



Investigation of Bovine Respiratory Disease in Dairy Calves in Bahri Locality, Sudan

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Authors' contributions

This work was carried out in collaboration between both authors. Author EAAA designed the study, performed the statistical analysis and wrote the protocol and wrote the first draft of the manuscript. Author EKGA managed the analyses of the study, managed the literature searches. Both authors read and approved the final manuscript.

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ABSTRACT

Bovine respiratory disease (BRD) is considered the major cause of economic losses in dairy and beef cattle production due to its high morbidity and mortality rates. Opportunistic bacteria are factors for the development of BRD. This study was aimed at investigation of BRD in dairy calves in Bahri locality, Sudan. The study was carried out during six months of the years 2019 and 2020. This simplified scoring system which used assessed six clinical signs. When present, a specific number of points were assigned for each sign. A total score of 5 or higher classified an individual as a BRD case. Among 450 dairy calves investigated for presence of BRD, 40 calves (8.9%) were positive for the scoring system. Out of the 40 dairy calves positive for BRD scoring system, 24 (60%) were male calves. Out of 40 dairy calves positive for BRD scoring system, 25 (62.5%) 1 month old, 5 (12.5%) were 1.5 month, 4 (10.0%) were 2 months, 2 (5.0%) were 2.5 months and 4 (10.0%) were 3 months. Out of 40 dairy calves positive for the scoring system in Bahri Locality, 40 (100.0%) appeared nasal discharges, 31 (77.5%) eye discharges, 33 (82.5%) coughing, 25

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(62.5%) difficult breathing, 13 (32.5%) fever and 29 (72.5%) Ear drop or head tilt. In this investigation a total of 43 bacterial isolates were obtained from 40 nasal swab samples collected from pneumonic calves in Bahri locality. The identified bacteria according to bacteriological examinations were: 8 *S. aureus* (18.6%), 3 *S. epidermidis* (6.9%), 2 *S. chromogenes* (4.7%), 5 *Str. Pneumoniae* (11.6%), 2 *Str. uberis* (4.7%), 3 *K. pneumoniae* (6.9%), 7 *E. coli* (16.3%), 6 *Ps. aerogenosa* (13.9%), 2 *B. subtilis* (4.7%), 2 *M. variens* (4.7%) and 3 *M. luteus* (6.9%). Staphylococci represented the predominant bacteria (30.2%) isolated from nasal swabs compared to other bacteria *E. coli* (16.3%), Streptococci (16.3%), *Ps. aerogenosa* (13.9%), Micrococci (11.6%) *K. pneumoniae* (6.9%) and *B. subtilis* (4.7%).

Keywords: BRD; scoring system; dairy; Bahri; bacteria; nasal swabs.

1. INTRODUCTION

Bovine respiratory disease (BRD) is a multifactorial disease in cattle caused by a variety of pathogens. It affects both the upper and lower respiratory tract and the lungs of cattle [1]. BRD is the leading cause of death in weaned dairy heifers and the second most common cause of in pre-weaned calves in cattle herds in the United States. It is a major source of economic loss for the cattle industry [2,3]. It is estimated that BRD is responsible for the loss of more than one million animals and approximately US \$700 million annually [4,5]. The infection is usually a sum of three codependent factors: stress, an underlying viral infection, and a new bacterial infection [6,7,8]. Infections of the upper respiratory tract such as rhinitis typically present with ocular and nasal discharge. Infections of the lower tract, in contrast, may be challenging to detect earlier in the disease course. In addition, both upper and lower tract infections may vary in severity. Despite the variability of presentation, the observation of clinical signs is the most common method used to identify cattle in need of treatment for BRD. However, the specific criteria used to detect BRD are subjective and vary widely among dairies and observers, leading to deleterious effects on animal welfare and unnecessary treatments with antimicrobial drugs [9]. The diagnosis of the disease is complex since there are multiple possible causes [10]. The disease manifests itself most often in the calves within four weeks of weaning, when calves are sorted and often sold to different farms. This gives it a common nickname, "shipping fever." It is not known whether the stress itself, co-mingling, or travel conditions are at most to blame, and while studies have identified general stressing factors like transport and cold weather conditions, there is still no conclusive evidence on more specific factors (e.g. distance, transport mode, temperature, or temperature volatility) [11,12]. In Sudan [13,14]

reported that pneumonia is the most important cause of calf mortality beside diarrhoea in dairy farms at Khartoum State.

This study was aimed at investigating Bovine respiratory disease in dairy calves in Bahri locality, Sudan.

2. METHODOLOGY

2.1 Area of the Study

The study was carried out in the Bahri locality between February 2019 and July 2020.

2.2 Bovine Respiratory Disease (BRD) Scoring System (University of California)

This simplified scoring system assesses six clinical signs. When present, a specific number of points are assigned for each sign. A total score of 5 or higher classifies an individual as a BRD case.

Cough = 2 points
 Eye discharge = 2 points
 Fever (> 39.2C°) = 2 points
 Abnormal respiration = 2 points
 Nasal discharge = 4 points
 Ear droop or head tilt = 5 points

One of the advantages of this system is that a rectal temperature is not needed for every calf; it is only needed if the total score for the visible signs is 4. A temperature could then tip the score over the cutoff of 5 [15] (Fig. 1).

2.3 Source of Samples

A total of 40 naso-pharyngeal swabs were collected from dairy calves positive for BRD scoring system and didn't receive any treatment.


2.4 Isolation, Identification and Characterization of Bacterial Isolates

All media (Oxoid media) were prepared and sterilized according to the manufacturer instructions. For the primary isolation of bacteria, a loop full of the enriched broth streaked onto blood agar, McConkey's agar, and nutrient agar using sterile wire loop. The cultures were incubated aerobically at 37°C for 18-24 hours. Cultures on semi-solid media were examined grossly for colonial morphology and haemolysis on blood agar. Whereas, broth media were checked for turbidity, change in colour, accumulation of gases in CHO media and for sediment formation. One half colony from each plate was used for performing gram staining. Purification was based on the characteristics of colonial morphology and smear. This was

obtained by sub culturing of a typical discrete colony on blood agar plate. Pure cultures were preserved on slants of blood agar and egg media at 4°C.

2.5 Biological and Biochemical Identification of the Bacteria

The purified isolates were identified as previously described by [16,17]. The identification includes: Gram's reaction, presence or absence of spores, shape of organism, motility, colonial characteristics on different media, aerobic and anaerobic growth, sugars fermentation ability and biochemical tests (staining of smear, catalase test, oxidase test, coagulase test, oxidation fermentation test, motility test, glucose breakdown test and fermentation of carbohydrates).















Calf Health Scoring Criteria			
0	1	2	3
Rectal temperature			
100-100.9	101-101.9	102-102.9	≥103
Cough			
None	Induce single cough	Induced repeated coughs or occasional spontaneous cough	Repeated spontaneous coughs
Nasal discharge			
Normal serous discharge	Small amount of unilateral cloudy discharge	Bilateral, cloudy or excessive mucus discharge	Copious bilateral mucopurulent discharge
			
Eye scores			
Normal	Small amount of ocular discharge	Moderate amount of bilateral discharge	Heavy ocular discharge
			
Ear scores			
Normal	Ear flick or head shake	Slight unilateral droop	Head tilt or bilateral droop
			

Fig. 1. Clinical Assessment and Scoring according to BRD scoring system

3. RESULTS

3.1 Prevalence of Bovine Respiratory Disease in Bahri Locality

Among 450 dairy calves investigated for presence of BRD, 40 calves (8.9%) were positive for the scoring system.

3.2 Sex of Dairy Calves Positive for the Scoring System

Out of 40 dairy calves positive for BRD scoring system, 24 (60%) were male calves (Fig. 2).

3.3 Ages of Calves Dairy Calves Positive for the Scoring System

Out of 40 dairy calves positive for BRD scoring system, 25 (62.5%) 1 month old, 5 (12.5%) were 1.5 month, 4 (10.0%) were 2 months, 2 (5.0%) were 2.5 months and 4 (10.0%) were 3 months (Fig. 3).

3.4 Different Clinical Signs among Dairy Calves Positive for the Scoring System

Out of 40 dairy calves positive for the scoring system in Bahri Locality, 40 (100.0%) appeared

nasal discharges, 31 (77.5%) eye discharges, 33 (82.5%) coughing, 25 (62.5%) difficult breathing, 13 (32.5%) fever and 29 (72.5%) Ear drop or head tilt (Fig. 4).

3.5 Bacteria Isolated from Pneumonic Calves' Nasal Swabs Collected from Bahri Locality

In this investigation a total of 43 bacterial isolates were obtained from 40 nasal swab samples collected from pneumonic calves in Bahri locality. The identified bacteria were: 8 *S. aureus* (18.6%), 3 *S. epidermidis* (6.9%), 2 *S. chromogenes* (4.7%), 5 *Str. pneumoniae* (11.6%), 2 *Str. uberis* (4.7%), 3 *K. pneumoniae* (6.9%), 7 *E. coli* (16.3%), 6 *Ps. aerogenosa* (13.9%), 2 *B. subtilis* (4.7%), 2 *M. varians* (4.7%) and 3 *M. luteus* (6.9%).

Gram positive Bacteria represented the higher percentage (62.8%) compared to gram negative bacteria which represented 37.2% of the total bacteria isolated from nasal swabs (Fig. 5).

Staphylococci represented the predominant bacteria (30.2%) isolated from nasal swabs compared to other bacteria *E. coli* (16.3%), Streptococci (16.3%), *Ps. aerogenosa* (13.9%), Micrococci (11.6%) *K. pneumoniae* (6.9%) and *B. subtilis* (4.7%) (Table 1).

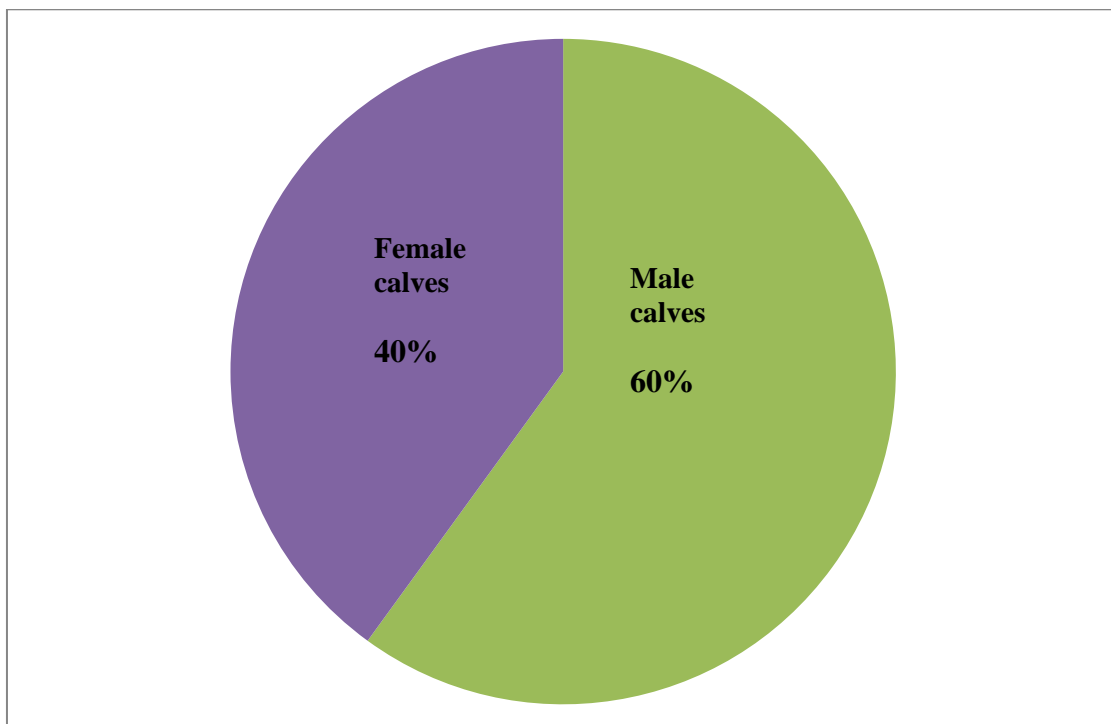


Fig. 2. Male and female calves positive for calf pneumonia in Bahri locality

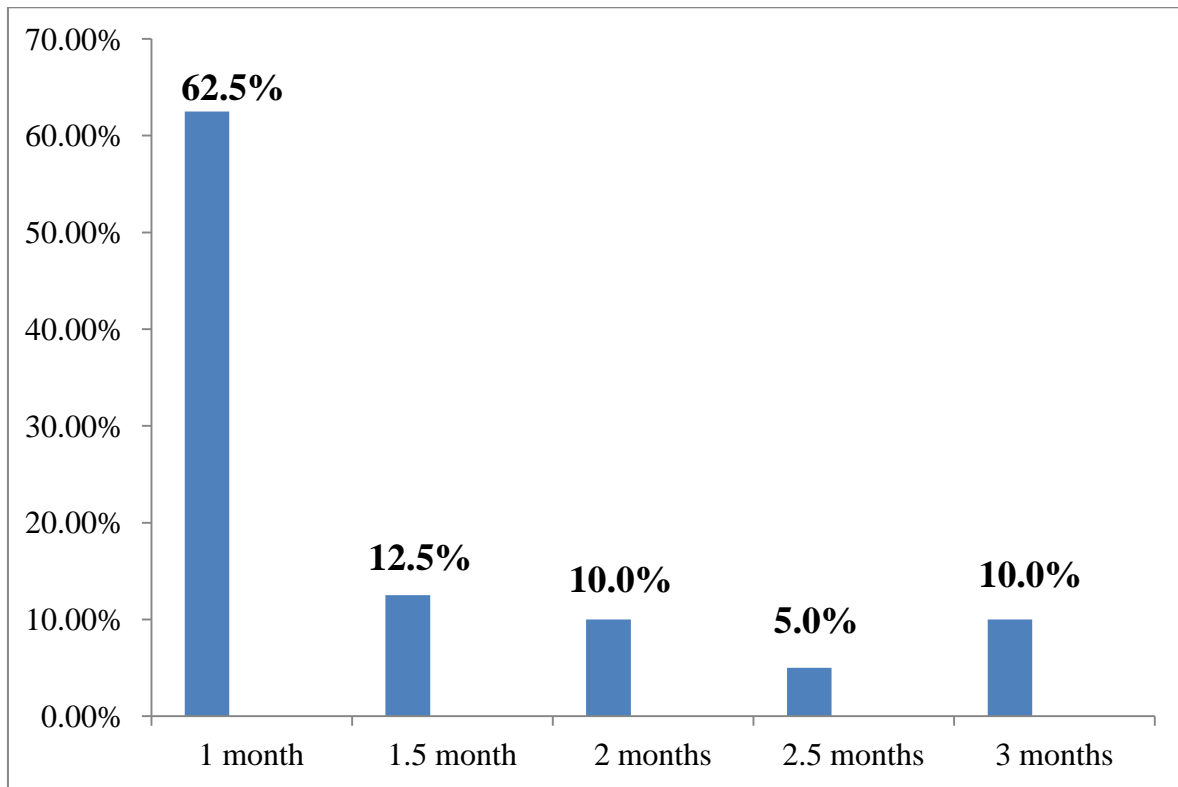


Fig. 3. Ages of calves affected with calf pneumonia

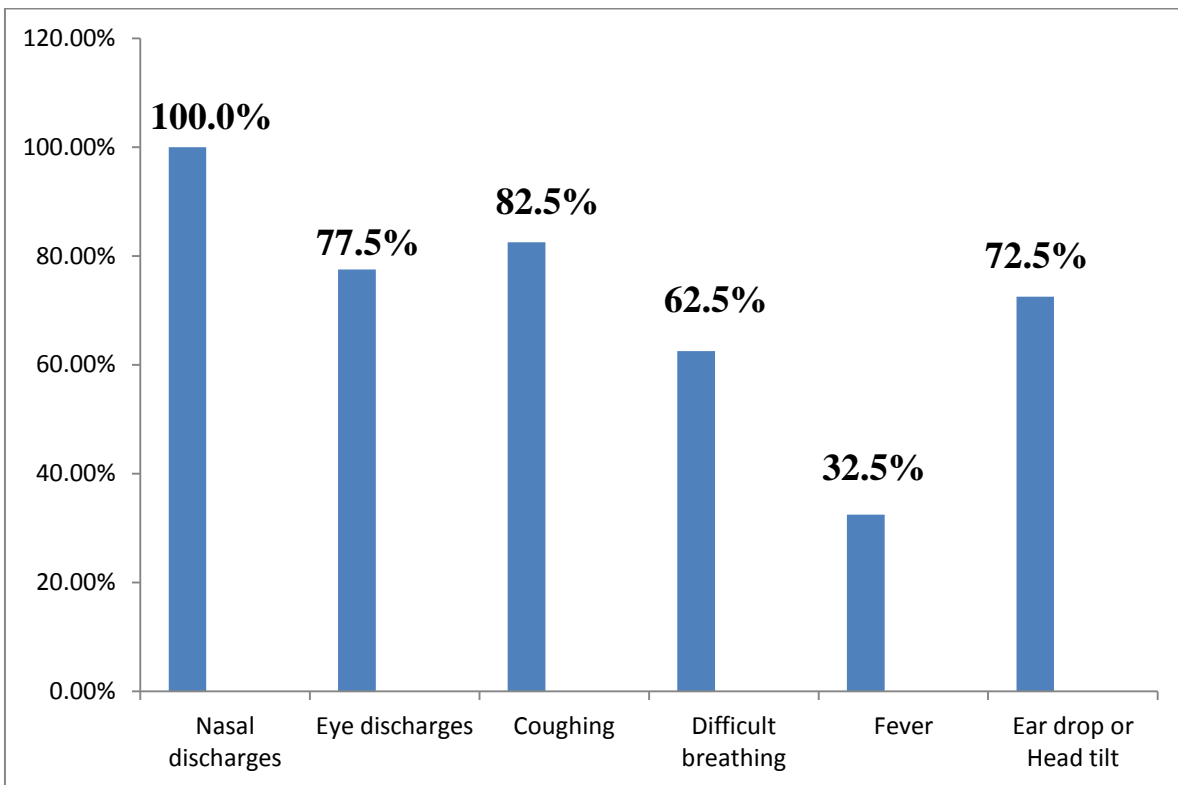


Fig. 4. Different clinical signs among pneumonic calves

Table 1. Bacteria isolated from pneumonic calves' nasal swab samples collected from Bahri Locality

Bacterial species	Number	Percentage
<i>S. aureus</i>	8	18.6%
<i>S. epidermidis</i>	3	6.9%
<i>S. chromogenes</i>	2	4.7%
<i>Str. Pneumoniae</i>	5	11.6%
<i>Str. uberis</i>	2	4.7%
<i>M. luteus</i>	3	6.9%
<i>M. variens</i>	2	4,7%
<i>B. subtilis</i>	2	4.7%
<i>E. coli</i>	7	16.3%
<i>Ps. aerogenosa</i>	6	13.9%
<i>K. pneumoniae</i>	3	6.9%
Total	43	100.0%

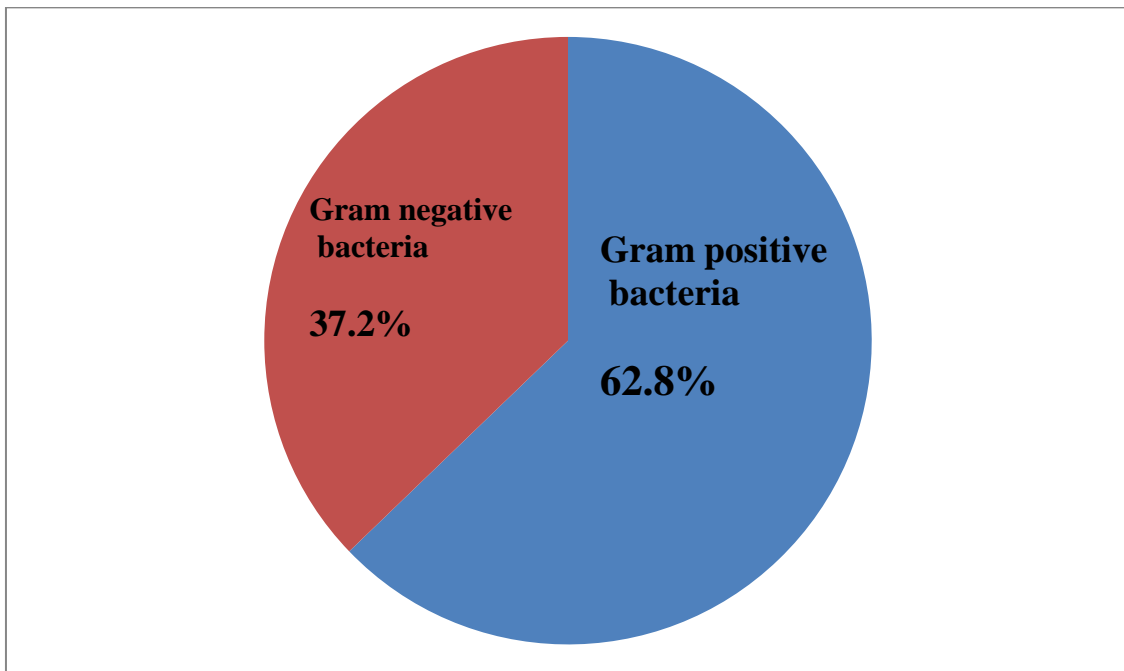


Fig. 5. Gram positive and gram negative bacteria isolated from pneumonic calves' nasal swabs collected from Bahri locality

4. DISCUSSION

Bovine respiratory disease (BRD) is the most common and costly disease affecting beef cattle in the world. It is a complex, bacterial infection that causes pneumonia in calves and can possibly be fatal. The infection is usually a sum of three codependent factors: stress, an underlying viral infection, and a new bacterial infection. The diagnosis of the disease is complex since there are multiple possible causes [10]. In this study the prevalence of BRD was

8.9%, which is lower than the prevalence of the disease (21.2%) in USA which reported by [18] and may be due to cold weather in USA than Sudan. [13] reported that the prevalence of 1.08% for pneumonia in Khartoum State, which is lower than the prevalence detected in this study. In this study the male calves affected with BRD represented 60% of the total affected calves. [19] also found lower mortality rate for females with the disease compared to males. In this study the affected calves with BRD at the age of 1 month represented 62.5% compared to

the other older ages and that because the first week of the calf live is more critical. [11] also reported that BRD affects mainly calves at the age of four weeks. In this study 40 (100.0%) of the affected calves with BRD appeared nasal discharges, 31 (77.5%) eye discharges, 33 (82.5%) coughing, 25 (62.5%) difficult breathing, 13 (32.5%) fever and 29 (72.5%) Ear drop or head tilt. [15] mentioned that symptoms of BRD include Coughing, Eye discharge, Fever, Abnormal respiration, Nasal discharge and Ear droop or head tilt. [20] reported that the main clinical signs of BRD are coughing, decreased appetite, and breathing difficulty. [15] mentioned that BRD can be divided into three main categories: upper respiratory tract infections, diphtheria and pneumonia. [8] reported that *Pasteurella multocida*, *Mannheimia haemolytica* and *Mycoplasma bovis*, are the major bacterial pathogens of BRD but there is other opportunistic bacteria. [21,8,22] reported that opportunistic bacteria are factors for the development of BRD. In this investigation a total of 43 bacterial isolates were obtained from 40 nasal swab samples collected from pneumonic calves in Bahri locality. The identified bacteria were: 8 *S. aureus* (18.6%), 3 *S. epidermidis* (6.9%), 2 *S. chromogenes* (4.7%), 5 *Str. Pneumoniae* (11.6%), 2 *Str. uberis* (4.7%), 3 *K. pneumoniae* (6.9%), 7 *E. coli* (16.3%), 6 *Ps. aerogenosa* (13.9%), 2 *B. subtilis* (4.7%), 2 *M. varians* (4.7%) and 3 *M. luteus* (6.9%). [23] also isolated *Staphylococcus* spp. and *Pasteurella haemolytica* from cases of calf pneumonia. [24] [25,26,21,15] isolated *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Streptococcus pneumoniae*, *Proteus vulgaris*, *Pasteurella multocida*, *Escherichia coli*, *Corynebacterium* spp., *Salmonella* spp, and *Enterobacter* spp. from cases of calf pneumonia in Nigeria. [27] reported that facultative anaerobes; *Mannheimia (Pasteurella) haemolytica*, *Pasteurella multocida*, *Haemophilus somnus*, *Arcanobacter pyogenes*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Actinobacillus pleuropneumoniae*, *Streptococcus* spp., *Staphylococcus* spp., *Moraxella* spp, *salmonella* spp. are the most common bacteria involved in pneumonia. [28] the majority of pathogens of the respiratory tract of cattle belong to the *Streptococcus*. [29] isolated *Streptococcus* species from cattle in Khartoum State suffered from pneumonia. [30] reported that *Klebsiella* spp. can be found associated with some diseases as secondary invaders but may also act as primary aetiological agents of diseases. [30] reported that *Pseudomonas aeruginosa* isolated frequently

from wound infections in a number of domestic animals.

5. CONCLUSION AND RECOMMENDATIONS

According to the results of scoring system the prevalence of BRD was 8.9% in Bahri locality. Application of scoring system for detection of BRD in beef and dairy calves in Sudan is very necessary. Further studies should include a survey of more animals in different farms and an extensive study of the significance of bacteria in calf pneumonia.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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