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Research Article

Caprine Arthritis is a Hidden Sickness Requiring Early Detection and Management

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ABSTRACT

Caprine arthritis may lead to impair performance and reduce productivity. Physical and chemical examination of synovial fluid (SF), and isolation of microbes from the SF were carried out in this study. Clinically 5% goats showed signs of arthritis with enlarged joints, pain response, farmer, warmer and hairless skin over the joints. Volume of SF of inflamed joints of the arthritic goats were significantly (0.42±0.123 ml, p> 0.012) higher than the healthy animal (0.143±0.013 ml). The color of the SF of inflamed joints became brownish to reddish and cloudy in nature. Mucin clot test of SF revealed absent of reaction in 05% cases, whereas, strong ring was formed with healthy (95%) SF. The cellular infiltrations and densities of leukocytes in arthritic goats ranged from 2,200 to 75,400 cells/ml in contrast to 21 to 34 cells/ml in apparently healthy SF. Neutrophilia (55% to 80%) was seen in three arthritic SF and monocytosis (75% to 80%) in two cases. Two joints were infected with *Staphylococci sp*, one with *E. coli* and one with *Aspergillous fungus*. The cause of a arthritis could not identify in this study. This study provide evidence that 05% of goats were affected with arthritis. The cytology of synovial fluid, biochemical tests and cultural isolation techniques adapted in this study can be used to detect specific cause of arthritis whether at sub-clinical or clinical stage. Early detection of specific cause of arthritis in goat may provide accurate therapeutic option, alleviate pain and enable higher productivity of milk, meat or offspring.

Introduction

Goats on pasture are susceptible to arthritis and the technology to detect the arthritis is yet to standardize in Bangladesh at field condition. The key constraint for successful goat production in Bangladesh is the infection by various microbes. Among systemic and local infection, arthritis is frequently noted as an imperative illness. Arthritis is the inflammation of joint that include synovial membrane, synovial fluids (SF) and fluid contents. Cells infiltrated in synovial fluid are mostly blood leukocytes. Inflammatory arthritis induces pain, deformity and disability of goats that hampers the growth and production performance. Arthritis may extend to bone and may damage bone and cartilage (Ostrowska et al., 2018). Arthritis is reported to increase synovial fluid and thickening of joint capsule. Moreover, numerous inflammatory cytokines and chemokines are involved in the inflammatory arthritis (Waseem et al., 2016). Joint diseases are caused by bacteria, virus, fungus and/or mycoplasmas, in which leukocytic infiltration from the blood is an important indicator. In each of the inflamed joints, the volume of synovial fluid is increased, leading to immobilization of the joint (de Grauw et al., 2009) There is viral arthritis popularly known as caprine arthritis encephalitis (CAE), multi systemic disease and distributed globally.

Caprine arthritis is a neglected illness of rural goat at present time. The major evaluation component of arthritis is to detect microbes and analyse synovial fluid. Synovial fluid usually showed a predominance of polymorphonuclear cells. However, high cellularity may sometimes be associated with a predominance of

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lymphocytes, monocytes, and eosinophils. Lymphocytes, a subset of leukocytes, are found infiltrating in the SF in viral arthritic joints of small ruminants (Turchetti et al., 2013; Wilkerson et al., 1995). In this situation, such a simple evaluation (differential cell count analysis) is very helpful in making a presumptive diagnosis of arthritis (Dougados, 1996). Idiopathic polyarthritis (IPA) is common in dogs (Bennett, 2010) and is diagnosed mainly by detecting increased number of leukocytes in the SF but such lesions is not documented in small ruminants. The total protein concentration of SF contributing as causal agents for IPA dogs. Total protein concentration in SF may be a diagnostic marker of canine IPA (Murakami et al., 2016).

Presence of neutrophil, lymphocyte, monocytes in the SF of the joints indicates infectious/septic arthritis. Detection of cellularity and absence of crystals in the SF helps in the diagnosis of septic arthritis (Ferreyra et al., 2017). High volume of synovial fluid with increased alkaline phosphatase, transaminase, total leucocytes, and protein with significant reduction of sugar found in the infected joints of goat (Nayak and Bhowmik, 1989). However, detection of specific cause of arthritis requires isolation and identification of microbes including bacteria, viruses, fungus and chlamydia. This study was attempted to standardize protocols to detect bacterial and fungal causes of caprine arthritis and help farmers to apply this protocols to detect and manage arthritis at their door steps.

Materials and Methods

In order to identify causes of caprine arthritis, knee and elbow joints of 100 goats were clinically examined. Synovial fluid from the arthritic and non-arthritic goats were collected from the patients brought to the Veterinary Teaching Hospital, Bangladesh Agricultural University (BAU), Goat Farm, BAU and Department of Surgery and Obstetrics, BAU. The samples were collected during the period from January to June 2020. The joints (Knee and elbow) of the goats showed signs of inflammation and lameness. Hairs over the skin of suspected joints were clipped, shaved and disinfected by painting with Povidone Iodine. Using the **s**terile tuberculin syringe single aspiration from the joint was collected, its volume was measured and dispensed in eppendorf tube for further investigation.

The physical characteristics (volume, viscosity, color and odor) of the synovial fluid (SF) were examined immediately after the collection of fluid. The volume of the SF was measured by observing the volume marking of tuberculin syringe. Aspirated synovial fluid of about 0.1 to 0.2 ml/ knee and elbow joint was considered as healthy. The viscosity of SF was considered as a state of normal fluid. Normal synovial fluid is clear, colourless or straw in nature. Presence of cloudy or coloured (yellow, pink, red) fluid also indicates a state of inflammation following collection in a clear vial. Normal synovial fluid is odourless and the existence of bad odour was considered inflamed joint. Mucin clot test was performed (Cohen et al., 1975) by mixing one-part synovial fluid (synovial hyaluronate) with four parts of glacial acetic acid with the formation of mucin clot. Formation of whitish band in between two solutions was an indication of non-arthritic joint inflammation.

Synovial fluid was subjected to microbiological examination to determine the presence and characteristics of microorganisms in caprine arthritis. For this purpose, synovial fluid was enriched overnight at 37ºC into nutrient broth followed by streaking and isolation of microorganisms onto different cultural media including nutrient agar, eosin methylene blue (EMB) agar and sabouraud dextrose agar (SDA) (Guinea et al., 2005; Merchant et al., 2005). The colony characteristics were recorded and morphology was revealed by Giemsa and Gram's staining techniques (Luna, 1968; Wilson, 1992).

The characteristics of leukocytes and their total number was determined by using Giemsa's staining (Wilson, 1992). About 10µl of synovial fluid was taken onto the middle of the glass slide and smeared with the edge of other clean edge slide. The slides were air dried, fixed for 30mins in ice cold methanol, air dried and stained with Giemsa's stain for 45mins. The stained onto the slides were gradually removed by placing the slide under running tape water. The slides were air dried and examined at 100X objectives. About 100 cells per samples were counted and differential cells present in the SF were analyzed. The total leukocytes count in the SF was carried out using a hemocytometer. The fresh synovial fluid was taken into a white blood cell (WBC) counted pipettes and filled in up to 0.5 marks. The WBC pipette was filled in up to 11 marks with 1% HCl, and shaken for 3mins. A drop of mixture from the WBC counting tube was discarded and a small drop of mixture was placed on one side of the counting chamber and allow the fluid to pass through the counting chamber and cover slip. All the four square chambers of WBC from the four corners were counted and were recorded. Total number of cells counted were multiplied by 50 and the resulted number of cells represented as thousands of cells per cumm of SF. Comparisons of cellular infiltrates and synovial microbes between infected and non-infected groups of goats were performed using a single classification ANOVA and the data obtained were analyzed by using SPSS statistical package 24.

Results

Synovial fluid was investigated to identify causes of abnormalities. This study investigated elbow and knee joints of randomly selected 100 goats and found 5 goats were lame (Table 1). Joints of three goats showed acute phase of illness whereas, two were chronically inflamed. Out of total goats investigated, 43 were male and 57 were female age ranging from 1.0 to 3.5 years. The cases of arthritis was detected in three female and two male goats. The affected goats showed inflammatory swelling of elbow (n=04) and knee (n=01) joints.

Physical properties of joints and synovial fluid

Warm, swelling, tender and pain response on pressure were the common clinical manifestations of inflamed joints. An affected joint showed massive swelling of knee. The chronically inflamed joints also revealed swelling, and hairless skin over the joint with lameness. The synovial fluid collected and examined were mostly clear in non-arthritic joints but cloudy, voluminous, thick and reddish in arthritic joints (Table 1). In three cases, the fluids appeared thicker, reddish and foul smelling.

Table 1. Physical characteristics of arthritic joints and SF of Black Bengal goats

Goats number	Description of Goats with its joints						
	Age (year)	Sex	Signs/ lesions	Legs/ joints affected	Volume of SF collected	Results of Mucin clot test	Nature of lesion
1	3.0	Female	Painful swelling and lameness	Left elbow joint	0.40ml	-Ve	Acute
2	2.5	Male	Swelling and lameness	Left elbow joint	0.30ml	-ve	Acute
3	2.0	Female	Hairless and dried skin over the joint, and lameness	Right elbow joint	0.60ml	-Ve	Chronic
4	2.5	Female	Painful swelling, and lameness	Left knee joint	0.30ml	-Ve	Acute
5	1.0	Male	Massive swelling of joint, rough and hairless skin and lameness	Left elbow joint	0.50ml	-Ve	Chronic
6 (Healthy control)	2.0	Male	Healthy joints (lack of swelling and pain)	None	0.13ml	+ve	Normal

Chemical examination of synovial fluid

Synovial fluid (SF) was collected ranging from 0.07 to 0.6 ml (Table 2). The SF was subjected to mucin clot test by using glacial acetic acid. A healthy SF lead to a congealing of the hyaluronic acid, forming a white ring popularly termed as mucin clot (Figure 1). In inflammatory joints, a mucin clot is not formed (the hyaluronic acid is degraded) or may poorly developed in a mildly inflamed case, a differential point to diagnose arthritic joint from non-arthritic joint. Out of 05 suspected arthritic fluid examined, 04 fluid samples did not form clot, a SF formed very thin clot indicating early inflammatory nature of the fluid.

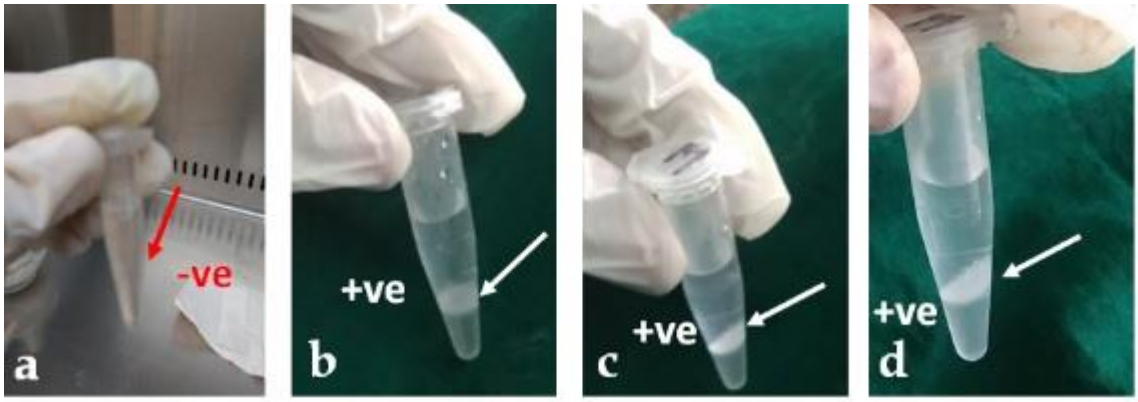

Figure 1. Mucin clot test of synovial fluid. In non-arthritic cases (b. c and d) sharp whitish ring was developed between the junction of SF and glacial acetic acid in 95 cases. The SF of arthritic joint did not form whitish ring (a) in five cases.

Table 2. Examination of physiochemical characteristics of synovial fluid of selected five healthy and five infected goats

Goat No.	Volume (ml) of SF collected	Color	Odor	Viscosity	Results of mucin Clot Test
1	0.2	Clear	-	-	-
2	0.16	Clear	-	-	-
3	0.05	Clear	-	-	-
4	0.2	Clear	-	-	-
5	0.15	Clear	-	-	-
6	0.4	Reddish	Foul smelling	Less viscous	-Ve
7	0.30	Cloudy	Foul smelling	Viscous	-Ve
8	0.6	Reddish	Foul smelling	Less viscous	-Ve
9	0.30	Cloudy	Foul smelling	Less viscous	-Ve
10	0.5	Reddish	Foul smelling	Less viscous	-Ve

Randomly selected goats No. 1, 2, 3, 4 and 5 were apparently healthy and goat No. 6, 7, 8, 9 and 10 were arthritic.

Cellular Infiltrates in SF

Synovial fluid collected from the apparently healthy and susceptible goats were smeared onto clean slides, stained with Gram's and Giemsa's stain and observed under microscope. The cells investigated were mostly leukocytes. Differential leukocyte count of the synovial fluid from arthritic and non-arthritic goats showed significant variation. There was a marked increase in the neutrophil count in three cases and monocyte count in two affected joints. In a case about 80% cells infiltrated were neutrophils and total leukocyte count was 75,400/ml. In an infected fluid lowest concentration of cells counted was 2,200 leukocyte/ml and the cells predominated were monocytes (78%). Total leukocytes count in inflamed joints was, therefore, ranged from 2200 to 75400 cells/ml. In uninfected cases, leukocytes counted in SF was ranged below 200/ml and cells dominated was monocytes (47 to 65%) and lymphocytes (33 to 50%) with 1-7 epithelial cells/ml.

Isolation and Identification of Microbes

Results of mucin clot test and leukocyte count in synovial fluid yield an indication of infectivity but the specific cause of illness could not be identified. Hence, SF was used to grow in culture for identification of bacteria and fungus. Direct smear from the synovial fluid on the clean slide stained with Giemsa's and examined under microscope also give an indication of bacterial or fungal infectivity (Figure 2) but at lower extent. The SF, therefore, was directly used to grow in nutrient broth, nutrient agar, EMB agar and SDA plate for the detection of specific cause of arthritis. This study revealed the presence of *Staphylococcus* spp (n=02), *E. coli* (n=01) and *Aspergillus* sp (n=01) in the arthritic joints (Figure 3).

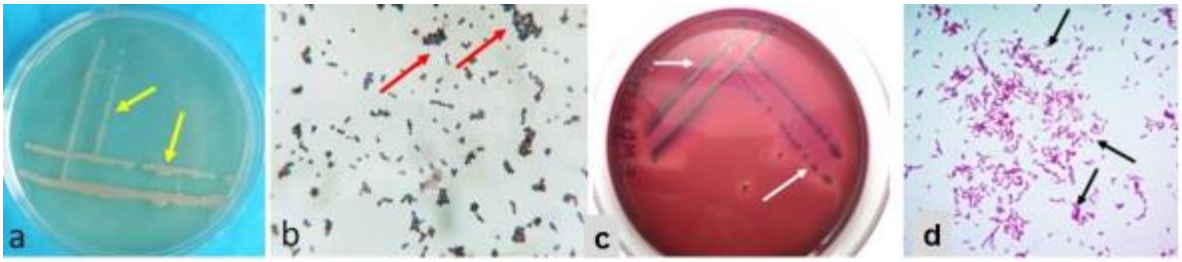

Figure 2. Isolation of bacteria onto nutrient (a) and EMB (c) agar medium. *Staphylococcus* spp. produced light cream colored colonies on nutrient agar and stained Gram +ve cocci arranged in clusters (a, b). On the other hand, *E. coli* produced bluish purple colonies with metallic Sheen onto EMB agar medium which revealed Gram -ve thin rod, arranged in single or short chain (c, d).

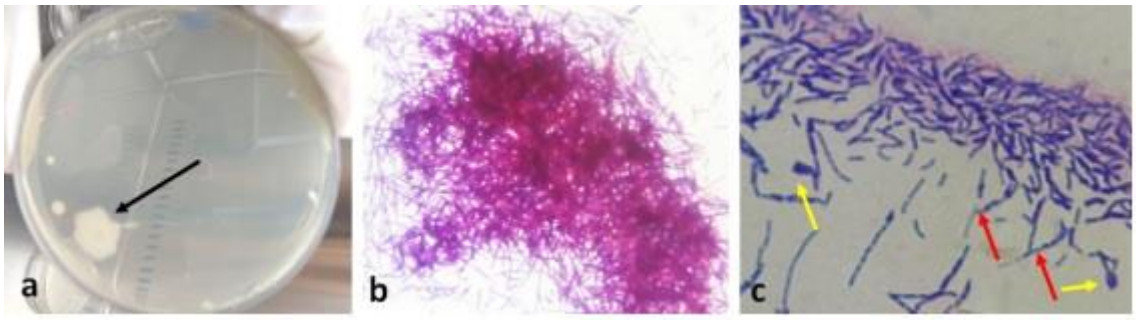

Figure 3. Isolation of fungus on SDA media from the joint of a 2.5 years old female goat (a, arrow). Culture of the SF revealed whitish colonies on SDA (a) which revealed pink color thin filamentous fungal hyphae on Gram's staining (b, 100x). owed. Giemsa staining of the fungal colony showed deep blue color branched and septate hyphae (c, red arrow, 300x) characteristics to the sporungium of *Aspergillus* spp. (c, yellow arrow).

Discussion

Arthritis a frequently noted illness of small ruminants is not a single entity, arthritis is an informal way of referring to joint illness or joint disease. The sophisticated laboratory techniques used to diagnose arthritis are X-ray imaging, serological tests, microbiological examination. However, detection of the causes of arthritis in animals is of major constraint. Arthritis in goats can be diagnosed by examining swollen joints and analyzing changes in the synovial fluid (Singh, 1991). Caprine arthritis encephalitis virus (CAEV) is the common case of arthritis and is diagnosed by examining joints and synovial fluids. Goats develop arthritis in joints with initiation of inflammation and thereby accumulation of fluid producing swollen joints (Maclachlan and Dubovi, 2010). Macroscopically, there is thickening of synovial membrane, infiltration of reactive cells from blood. The sub-synovia and connective tissue are also infiltrated with mononuclear cells, mainly lymphocytes, plasma cells and monocytes (Pérez et al., 2015), similar pattern of cellular infiltration was found in SF in this study.

Septic arthritis is an inflammatory condition that is associated with the invasion of microbial pathogens into the joint space. The predominant causes of arthritis in farm animal are septic arthritis. It has been reported that the origin of bacterial infections is from contamination, hematogenouse seeding, adjacent infection or direct trauma. The clinical presentation of bacterial arthritis includes lameness, in appetence, pain, joint swelling, recumbence, muscle atrophy and the reduction of productivity (Jesse et al., 2017; Moraes et al., 2010). Diagnosis of the arthritic condition is normally characterized by clinical signs, analysis of changed blood and synovial fluid and diagnostic imaging (Desrochers et al., 2014). In small ruminants, the distal joints are most commonly affected by direct trauma and abnormalities of joints and synovial fluids (Paape and Capuco, 1997). Therefore, this study was aimed to investigate arthritis in farm and free ranged goats and adapt or apply test protocol to identify specific cause of arthritis at minimum laboratory settings.

Physical Examination of Goats

In this study, 5% goats manifested mild to moderate arthritis and locomotor disorders and various degree of lameness. Three females and two males were affected and the joints involved were elbow (n=04) and knee (n=01) joints. The reasons of involving elbow joint is not clear but assuming that the front legs may be violently traumatized during randomize movement and enabling establishment of infection into the joints (Marcela et al., 2010). The bacteria may enter into the joints through broken, wet, or softened skin or by extension from the systemic or local lesions. However, open wound in any of the joint was not observed and the infection in joints may be due to extension of infection from systemic or local infection (McCarty et al., 2011). In the present study, both acute and chronic phases of infection were found, but reasons for differences were not clear. In addition, signs of alopecia and dryness over the chronic joints were evident but absent in acute joints. Aspiration of foul smell was found in 3 goats in this investigation is similar to the record of earlier researchers (Jesse et al., 2017 conducted in sheep. Septic arthritis is a medical emergency that requires prompt diagnosis and treatment in order to avoid morbidity, mortality as well as enhancing the general welfare of the animal. To enabling accurate diagnosis, synovial fluid from the inflamed joints was collected and examined.

Examination of Synovial Fluid

Examination of synovial fluid (SF) is required to rule out some serious conditions that may require management or medicinal intervention. Many joints contain small

amounts of SF, which helps the joint to glide and enable to move smoothly and help in the assessments of inflammation and joint abnormalities. Large quantity of fluid reduced thickness of joint is an indication of inflammation (Moraes et al., 2010). Synovial fluid is viscous in nature, meaning that it is thick and sticky and appearance of abnormal color of this fluid may indicate inflammation, hemorrhages or suppuration (Anderson et al., 2018; Anderson et al., 2019). Biomarker is not available for synovial fluid that indicates specificity for septic or non-septic arthritis. Therefore, synovial fluid based multiple analytical tools are required to adapt and to detect specific causes of arthritis.

The assessment of SF may be physical (color, volume, turbidity), chemical (concentration of total proteins and formation of mucin precipitate and cytological (nucleated cell count and swab analysis) is essential for joint health in dogs (Boon, 1997; Parry, 1999), humans (Sugiuchi et al., 1974; Brannan and Jerrard, 2006), and goats (Woodard et al., 1982; Maclachlan and Dubovi, 2010). The color of the synovial fluid samples in this study ranged from colorless, light yellow or pink color. Here, non-inflamed fluids (n=15) appeared colorless and viscous. The inflamed or infected synovial fluids were yellowish to reddish in color. Among the infected goats five manifest cloudy fluid and reddish in three cases. A study noted that (Moraes et al., 2010), infected caprine SF is colorless, dark yellow and light yellow. The color of the synovial fluid depends on the growth of infectious materials with leukocytes and blood. Normal synovial fluid is limpid and its coloration ranges from colorless to light yellow (Boon, 1997). Development of dark yellow to reddish is an indication of arthritis. The enhance volume of synovial fluids in the affected joints is also an indication of pathological condition and require addressing to support the healthy life.

In this investigation 0.07 ml to 0.20 ml volume of synovial fluids were collected from the knee or elbow joints of healthy goats (n=95). On the other hand, 0.30 ml to 0.60 ml of SF were aspirated from the arthritic joints. Higher volume of synovial fluid (0.94 ± 0.26 mL) was collected from the infected knee joints (Moraes et al., 2010). In joints, infections bacteria, viruses, mycoplasma or fungus were found. The viscosity is reduced in inflammatory states due to the reduction in hyaluronic acid polymers and increased production of neutrophils (Brannan and Jerrard, 2006; Francoz et al., 2005). This observation supports the evidence that increased synovial fluid is an indication of inflammatory conditions of joints. Reddish color synovial fluids recorded in joints indicated various degree of hemorrhages. Synovial fluid from a joint of arthritic goat appeared too much cloudy and less viscous. An elbow joint showed increase volume of synovial fluid (0.3 ml) and that is higher than control (0.07 to 0.20). The result of higher volume of SF in joints can be a good indicator of arthritic joint and reported by another group of researchers (Francoz et al., 2005).

Mucin clot test of synovial fluid

The quality of the mucin clot test is dependent upon the quality and concentration of hyaluronic acid present in the SF (Boon, 1997). Clot is rarely developed in a variety of inflammatory conditions of joints including septic arthritis, gouty arthritis, and rheumatoid arthritis and this test is, therefore, becoming a reliable indicator of differentiating arthritis from non-arthritic joints (Krebs et al., 2017; Moraes et al., 2010). In this study, knee and elbow joints of 100 goats were investigated, the SF was collected from the joints and analysed. Increased volume and viscosity of SF was seen in five cases but mucin clot test appeared negative indicating broken down of hyaluronic acid during arthritic processes and are arthritic joints. Faint band was seen in a case and this case may be due to an acute illness. All other SF (n=95) developed strong band during mucin clot test indicating a successful adaptation of mucin clot test and detection of arthritis. Mucin clot test is rapid and easily reproducible test to assess the quality of SF in arthritis (Moraes et al., 2010) and visual assessment of the strength of mucin clot band may be an early indication of detecting arthritis. However, analysis of leukocytes, blood cells and other cells in SF may indicate the extent and nature of arthritis.

Leukocyte count of SF

Normal synovial fluid consists of transudate of plasma from synovial blood vessels. Changes occur to the cell numbers and cell type in the fluid forming the basis of a diagnostic test (Hermansen and Freemont, 2017). An increase in turbidity suggests an increase in white blood cell count and a change in color indicates either a hemorrhage or iatrogenic contamination by blood at the time of collection. The fresh SF was collected and subjected to total leukocyte count and differential leukocyte count (Luna, 1968) and display the evidence of inflammation.

Lymphocytes is the predominant cell type (48.34±17.2%) found in the synovial fluid, whereas monocytes, macrophages and neutrophils constituted 36.52±3.5%, 12.75±5.9% and 2.28±1.18% in the synovial fluid, respectively (Ameri and Gharib, 2005). Lymphocytes reported to dominate over all other cell types present in the SF. In this study, mononuclear phagocytes (47 to 65%) were dominated in the fluid followed by lymphocytes (33 to 50%) and other cells. The total leukocytes count (TLC) was 18 to 34 cells/ ml of healthy SF and 2,200 to 75,400 cells/ ml in infected SF. Higher neutrophil count was observed in SF infected with bacteria. However, a goat infected with *E. coli*

showed the highest number of neutrophil count (75,400/ml) compared to 2,300 cells/ml in a fungal infected joint. Bacterial infection of the joint may trigger an acute inflammatory response leading to an influx of leukocytes. However, the recruitment of these inflammatory cells in SF depends on gradients of chemoattractants derived from the infectious agent or dying cells, host-derived leukotrienes, complement proteins and chemokines. Neutrophils are of major importance and play a dual role in the pathogenesis of septic arthritis. Neutrophil is indispensable member of cell in the first-line defense to kill invading pathogens in the early stage of bacterial arthritis. Since the elimination of inflammatory neutrophils from the site of inflammation is prerequisite for resolution of the acute inflammatory response, the prolonged stay of these leukocytes at the inflammatory site can lead to irreversible damage to the infected joints (Boff et al., 2018). In this study, the nutrophilic infiltration in the joint was reduced by withdrawing infected SF through tuberculin syringe but the fate of the arthritic joints was not evaluated due to shortage of resources and time limit for thesis preparation.

Joint infected with fungi showed higher percentage of monocyte count (75%) and joints infected with bacteria showed higher neutrophils (55 to 80%) count in the SF. From an infected joint the specific cause of illness is monocytes (78%) with TLC of 2,200/ml and this joint could have infected with other causes like mycoplasma, chlamydia or caprine arthritis encephalitis virus. It was reported that (Harkiss et al., 1991) sheep infected with visna virus (inflammatory synovitis) showed increase representation of lymphocytes followed by macrophage, and dendritic cell in SF. Goat infected with caprine arthritis encephalitis virus (CAEV) has dominancy of lymphocytes in joints. The activated lymphocytes in joints produced no dialyzable lymphokines mitogenic for macrophages. The activated macrophages in SF produce cytokines that destroy joint capsule leading to chronic arthritis (Jutila and Banks, 1988). In this study, lymphocyte dominating SF was not detected and joints infected were mostly due to fungus or bacteria. Specific causes of arthritis were therefore, identified by isolating bacteria and fungus from the cultures.

Isolation and identification of organism in culture

The most common microbes involved in septic arthritis (SA) are *Staphylococcus aureus, Streptococcus pneumoniae*, *E. coli, Proteus* sp.*, Salmonella* sp.*, Serratia marcescens, Erysipelothrix rhusiopathiae*, *Chlamydia sp.* and *Neisseria* sp. It has been estimated that 50% of total arthritis is caused by bacteria (Colavite and Sartori, 2014). Bacterial arthritis occurs most commonly in food animals (Desrochers and Francoz, 2014), especially in young animals. The bacteria enter into the joint mostly through trauma but application of specific tools for the accurate detection of specific cause is the major hurdle. A delay in the detection of arthritic may enable expansion of the lesions and damaging bones and joints.

The diagnosis of arthritis is usually made by clinical signs and by component of synovial fluid. Culture and sensitivity may yield false-negative results but inoculating the SF into blood agar medium may increases the chances of positive result. Gram staining of the smeared of the fluid sometimes revealed a very high (90%) neutrophils (MacWilliams and Friedrichs, 2003). Neutrophils may be degenerated by intra and extra-cellular bacteria. However, aseptic and careful aspiration of SF for microbial culture of nutrient or blood agar medium may flash a light for diagnosis and treatment. In this study synovial fluid from five joints were grown in nutrient broth, nutrient agar, blood agar, EMB agar and SDA medium to isolate bacteria and fungus. Following Grams staining and Giemsa's staining of the smears collected from the agar plate are *Staphylococcus* spp. in 2 cases, *E. coli* in one case and *Aspergillus* spp. from one case. In one case of arthritis no bacterial or fungus was revealed leaving the cause of arthritis undetermined in this study. However, attempt was not taken to identify either viral, mycoplasma or chlamydial cause of arthritis. *Chlamydia pecorum* causes a range of clinically important diseases in cattle, sheep, goats, and pigs manifesting as encephalomyelitis, reduced fertility, vaginitis and endometritis, enteric infections, mastitis, pneumonia, conjunctivitis, and arthritis (Jelocnik et al., 2013). The isolation and identification of virus (CAE), Mycoplasma and *Chlamydia pecorum* requires selective medium with sophisticated laboratory techniques, hence these diseases in arthritic goats was left unidentified.

Conclusion

This study investigated elbow and knee joints of 100 randomly selected free ranged goats and arthritic joints in five cases. Arthritis was seen in three females and two male goats. Three goats showed acute form of arthritis, whereas, two had chronic form. Elbow joints were affected more (n=04) than knee joint (n=01). The reason(s) of higher infectivity in elbow joints could not identified in this study but may be due to increase exposure to trauma and invasion of microbes. Synovial fluid (SF) of the infected joints appeared less viscous, yellowish to reddish in color. All of the non-infected SF showed solid ring in mucin clot test but the infected SF showed very weak band or did not form clot indicating successful adaptation of the test protocol. The synovial fluid from an infected joint recorded highest infiltration of monocytes and was infected with *Aspergillus* spp.

The causation of an arthritis was not identified although there was higher (2,200/ml) number of leukocytes in SF; this got may have infected with either Mycoplasma or Chlaymidial species require further investigation. The mucin clot test protocols adapted in this study for diagnosis of arthritis appeared successful. The neutrophilic infiltration (more than 2200/ml in the SF) in the SF may be an indication of bacterial arthritis. Isolation and identification of the causal agents is a prime goal to identify possible etiology and management of arthritis in the field levels. However, it needs to adapt test protocols to identify other causes of arthritis like Mycoplasm, Chlamydia or lenti virus (CAE virus) infection in the joints and accurately management of caprine arthritis.

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Competing interests

All the authors declare that they have no competing interests that might be sensed to influence the results and/or discussion of this manuscript.

Author contribution

AST and BSMSH conducted the research work. RMM and KMAHNA supervised the study.

Ethical approval

The research project titled "Caprine Arthritis is a Hidden Sickness Requires Early Detection and Management" has been approved by the Ethical Standard of Research Committee, Bangladesh Agricultural University Research System (BAURES), Bangladesh Agricultural University, Mymensingh-2202, Bangladesh.

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