

Epidemiological study on enterotoxaemia in small ruminants from Namakkal and Karur districts of Tamil Nadu

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Received: 20.11.25; Accepted: 28.12.25

ABSTRACT

Enterotoxaemia remains one of the most economically important *Clostridial* diseases of small ruminants, which often occurs in peracute form with sudden mortality without any premonitory signs. Thus, the present study was undertaken to determine the occurrence of enterotoxaemia in sheep and goat flocks with the history of sudden death in Namakkal and Karur regions of Tamil Nadu. Totally, 62 flocks with the history of sudden death were investigated and samples like intestinal contents in necropsied animals and faecal swabs from live animals were collected. Among these, 22 flocks were found positive for different toxinotypes of *Clostridium perfringens* by multiplex PCR, which include 14 sheep flocks and 8 goat flocks. The study revealed that the major etiological agent for sudden death in small ruminants was *C. perfringens* type D and highest occurrence was recorded in sheep. Similarly, male lambs and kids were more susceptible than females. With respect to age, lambs and kids in the age group of 4 to 6 months were mostly affected. The findings also revealed, enterotoxaemia is more prevalent in the extensive system of management. Among the different toxinotypes, Type D is most commonly observed in both dead and live animals, followed by Type A. Thus, the present study highlights the continued endemicity of enterotoxaemia in this region and stresses the need for periodical vaccination, proper dietary management, disease surveillance and regular monitoring for effective disease control.

Keywords: Enterotoxaemia, small ruminants, occurrence, mPCR

INTRODUCTION

Sheep and goat husbandry in India remains largely traditional, with animals reared under extensive grazing systems that predispose them to soil-borne infections and parasitic infestations. Among these, enterotoxaemia is one of the most devastating diseases of young feedlot sheep and goats, leading to high mortality and significant economic losses¹. Seasonal outbreaks, particularly during the onset of the monsoon, are common even in vaccinated flocks².

Enterotoxaemia in small ruminants is caused by *Clostridium perfringens*, a Gram positive, spore forming anaerobe, usually a commensal organism of the intestinal tract that proliferates rapidly under favourable intestinal conditions, resulting in the production of potent toxins. Of its five toxinotypes (A–E), type D, producing alpha and epsilon toxin, is mainly responsible for enterotoxaemia in small ruminants³.

While case history, clinical signs and gross pathological findings are useful for arriving at presumptive diagnosis of enterotoxaemia by *C. perfringens* in sheep and goats, definitive diagnosis of these diseases cannot be established without molecular confirmation of relevant toxin genes⁴. Recent studies have investigated the role of different toxin genes of *C. perfringens* in the intestinal contents in the definitive diagnosis of the diseases⁵. Based on these studies, the definitive diagnosis of enterotoxaemia relies on identification of specific toxin genes in the intestinal contents in conjunction with characteristic clinical signs and gross lesions of the affected animals.

The disease occurs sporadically or in outbreaks, often influenced by sudden dietary changes, climatic stress and management practices that favour the rapid intestinal proliferation of *C. perfringens*. The occurrence varies widely between regions and seasons, reflecting differences in husbandry systems,

How to cite this article : Sivaraj, S., Sasikala, M., Ramya, K. and Srinivasan, P. 2026. Epidemiological study on enterotoxaemia in small ruminants from Namakkal and Karur districts of Tamil Nadu. Indian J. Vet. Pathol., 50(2) : 150-154.

feeding patterns and vaccination coverage. Young and fast-growing animals are particularly vulnerable and the disease often remains underreported due to its per-acute nature.

Therefore, evaluating the occurrence of enterotoxaemia across different age groups, management systems and different seasons remains crucial for understanding disease dynamics and formulating evidence-based control measures and effective vaccination programmes. With this background, the present communication reports the occurrence of enterotoxaemia in

sheep and goats of Namakkal and Karur districts of Tamil Nadu, a well-known hub for small-ruminant farming.

MATERIAL AND METHODS

Study area

Occurrence of enterotoxaemia was assessed in sheep and goat flocks located in and around the Namakkal and Karur districts of Tamil Nadu during the period from December 2024 to September 2025.

Study population

During the study period, a total of 75 dead animals from 62 flocks were investigated and 31 flocks were suspected to have enterotoxaemia outbreaks. With respect to live animals, a total of 181 faecal samples were collected from the flocks confirmed with enterotoxaemia outbreak. Data like species, sexes, age, month of occurrence and different toxinotypes were recorded.

Sampling

Case history

Dead animals with a history of sudden death, neurological signs, abdominal bloating along with good body condition were considered for sample selection and subsequent screening for enterotoxaemia by multiplex PCR. Meanwhile, samples from live animals were collected based on the presence of clinical signs such as diarrhoea and abdominal distension in flocks with a confirmed enterotoxaemia outbreak.

Sample collection

During the study period, a total of 75 animals from 62 flocks were reported to have sudden death and postmortem samples were collected from these cases. The collected samples were subjected to isolation by culture method and confirmed by molecular detection of *C. perfringens* toxin genes associated with enterotoxaemia by multiplex PCR. Faecal swabs and blood samples were collected from 181 live animals that manifested clinical signs such as depression and diarrhoea and subjected to bacteriological and molecular detection of *C. perfringens*.

Laboratory diagnostics

The collected samples, such as intestinal contents and faecal swabs, were inoculated into Robertson's cooked meat medium and milk medium, followed by incubation at 37 °C for 24 hours. Subsequently, the positive samples

were then subcultured onto blood agar and egg yolk agar and then incubated for 24 hours⁶. Colonies exhibiting characteristic growth were then subjected to molecular detection of different toxinotypes of *C. perfringens* by multiplex PCR⁷. The intestinal contents were also tested for the presence of epsilon toxin for confirmation of type D enterotoxaemia by using commercial ELISA kit (Bio-X, Belgium; BIO K 268).

RESULTS AND DISCUSSION

Laboratory Diagnosis of Enterotoxaemia Using PCR and ELISA

DNA was extracted from the positive cultures of *C. perfringens* by the hot and cold lysis method. Multiplex PCR was carried out for the detection of different toxin genes of *C. perfringens* using the extracted DNA and specific primers. Polymerase chain reaction yielded positive results in 29 out of 75 intestinal samples collected during necropsy and 69 out of 181 clinical samples collected from enterotoxaemia suspected animals.

ELISA performed on intestinal contents of PCR positive samples for the detection of epsilon toxin revealed varying levels of epsilon toxin, ranging from 27.2% to 87.6%, which were significantly than the levels detected in other clinically suspected animals.

Species-wise occurrence

In the present study, the distribution of enterotoxaemia differed between sheep and goats, indicating a species-related variation in susceptibility to *C. perfringens* infection. The occurrence of enterotoxaemia was more in sheep with 19 cases (45.23%) from 14 flocks than in goats with 10 cases (30.30%) from 8 flocks (Table 1). Similarly, in live animals, out of 181 samples screened, 69 samples were positive for enterotoxaemia, with 52 (40.94%) samples from sheep and 17 (31.48%) samples from goats (Table 2). Such differences have been documented in earlier reports as well. 25% of sheep from 20 flocks in Egypt exhibited clinical signs of enterotoxaemia, with a mortality rate of 16.25%, highlighting the significant disease burden in ovine populations observed by⁸. Similarly,⁷ reported a higher prevalence of *C. perfringens* in sheep (72.36%) than in goats (60%), suggesting that sheep may be comparatively more prone to infection and clinical disease expression.

Table 1. Species-wise occurrence of Enterotoxaemia in affected flocks (Dead animals)

S.No.	Species	No of animals died with history sudden death	No of ET positive animals	Per cent positivity
1.	Sheep	42	19 (14 flocks)	45.23
2.	Goats	33	10 (8 flocks)	30.30
	Total	75	29 (22 flocks)	38.66

Table 2. Species-wise occurrence of Enterotoxaemia in affected flocks (Live animals)

S.No.	Species	No of sample collected	No of ET positive samples	Per cent positivity
1.	Sheep	127	52	40.94
2.	Goats	54	17	31.48
	Total	181	69	38.12

Table 3. Sex-wise occurrence of Enterotoxaemia in affected flocks

S.No.	Sex	Total no. of samples		No. of Positives		Per cent positivity	
		Dead	Live	Dead	Live	Dead	Live
1.	Male	41	110	17	48	41.46	43.63
2.	Female	34	71	12	21	35.29	29.58
	Total	75	181	29	69	38.66	38.12

Sex-wise occurrence

In the present study, males showed a higher occurrence of enterotoxaemia than females, *viz.*, 41.46% of dead animals and 43.63% of live animals were males (Table 3). Similar findings

were reported earlier^{9,10} who also observed a higher prevalence of enterotoxaemia in males. This pattern might be due to managerial practices where males are often maintained for fattening and are commonly preferred in intensive farming systems. Such management conditions may expose them to higher nutritional stress and sudden dietary changes, making them more susceptible to *C. perfringens* infection.

Age-wise occurrence

In the present study, enterotoxaemia occurred in all age groups, but the highest occurrence was seen in animals aged 4–6 months, both in dead (57.14%) and live (46.47%) animals. The 7–12 month group followed this, while the lowest occurrence was recorded in animals above one year of age (Table 4). These findings indicate that young growing animals are more susceptible to the

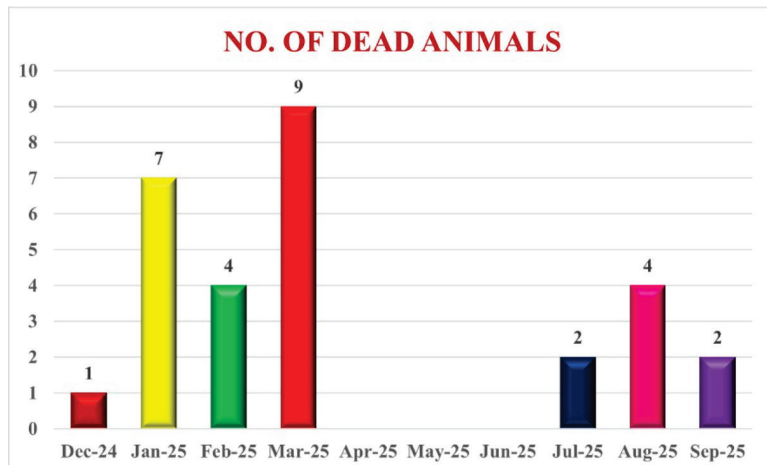


Fig 1. Month-wise occurrence of Enterotoxaemia among dead animals

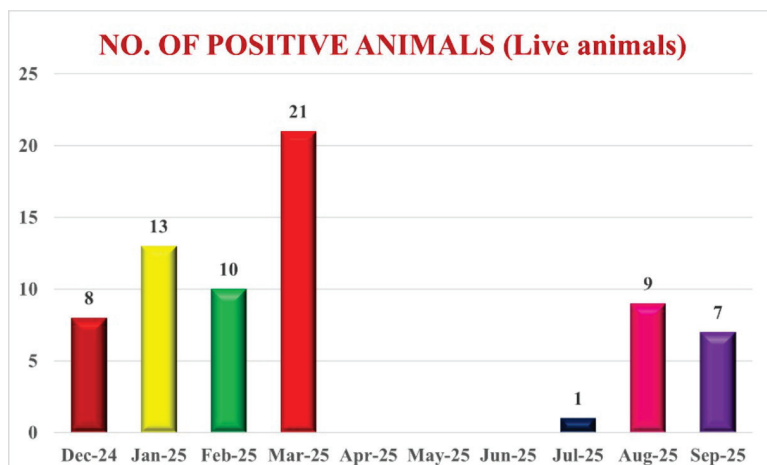


Fig 2. Month-wise occurrence of Enterotoxaemia among live animals

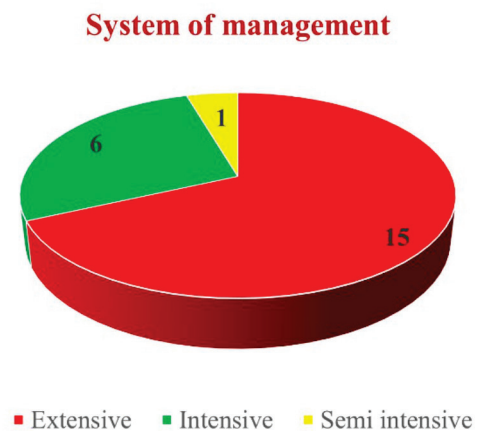


Fig 3. Occurrence of Enterotoxaemia based on the System of Management

Table 4. Age-wise occurrence of Enterotoxaemia in affected flocks

S. No.	Age group	Total no. of samples		No. of positive		Per cent positivity	
		Dead	Live	Dead	Live	Dead	Live
1.	0 – 3 Months	21	52	5	16	23.80	30.77
2.	4 – 6 Months	28	71	16	33	57.14	46.47
3.	7 – 12 Months	12	48	6	19	50.00	39.58
4.	Above 12 Months	14	10	2	01	14.28	10.00
	Total	75	181	29	69	38.66	38.12

Table 5. Occurrence of different toxinotypes of *C. perfringens*

S. No.	<i>Clostridium</i> toxinotypes	Dead			Live			Total no of animals
		Sheep	Goats	Total	Sheep	Goats	Total	
1.	<i>C. perfringens</i> type A	7	3	10	21	6	27	37
2.	<i>C. perfringens</i> type B	0	0	0	0	0	0	0
3.	<i>C. perfringens</i> type C	0	0	0	0	0	0	0
4.	<i>C. perfringens</i> type D	12	7	19	31	11	42	61
	Total	19	10	29	52	17	69	98

disease. Seroprevalence of epsilon toxin was highest in sheep aged 6–12 months, followed by animals below 6 months and lowest in adults¹⁰.

Month-wise occurrence

In the present study, the highest number of enterotoxaemia-related mortality occurred during March, with 9 animals out of 14 animals examined (64.28%), followed by January with 7 mortality (50.00%) and February and August with 4 mortalities each (Figure 1). A similar temporal trend was noted in live animals, with 21 positive animals (35.00%) in March, followed by January with 13 positive animals (38.23%) and February with 10 positive animals (50.00%) (Figure 2). The clustering of cases during the late winter and early summer months indicates that seasonal factors may influence disease expression.

The present findings agree previous worker¹¹ who reported that *C. perfringens* infection in small ruminants was more frequent during winter and autumn, suggesting that cooler seasons favour disease occurrence. Likewise, ¹²emphasized that humid conditions and rainy periods predispose sheep to enterotoxaemia, indicating that fluctuations in environmental moisture may enhance bacterial proliferation or toxin expression. Peak occurrence during the monsoon season in unvaccinated lambs from Andhra Pradesh, highlighting the role of climatic stressors and management conditions documented by ².

System of management

In contrast to the earlier worker¹⁴ who observed a higher prevalence of *Clostridium perfringens* in closed (intensive) farms, the present study showed a different distribution

pattern. Among the 22 positive flocks detected, 15 (68.18%) were from extensive systems, 6 (85.71%) from intensive farms and 1 (50.00%) from a semi-intensive system (Figure 3). The higher occurrence of enterotoxaemia in extensive (open) farms may be due to frequent grazing, wider environmental exposure, irregular dietary changes during grazing shifts, the ingestion of lush pasture or high-carbohydrate feeds and improper vaccinations.

Toxinotypes

In the present study, *C. perfringens* type D was the predominant toxinotype in both dead (65.51%) and live (60.86%) animals, followed by type A, while types B and C were not detected (Table 5). The higher occurrence of type D in clinically affected animals coincides with earlier observations, who reported that type D constituted 45.76% of isolates from diseased sheep¹³. Similarly, consistent involvement of type D in field outbreaks of enterotoxaemia has been reported^{2,14}. The detection of type A in both live and dead animals also agrees with the reports^{2,8} who observed substantial proportions of type A in suspected and healthy animals, suggesting that type A remains a common intestinal inhabitant that may become pathogenic under conducive conditions. With agreement to this, in our study, a total of 10 dead animals were positive for *C. perfringens* type A and 27 live animals were positive for *C. perfringens* type A. The absence of toxinotypes B and C in the present work is also in agreement with recent studies, which also documented their limited or negligible occurrence^{2,8}.

CONCLUSION

In conclusion, the present study provides

a comprehensive overview of the occurrence of enterotoxaemia in small ruminants across different species, age groups, sexes, seasons and management systems. The distribution of toxinotypes further highlights the epidemiological complexity of the disease. These findings emphasize the continued vulnerability of young animals and the critical role of management and environmental factors in disease occurrence. Overall, the insights gained from this study can support the development of more targeted vaccination programmes, strengthen preventive health strategies, and guide improvements in farm management practices to mitigate the burden of enterotoxaemia in endemic areas.

Financial support & sponsorship: None.

Conflicts of interest: None.

Use of artificial intelligence (AI)-Assisted Technology for manuscript preparation: The authors confirm that there was no use of AI-assisted technology for assisting in the writing of the manuscript and no images were manipulated using AI.

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