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Journal of Medical

Bacteriology



## Journal of Medical Bacteriology

# **Causes of Lamb Mortality in the Lacaune Sheep Breed in Iran**

Hossein Esmaeili<sup>1\*</sup>, Mohammad Mahdi Safari<sup>1</sup>, Mona Hamedi<sup>1</sup>

1 Department of Microbiology and Immunology, Faculty of Veterinary Medicine, University of Tehran, Tehran, Iran.

ARTICLE INFO	ABSTRACT			
<i>Article type:</i> Research Article	<b>Background</b> : Neonatal mortality is a significant multifactorial problem that affects flock productivity. Different infectious and non-infectious factors have been attributed to lamb losses. The			
Article history:Received:23Aug2023Revised:06Sep2023Accepted:28Oct2023Published:14Nov2023	objective of the present study was to identify the major causes of neonatal mortality. <i>Methods:</i> In a 3-months period, 114 Lacaune breed lambs died of which 52 animals were lost with clinical signs of infection. Heart and lung samples were collected and transmitted to the laboratory. Microbiological cultures following biochemical tests were conducted to identify bacterial infection of the lambs.			
Keywords: Bacteria, Escherichia coli, Lacaune, Lamb Mortality, Sheep.	<b>Results:</b> A total of 52 out of 114 lambs (45.6%) were infected with bacteria. The bacteria including <i>Escherichia coli</i> (30.8%), <i>Proteus mirabilis</i> (19.2%), <i>Pasteurella multocida</i> (1.5%), <i>Corynebacterium pseudotuberculosis</i> (7.7%), <i>Mannheimia haemolytica</i> (3.9%), <i>Staphylococcus epidermidis</i> (1.9%) were isolated. The role of the identified pathogens was more significant in lamb death compared to non-infection causes. Diarrhea was the most infectious disease followed by septicemia and pneumonia. Escherichia coli was isolated significantly more than other bacterial agents (30.8%). <i>Conclusion:</i> The present findings showed Escherichia coli as the most common pathogen leading to lamb losses in the first two weeks of life. The hygienic practice which encompasses both environment and milking equipment, periodical flaming, and providing dry and clean bedding can dramatically decrease the infection among newborn animals. Reduction of the birth density and avoiding synchronization until the expert and labor capacity has reached the optimal level are necessary actions to reduce infectious diseases.			

• Please cite this paper as: Esmaeili H, Safari MM, Hamedi M Causes of Lamb Mortality in the Lacaune Sheep Breed in Iran. J Med Bacteriol. 2023; 11 (5, 6): pp.8-16.

#### Introduction

Neonatal mortality is a significant multifactorial problem which affects a flock productivity (1, 2) and has serious constraints to profitability of small ruminant's farms. Iran has a very strong production potential to provide food security from sheep and goat meat, though this opportunity has not been fully achieved. High neonatal loss especially in extensive rearing systems is directly associated with this defect since the production level is defined as the number of lambs per ewes in a flock (3).

The highest mortality rate is usually close to the time of birth, especially within the first 3 days (4) and it has been estimated to occur in the range of 8% to 30% worldwide (5). In a retrospective study during 2015-2016 in Ethiopia, annual birth-to-weaning losses were reported alarmingly high which ranged from 14.9–33.5% in lambs, and 17.6–24% in kids (6). As a result, the factors affecting lambs health during this period should be determined to apply appropriate control programs based on the common causes of death.

Different infectious and non-infectious factors have been attributed to lambs and kids losses worldwide. However, depending on the region and environmental risk factors, predominant causes, and their prevalence may be different (1, 7). In addition, the interaction of factors such as management practices, the environment, infectious agents, and the animal condition determines neonatal survival in a region (8).

Feeding insufficient colostrum, starvation, cold stress, dystocia, and mismothering are some of the non-infectious factors which significantly increase lamb mortality (9). Diarrhea and pneumonia are the major consequences of defects in both animal husbandry and infectious diseases which lead to pre-weaning mortality (6, 8). Diarrhea is the most devastating disease in newborn animals under 21 days of age which is accompanied by enteritis, abdominal pain, and an increase in fecal fluidity (10). It is reported that in Mexico one of the two main causes of mortality in goat kids is diarrhea (11). In addition, growth retardation in lambs and kids that have recovered from the disease and the costs of treatment are the other economic losses due to diarrhea (12). Major microbial agents that have been isolated in neonatal diarrhea and pneumonia include *E. coli, Salmonella* spp., *Pasteurella* spp., *Klebsiella* spp., *Staphylococcus, Streptococcus*, and *Pseudomonas* spp. (13).

Reduction in lambs losses can be accomplished through the detection of major causes and the establishment of specific preventive measures based on the laboratory findings. In addition to pathogens identification, the influence of noninfectious factors and other conditions such as environment, gender, age, and breed on neonatal mortality should be further assessed. Nonindigenous breeds such as Lacaune are usually kept in intensive rearing systems, and in some cases, they have different requirements than native breeds which are more adapted to the local conditions. Since there is a lack of information about the newly entered breeds in Iran, the aim of this study was to evaluate the major causes of lambs mortality in the Lacaune breed that have been imported in large numbers to Iran.

## **Materials and Methods**

## Study Setting and Sample Collection

In an intensive sheep-rearing farm in a 3-month period, the causes of death were evaluated in 114 Lacaune lambs from birth to the age of weaning. The clinical signs of infection were visible in 52 lambs before death. The history, age and sex of each lamb were recorded. The animals were necropsied and heart and lung samples were aseptically collected. Then the specimens were packaged with ice packs and transmitted to the laboratory.

#### Bacterial Isolation and Identification

The samples were dissected using a sterile loop initially streaked on general microbiological media including blood agar and MacConkey agar. Then, the cultured plates were incubated at 37 °C for 48 hours. The colonies which were grown in the media were morphologically examined and stained by the gram staining method. According to the bacteria observed under a light microscope, the shape of colonies and their growth on MacConkey agar were assessed by differential media and biochemical tests. According to suspected colonies on the general media, the bacteria were further assessed by the biochemical media including Nitrate, Eosin Methylene Blue, TSI, Urea, and SIM. Moreover, the colonies were examined using IMViC, catalase, oxidase, and coagulase tests.

## Statistical analysis

The effects of age, sex and infectious agents on lamb mortality were analyzed using spss version 21. Chi-square and one-way ANOVA statistical tests were performed and  $p\leq0.05$  was considered significant.

## Result

A total of 52 out of 114 lambs (45.6%) were infected with bacteria including *Escherichia coli* (30.8%), *Proteus mirabilis* (19.2%), *Pasteurella multocida* (1.5%), *Corynebacterium pseudotuberculosis* (7.7%), *Mannheimia haemolytica* (3.9%), *Staphylococcus epidermidis* (1.9%), and finally while the signs of diarrhea and septicemia were visible before death, in 25% of 52 samples no bacteria were isolated.

Diarrhea was the sign of 31 animals before the death of which *E. coli*, *Proteus mirabilis*, *Pasteurella*, and *Corynebacterium* were isolated from 14, 8, 2 and 2 lambs respectively. Septicemia was the sign of 11 animals before death and *E. coli*, *Pasteurella*, *Mannheimia*, *Corynebacterium*, and

*Proteus mirabilis* were isolated from 2, 1, 1, 1, and 1 lambs respectively. *Pneumonia* was visible in 10 animals before the death of which *Pasteurella*, *Mannheimia*, *Corynebacterium*, *Staphylococcus epidermidis*, and *Proteus mirabilis* were isolated from 3, 1, 1, 1, and 1 lambs respectively. The rest of the lambs died due to non-infectious causes and trauma was more predominant (Table 1).

According to Table 1, infection agents were significantly the most causes of lamb mortality ( $p \le 0.05$ ), and *Escherichia coli* was isolated significantly more than other bacterial agents ( $p \le 0.05$ ) (Table 2 and Figure 1).

The average age of the dead lambs was 21.5 days. Among 52 infected lambs, 22 (42.3%) were female and 30 (57.7%) were male. The average age of death in female lambs was 22.8 days and for males was 20.5 days. There wasn't a significant difference in the sex of the dead lambs.

According to Figure 2, the highest number of deaths is related to the age range of fewer than 15 days with 10 animals (46.1%) which was significant ( $p \le 0.05$ ). Moreover, the most mortality rate was seen in the first month of life (78.8%). In the age of fewer than 15 days, the most isolated agent was *Escherichia coli*, which was detected in 10 lambs was equal to 19.2% of all the infectious deaths and 41.7% of deaths in the first two weeks of lambs' life

#### Discussion

In the current study, bacterial and non-infectious causes of 114 new-born deaths from birth to the age of weaning were assessed which showed the highest frequency of bacterial infection (Table 2). This result was in agreement with the study conducted in nomadic flocks of Isfahan province in 2014, in which 45.03% of lamb mortality was related to infectious diseases (14). Another eight-year investigation (2002-2010) in India showed that infectious agents were significantly the most important causes of lamb death (3). Holmøy and Wage in 2017 and Shiels et al., in 2022 also found

infectious pathogens among the most important causes of neonatal mortality (36% and 16% respectively) (5, 15).

It is evident that in intensive rearing systems like the studied flock in which lambing takes place indoors, the main lamb losses occur following infectious diseases and dystocia is the second cause of production inefficiency (5). Overcrowded closed pens, especially during lambing time, the lead transmission of pathogens easier. In such flocks, diarrhea and pneumonia jeopardize animals' health in situations where proper ventilation is not established (16).

In intensive flocks as the lambing process and individual lamb health are usually monitored closely, non-infection causes of neonatal mortality such as starvation are detected earlier and appropriate health care is taken to reduce the risk of death (17). While for ewes lambing outdoors, the major causes of lamb mortality are hypothermia, starvation, and predation (18). In the current study, the lambs were separated from the ewes immediately after birth and received a milk replacer, so there were very few cases of starvation and subsequent hypoglycemia and hypothermia (7 cases). According to the study of Refshauge et al. in 2016, half of neonatal mortality in traditional rearing practices was related to mismothering and starvation (19). In another retrospective study in 2021, among lambs belonging to traditional smallholders, malnutrition was the most common cause of lamb mortality (20). Colostrum consumption is crucial immediately after birth. Adequate consumption of colostrum increases the chance of survival before weaning (21).

Intensive flocks use warm boxes which prevent the death of hypothermic lambs. While in extensively grazed flocks and nomadic rearing systems, newborn animals are exposed to wind and cold weather during lambing season which increases the risk of high mortality. Al-khaza et al. in 2019 indicated harsh climatic conditions in different parts of Jordan, adversely affected the performance and health of the newborn animals (16). Other non-infectious causes of lamb loss such as umbilical diseases, trauma, and congenital abnormalities have less morbidity and mortality in small ruminant flocks (20). We also found no significant differences in the incidence of the assessed non-infectious factors (Table 1).

The present study indicated the importance of the control of infections specifically in intensive rearing systems. Gastrointestinal diseases are the most common infection cause of death (22). Based on the current results, diarrhea was more prevalent (59.6%) followed by septicemia (21.2%) and pneumonia (19.2%). Aklilu et al. in 2013 in Ethiopia, found diarrhea responsible for 60% of neonatal problems (23). In the study by Hadgu et al. in 2021, 24% of the causes of lamb's mortality were related to diarrhea and 21.3% were due to respiratory problems (20). The role of bacteria in the development of intestinal problems in lambs is well known. Among the bacteria, Escherichia coli, Salmonella, and Clostridium are the dominant pathogens (22). We isolated Escherichia coli and Proteus mirabilis from 30.8% and 19.2% of lambs respectively which suffered diarrhea before death. Numerous infection agents are responsible for newborn diarrhea and E. coli is the commonest one (24). In a similar study that evaluated the frequency of bacterial infection in diarrheic lambs, E. coli, Klebsiella, Enterobacter, Citrobacter, Proteus, Serratia and Morganella morganii were respectively isolated the as members of Enterobacteriaceae (25).

*Escherichia coli* is the most harmful organism in the first two weeks of birth. Since the immune system of lambs has not been developed (26). In our study, the highest number of deaths occurred at the age of fewer than 15 days (46.1%) (Figure 2) and the difference was significant ( $p \le 0.05$ ). Moreover, during this period *E. coli* was the most common agent and was isolated from 10 lambs. In agreement with our results, Snigdha et al. in 2022 reported that the highest incidence of diarrhea caused by *Escherichia coli* (35%) was between 0 and 14 days old lambs (26). Another study conducted in Ethiopia in 2013, found lambs less than 1 month significantly more susceptible to diarrhea and *E. coli* was isolated from 84% of the diarrheic animals (23).

We examined the causes of death in the Lacaune breed that have a considerable population among imported sheep to Iran. It is evident that highyielding non-indigenous sheep breeds are more sensitive to E. coli infection. On the other hand, most of those breeds are kept in intensive rearing systems, so unclean lamb houses, lack of proper environmental and milk equipment hygiene have more effects on the health of such animals (23). Escherichia coli was also isolated from 18.2% of the septicemic lambs and septicemia was the second cause of neonatal mortality. Low-weight lambs and lambs born from weak ewes are considered at higher risk for septicemia (27). In intensive rearing systems, lambs with lower birth weight are removed so they usually are not calculated in the lambs' mortality rate. Since they are born weaker than other lambs and are more sensitive to diseases than others.

Among the non-infectious factors, we found that 17.5% of lambs' death occurred among lowerweight animals. Finally, pneumonia was the third cause of death in our study and Pasteurella was isolated as the most common pathogen from the specimens. Histopathological findings of a study in 2016 in the US, revealed pasteurellosis as the most predominant cause of pneumonia in younger lambs Mycoplasmosis typically and was more predominant in older lambs (28). Although like the present findings Babu et al., in 2019 isolated E.coli, Pasteurella and Mannheimia from the lungs belonged to lamb losses cases, in contrary, the frequency of the bacteria was different from the current results. In a way that E. coli was isolated from 46.51% of samples while Pasteurella multocida was the least frequent bacteria (9.3%) (29).

Respiratory diseases in lambs may depress the health statues of the animals for years (30). *Mannheimia haemolytica* is frequently isolated

from infected lungs of dead lambs (31), though in our study *Pasteurella* was more prevalent. Other bacteria like *Corynebacterium* are less frequently isolated in pneumonic lambs (32) and in our study, only 10% of the cases were due to the less common agents.

In the present study, respiratory signs were observed in older lambs compared to diarrheic animals and the average age of pneumonic lams was 38 days old. Similarly, in a study in 2019 in Spain, the first respiratory symptoms started when the average age of lambs was 32 days old (33). Accordingly, it is necessary to know the times of the lamb's life when they are more susceptible to each disease so that the breeder can prepare to deal with them at different periods.

We found no significant differences between female and male lamb mortality which was in accordance with the study of Swarnkar et al., in 2019 in India. The authors indicated the factor which affects the probability of lambs' death was age (27). In contrast, another study in 2019 in Jordan showed higher mortality in male lambs (16). Bangar et al. in 2016 in India, also found the risk of male lambs' death was higher than females (34). Factors such as higher susceptibility of males to diseases, higher probability of dystocia in ewes giving birth to male lambs, and poor health care and monitoring of males have all been considered as the causes of this difference between the two sexes (16).

The success of any breeding system depends upon the rate of survival of offspring. A three percentage reduction in lamb mortality is defined as 100.000 more living animals each year and it means a more efficient flock (15). In the current research, we emphasized infectious pathogens and *E.coli* had a high contribution to lamb losses in Lacaune breeds that were reared in intensive conditions. Several studies have shown the bacterium is an indicator of sheep house hygiene, so the lower the level of hygiene, the greater the disease and death caused by *E. coli* (10, 23, 25, 29).

## Table 1. The evaluated causes of lambs mortality.

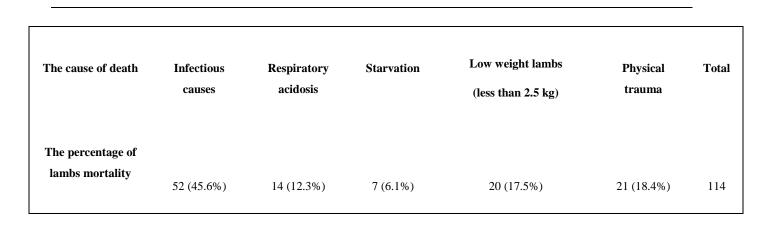


Table 2. Frequency of the isolated bacteria and the average age of the infected lambs.

Isolated bacteria	Frequency	Percentage	Cumulative frequency percentage	Average of the age (±1)
Escherichia coli	16	30.8	30.8	18
Proteus mirabilis	10	19.2	50	16
Pasteurella multocida	6	11.5	61.5	38
Corynebacterium pseudotuberculosis	4	7.7	69.2	30
Mannheimia haemolytica	2	3.9	73.1	20
Staphylococcus epidermidis	1	1.9	75	17
Negative	13	25	100	20
Total	52	100		21

Pathogens that are a threat to lamb health can be eliminated by the implication of appropriate health plans and efficient management (10). Due to the seasonal estrus of sheep and the density of intensive rearing systems, one of the management practices is to avoid synchronizing and intensive lambing until the working capacity and expertise of the flocks have their optimal situation. In the studied flock the sampling time was in the middle of births and the births were planned consecutively and densely. It caused the fatigue of the staff and decreased the accuracy in the process of milking, monitoring, and the lambs' health care.

## Conclusion

Expanding the lambing time throughout the year improves the level of hygiene and dramatically reduces the incidence of *E. coli* infection which is directly related to environmental contamination. Disinfection of the pen of pregnant ewes waiting for parturition, cleaning of teats and equipment. feeding sufficient colostrum, and timely change of bedding are among the measures that reduce E.coli infection. Finally, the authors recommend evaluating other factors that have higher risks for neonatal survival since safe passage through the neonatal period needs an appropriate management system.

## Acknowledgements

The authors acknowledge the members of the microbiology laboratory of the faculty of veterinary medicine, University of Tehran.

## **Funding Information**

This research received specific grant from faculty of veterinary medicine, University of Tehran.

## Ethics approval and consent to participate

This study did not require an ethics license.

## **Conflict of interest**

The authors declare that they have no conflict of interest.

## References

- Gama LT, Dickerson GE, Young LD, et al. Effects of breed, heterosis, age of dam, litter size, and birth weight on lamb mortality. *J Anim Sci* 1991; 69:2727-43.
- 2. Notter DR, Mousel MR, Leeds TD, et al. Effects of rearing triplet lambs on ewe productivity, lamb survival and performance, and future ewe performance. *J Anim Sci* 2018; **96**:4944-58.
- Makhdoomi DM, Tufani NA. Infectious and non-infectious diseases as cause of mortality in sheep. 3rd International Scientific Conference on Small Ruminant Development, Hurghada, Egypt, 12-15 April, 2010.
- 4. Alexander G, Peterson JE, Watson RH. Neontal mortality in lambs: intensive observations during lambing in a Corriedale flock with a history of high lamb mortality. *Aust Vet J* 1959; **35**:433-441.
- 5. Holmoy IH, Waage S, Granquist EG. Early neonatal lamb mortality: postmortem findings. *Animal* 2017; **11**(2):295-305.
- Fentie T, Temesgen W, Melaku A, et al. Assessment of young stock mortality in major livestock production systems of Ethiopia. Feed the Future USAID 2016. University of Gondar, Ethiopia.
- Dwyer CM, Conington J, Corbiere F, et al. Invited review: improving neonatal survival in small ruminants: science into practice. *Animal* 2016; 10:449-59.
- Lema M, Kassa T, Tegegne A. Clinically manifested major health problems of crossbred dairy herds in urban and periurban production systems in the central highlands of Ethiopia. *Tropical Animal Health and Production* 2001; 33(2):85-93.
- Shiferaw Y, Yohannes A, Yilma Y, et al. Dairy husbandry and health management at Holleta. The 16th Conference of the Ethiopian

Veterinary Association. Addis Ababa, Ethiopia, 5-6 Jun, 2002.

- Constable PD, Hinchcliff KW, Don SH, Gruenberg W. 2016. Veterinary Medicine. A Textbook of Disease of Cattle, Sheep, Pigs, Goats and Horses. 11th ed. Elsevier, Missouri.
- Cuellar OJA, Tortora PJ, Trejo GA, et al., 2012. La producción caprina mexicana, particularidades y complejidades. 1st ed. UNAM, México: Aridana.
- Zhong T, Wang Y, Wang X, et al. Diarrhea in suckling lambs is associated with changes in gut microbiota, serum immunological and biochemical parameters in an intensive production system. *Front Microbiol* 2022; 13 (17):1-10.
- Sharif L, Obeidat J, Al-Ani F. Risk factors for lamb and kid mortality in sheep and goat farms in Jordan. *Bulgarian J Vet Med* 2005; 8(2):99-108.
- Aubi M, Tabatabaei SN, Zamani F. Mortality of lambs at birth to weaning in nomadic flocks of Isfahan province. The 1st international conference on new ideas in agriculture. Isfahan, Iran, 26-27 Jan, 2014.
- 15. Shiels D, Loughrey J, Dwyer CM, et al. A survey of farm management practices relating to the risk factors, prevalence, and causes of lamb mortality in Ireland. *Animals* 2022.; **12** (30):1-14.
- Al-Khaza leh J, Megersa B, Obeidat B. Constraints and risk factors contributing to young stock mortalities in small ruminants in Jordan. *Small Rumin Res* 2019; **19**:1-19.
- 17. Ahmed A, Egwu GO, Garba HS, et al. Studies on risk factors of mortality in lambs in Sokoto, Nigeria. *Niger Vet J* 2010; **31**(1):56-65.
- Everett-Hincks JM, Dodds KG. Management of maternal-offspring behavior to improve lamb survival in easy care sheep systems. *J Anim Sci* 2008; 86(14):59-70.
- 19. Refshauge G, Brien FD, Hinch GN, et al. Neonatal lamb mortality: factors associated with the death of Australian lambs. *Anim Prod Sci*

2016; 56: 726-35.

- 20. Hadgu A, Lemma A, Yilma T, et al. Major causes of calf and lamb mortality and morbidity and associated risk factors in mixed croplivestock production system in Jamma district, south wollo, Ethiopia. *Vet Med Int* 2021:1-14.
- Agenbag B, Swinbourne AM, Petrovski K, et al. Lambs need colostrum: A review. *Livest Sci* 2021; 251:104624.
- 22. Uzzau S, Leori GS, Petruzzi V, et al. *Salmonella enterica* serovar-host specificity does not correlate with the magnitude of intestinal invasion in sheep. *Infect Immun* 2001; **69**:3092– 99.
- 23. Aklilu M, Sisay T, Tefera G, et al. Identification and biotyping of *Escherichia coli* from diarrheic lambs in and around Debrebirhan town, Ethiopia. *J Infect Dis Ther* 2013; **1**(115):1-3.
- 24. Mokhbatly AA, Elsheikh N, Ghazy EW, et al. Prevalence of Shiga toxin- producing *Escherichia coli* and *Salmonella* and some associated hematologic and biochemical profile alterations in lambs. *Vet Res Forum* 2022; **13**(2):155-62.
- Bkheet AA, Fadly RS, Elhoffy HR. Studies on some bacterial and parasitic causes of lamb diarrhea in Bohaira province and the subsequent biochemical changes. *Assiut Vet Med J* 2010; 56(127):203-22.
- 26. Shrivastava S, Kumar Mishra K, Shrivastava N, et al. Prevalence of caprine diarrhea due to *Escherichia coli* in the Vindhya region (Rewa) of Madhya Pradesh. *J Pharm Innov* 2022; 22(3):1694-98.
- Swarnkar CP, Gowane GR, Prince LLL, et al. Risk factor analysis for neonatal lamb mortality in Malpura sheep. *Indian J Anim Sci* 2019; 89(6):640-4.
- Wood ME, Fox KA, Jennings-Gaines J, et al. How respiratory pathogens contribute to lamb mortality in a poorly performing Bighorn sheep (*Ovis Canadensis*) herd. J Widl Dis 2016; 53(1):126-30.
- 29. Babu R SD., Balasubramaniam GA, Murthy T

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GK, et al. Isolation and identification of bacterial species from small ruminants affected with Pneumonia and their antibiogram pattern. *J Entomol Zool Stud* 2019; **7**(1):621-3.

- 30. Cassirer EF, Plowright RK, Manlove KR, et al. Spatiotemporal dynamics of pneumonia in bighorn sheep. *J Anim Ecol* 2013; **82**:518-28.
- Daniel JA, Held JE, Brake DG, et al. Evaluation of the prevalence and onset of lung lesions and their impact on growth of lambs. *Am J Vet Res* 2006; 67:890-4.
- 32. Oruc E. The pathologic and bacteriologic comparison of pneumonia in lambs. *Turkish J Vet Anim Sci* 2006; **30**(6):593-9.
- 33. Navarro T, Ramos JJ, Figueras L, et al. Epidemiology of ovine respiratory complex in lambs. *Small Rumin Res* 2019; **179**:70-4.
- 34. Bangar YC, Pachpute ST, Nimase RG. The survival analysis of the potential risk factors affecting lamb mortality in deccani sheep. J Dairy Vet Anim Res 2016; 4:266-70.