Escherichia coli* and subsequent antibiogram studies.

For isolation of *Escherichia coli* from faecal samples standard protocol by Quinn *et al.* (2011) was followed. All the samples were first inoculated on MacConkey's agar and the gram-negative lactose fermenting bacilli obtained were further plated into Eosin Methylene Blue media. Colonies with typical metallic sheen were preserved in Brain Heart Infusion (BHI) slants for biochemical tests *viz.* Indole test, Methyl Red test, Voges-Proskauer test, Citrate test, Urease Test and Motility test. Based on the cultural, morphological, staining characteristics and biochemical reactions the isolates were confirmed as *Escherichia coli*. Based on clinical signs and results of biochemical characterization the condition was diagnosed as colibacillosis.

*In-vitro* antibiotic sensitivity of isolates was studied using disc diffusion technique (Bauer *et al.*, 1966). A loopful of *Escherichia coli* inoculum was applied on the surface of a Mueller-Hinton agar plate using a sterile cotton swab and the plate was kept covered for 15 minutes at room temperature for drying the inoculum. The antibiotic discs were then placed 20 mm apart and they were gently pressed on to the surface of the agar to ensure contact.

Plates were then incubated at 37°C for 18 to 24hrs.

Himedia antibiotic discs Ampicillin (10 mcg), Ceftriaxone (30 mcg), Trimethoprim/Sulphamethoxazole (1.25mcg / 23.75mcg), Chloramphenicol (10mcg) and Tetracycline (30mcg) were used in this study

## Results and discussion

Antibiogram revealed susceptibility to Chloramphenicol (70.0 per cent), Trimethoprim/Sulphamethoxazole (60.0 per cent), Ceftriaxone (33.3 per cent), Ampicillin (33.3 per cent) and Tetracycline (3.3 per cent).

Resistance levels were Tetracycline (83.3 per cent), Ceftriaxone (50.0 per cent), Ampicillin (30.0 per cent) Trimethoprim/Sulphamethoxazole (16.7 per cent) and Chloramphenicol (6.7 per cent).

The antibiotic therapy was initiated with Sulphadiazine trimethoprim and with response and antibiogram results antibiotic was changed if necessary.

The sensitivity patterns of the isolates correlated well with clinical response. The antibiogram of the isolates is represented in Table 1.

Moreno *et al.* (2006) found out susceptibility of *Escherichia coli* against Trimethoprim/Sulphamethoxazole (70.7 per cent) which is well correlated with the present study in which results revealed a 60 per cent of Trimethoprim / Sulphamethoxazole.

Zhang *et al.* (2012) reported 89.69

**Table 1. Antibiogram of *Escherichia coli* isolates.**

| Antibiotic used                | Sensitive      |          | Intermediate sensitive |          | Resistant      |          |
|--------------------------------|----------------|----------|------------------------|----------|----------------|----------|
|                                | No. of samples | Per cent | No. of samples         | Per cent | No. of samples | Per cent |
| Chloramphenicol                | 21             | 70.0     | 7                      | 23.3     | 2              | 6.7      |
| Trimethoprim/Sulphamethoxazole | 18             | 60.0     | 7                      | 23.3     | 5              | 16.7     |
| Ceftriaxone                    | 10             | 33.3     | 5                      | 16.7     | 15             | 50.0     |
| Ampicillin                     | 10             | 33.3     | 11                     | 36.7     | 9              | 30.0     |
| Tetracycline                   | 1              | 3.3      | 4                      | 13.3     | 25             | 83.3     |

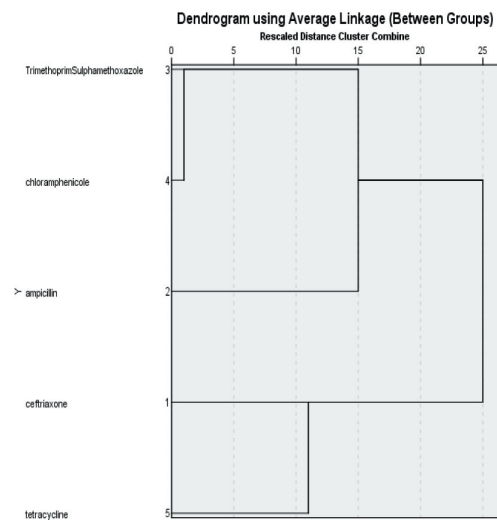
per cent resistance against tetracycline. This resistance is highly correlating with *Escherichia coli* (83.3 per cent resistance) isolates of Wayanad district.

White *et al.* (2000) reported 90 per cent of *Escherichia coli* isolates from Georgia were resistant against Chloramphenicol and that *flo* gene was responsible for the resistance. But in this study 70 per cent of *Escherichia coli* isolates were sensitive to Chloramphenicol and absence of *flo* gene in these isolates maybe the reason for sensitivity.

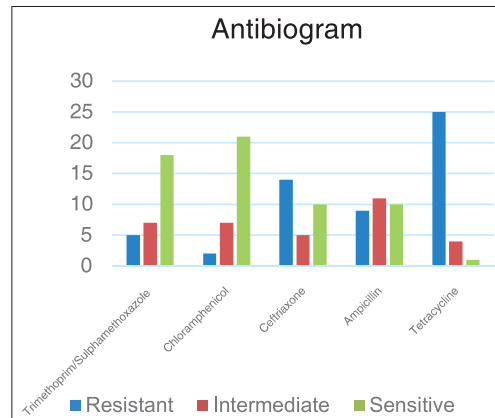
Li *et al.* (2017) reported 7.3 per cent resistance to Ceftriaxone against *Escherichia coli* from China which is not in accordance with the current study. In the present study, 50 per cent of the *Escherichia coli* isolates showed resistance to Ceftriaxone.

Mohammed *et al.* (2019) reported a marked resistance of *Escherichia coli* against Ampicillin, *i.e.* 100 per cent from Egypt. But in the present study, 33.3 per cent of the *Escherichia coli* isolates from Wayanad, showed sensitivity to Ampicillin.

Even though dendrogram analysis (Fig. 1) revealed the close relation of action between Trimethoprim/Sulphamethoxazole and Chloramphenicol, *i.e.* variance between these two antibiotics were below 10 per cent, the current study did not use chloramphenicol in treatment as it cannot be used in food



**Fig 1. Dendrogram comparison of anti-biogram results.**



animals. But as per current antibiogram result Chloramphenicol can be used in MDR *Escherichia coli* in human medicine.

To conclude, *Escherichia coli* isolated from Wayanad district, exhibited much sensitivity towards Trimethoprim/Sulphamethoxazole and Chloramphenicol and high resistance towards Ampicillin and Ceftriaxone.

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