Asian Journal of Medical and Biological Research ISSN 2411-4472 (Print) 2412-5571 (Online) www.ebupress.com/journal/ajmbr

Article

# Characterization of bacterial isolates from skin lesions of sheep, goat and cattle in different rearing condition

Tapu Kumar Saha<sup>1</sup>, Ferdousi Begum<sup>2</sup>, S. M. Lutful Kabir<sup>1</sup>, Md. Shafiqul Islam<sup>1</sup> and Md. Shahidur Rahman Khan<sup>1\*</sup>

<sup>1</sup>Department of Microbiology and Hygiene, Bangladesh Agricultural University, Mymensingh, Bangladesh <sup>2</sup>Veterinary Training Institute, Khakdohor, Department of Livestock Services, Dhaka, Bangladesh

\*Corresponding author: Professor Dr. Md. Shahidur Rahman Khan, Department of Microbiology and Hygiene, Bangladesh Agricultural University, Mymensingh, Bangladesh. E-mail: msrkhan001@yahoo.com

Received: 03 June 2019/Accepted: 25 June 2019/ Published: 30 June 2019

Abstract: The study was performed to isolate and characterize some selected bacteria from skin lesion of sheep, goat and cattle of different rearing condition. Intensive and semi-intensive housing systems were considered for this study. A total of (n=60) skin wound swab and pus samples were collected for this study during the study period. Among them (n=30) samples were collected from intensive farms and (n=30) samples were collected from semi-intensive farms. Staphylococcus aureus infections were found 70% skin lesion for sheep, 30% for goat and 60% for cattle and *Pseudomonas aeruginosa* infections were found 5% skin lesion for sheep, 15% for goat and 15% for cattle. Semi-intensive farm animals were more infected by bacteria than intensive farm animals. Staphylococcus aureus affected 53.33% in intensive farm animal and 66.67% in semi-intensive farm animal. Pseudomonas aeruginosa affected 10% animal in intensive housing system and 13.33% in semiintensive housing system. The organisms were isolated and characterized by morphological, cultural and biochemical characteristics and identified by polymerase chain reaction. Staphylococcus aureus produced vellowish colony in nutrient agar and golden vellow colony in mannitol salt agar. Positive isolates were produced  $\beta$ -hemolysis in blood agar. *Pseudomonas aeruginosa* produced characteristic colony in nutrient agar but not produce pigment. Pseudomonas aeruginosa produced colorless colony in MaCconkey agar, violet colony in EMB agar and produced hemolysis in blood agar. Staphylococcus aureus showed Gram positive, cocci shaped and grapes like arrangement and Pseudomonas aeruginosa showed Gram negative, short rod shaped. Staphylococcus aureus fermented five basic sugars and produced gas and Pseudomonas aeruginosa fermented dextrose and mannitol. Both organisms were catalase positive, only *Pseudomonas aeruginosa* was oxidase positive. Pseudomonas aeruginosa isolates were further characterized by PCR amplification of toxA gene primer. Staphylococcus aureus skin infection treatment was noticed more effective by erythromycin and gentamicin and Pseudomonas aeruginosa skin infection treatment more effective by ciprofloxacin and gentamicin. Both the organisms were found to be amoxicillin resistant. This is the first and preliminary report of molecular characterization of Pseudomonas aeruginosa from skin lesion of sheep, goat and cattle associated with a different rearing condition of the animals in Bangladesh.

Keywords: bacteria; skin lesion; cattle; sheep; goat; rearing condition

## 1. Introduction

The bacterial organisms that cause wounds on skins and hides of domestic animals have been investigated throughout the world (Sanousi *et al.*, 1989; Peregudon *et al.*, 1985). Bacteria such as *Staphylococcus aureus*, *Corynebacterium* spp., *Brevibacterium* spp. and *Acinetobacter* spp. live on normal skin and cause no harm. The most common bacteria to cause skin infections are: *Staphylococcus aureus* causes folliculitis, furunculosis (boils), impetigo, toxic shock syndrome etc. *Streptococcus pyogenes* cause cellulitis, erysipelas, impetigo,

necrotizing fasciitis and scarlet fever. Other less common bacterial infections that cause skin signs include: Ervsipelothrix insidiosa cause of ervsipeloid (usually an animal infection), Haemophilus spp. cause of cellulitis in young children, Klebsiella rhinoscleromatis cause of rhinoscleroma, Pseudomonas aeruginosa causes wound infections, athlete's foot, Gram negative folliculitis, chronic paronychia, Bacillus anthracis cause of anthrax, Clostridium perfringens and other species cause gas gangrene, Borrelia spp. cause Lyme disease, Mycobacterium spp. causes tuberculosis, leprosy and atypical mycobacterial infections, Serratia marcescens is a facultative anaerobic Gram negative Bacillus that may rarely cause skin infections such as cellulitis, abscesses and ulcers. These are more likely to arise in patients with immunodeficiency and *Tularemia* spp. a tick borne infection due to Francisella tularensis (Stulberg et al., 2002; Swartz, 2000; Wilkerson, 2002). Al-Harbi (2011) isolated Corvnebacterium pseudotuberculosis, Staphylococcus aureus subsp. anaerobius, Staphylococcus aureus, Streptococci, Pseudomonas aeruginosa, Actinomyces pyogenes were also isolated from infected abscesses from goat and sheep abscess. Intensive and semi-intensive housing systems are followed in domestic animal rearing in Bangladesh. The associated environment of farm causes various bacterial infections. Mazur et al. (2013) reported that the intensity of the epizootic situation in livestock-related pseudomonosis has increased due to changes in the methods of cultivation and animal keeping. In addition, in present conditions of intensive industries animals are exposed from birth to various stress factors which reduced their natural resistance, contributing to the rapid spread of pseudomonosis adaptation and reproduction of the pathogen in the body followed by isolation of bacteria in the environment. The methods of breeding, housing and feeding domesticated animals vary from species to species and differ according to custom and in accordance with geographical and climatic conditions. All kinds of wounds on skin and hides usually occur due to breach with very sharp instruments, imperfect brand marks, rubbing against course surfaces or incisions made by doctors during surgical operations. Breach in skins and hides, various kinds of wounds and injuries get contaminated by a big population of bacterial flora such as the pyogenic bacteria especially, Streptococcus pyogenes, Staphylococcus aureus and the Coliform group together with Proteus vulgaris welchii type A, Clostridium septicum, Clostridium diphtheriae, that cause damage to the qualities of leather which reflect in sense of losses to the leather industries and ultimately economy of the country. All kinds of abscess on skins and hides are the main cause of organisms as well parasites and become reason of contamination (Saini et al., 1992) Bangladesh has a fairly large livestock population to support a strong and growing tanning industry. Hides and skin Merchants Association Survey report 2005 showed that cow hides account for 56% of the production, goat skins for 3 30% and buffalo makes up the rest. The current output in Bangladesh is about 200 million sq. ft. of leather annually. Apart from bovine hides, buffalo, goat and sheep (Paul et al., 2013). Bacterial infections of skin degrade the quality of hides, skin and wool. That causes huge economic loss. Dermatitis, abscess, folliculitis, furunculosis (boils), impetigo, burns, parasitic infestation, cut, injury degraded hides and skin quality. The study of causal agent of skin infection helps to prevent degradation of hides and skin. Information on sheep and goat skin diseases in other countries is voluminous, but such information is very limited in Bangladesh. In addition to these, information on skin diseases of cattle is scanty in Bangladesh. Prevalence of bacterial skin diseases, lice, ticks and mite infestation in cattle, goat and sheep has been reported from Bangladesh (Huq and Mollah, 1972; Quader, 1973; Saikh et al., 1983). Most recently epidemiological and clinical features of skin diseases of cattle, goat, sheep caused by bacteria, lice, ticks and mite have been described by Nooruddin and Dey (1990). A work on Dermatophilosis in cattle was carried out by Mannan (2009) in Bangladesh. Moreover, Hossain et al. (2013) preliminarily reported *Pseudomonas aeruginosa* from abscess of cattle using cultural characterization instead of molecular characterization. The present research work was taken into consideration for further study focusing of cultural, biochemical and molecular characterization of selected bacterial isolates from skin lesions of sheep, goat and cattle in different rearing condition

#### 2. Materials and Methods

#### 2.1. Sampling areas and sample collection and transportation

The present research work was carried out considering the criteria of intensive and semi-intensive rearing system where 10 cattle, 10 goat and 10 sheep of Barisal Dairy and Breed Development Farm (BDBDF), Barisal Sadar, Barisal, Dairy Farm, BAU Mymensingh, Goat Farm, BAU, Mymensingh, sheep farm of Tangail sadar upazilla and 10 cattle, 10 goat and 10 sheep from semi-intensive farm of Narail sadar, Mymensingh sadar and Tangail sadar upazilla. Skin swab and pus samples of sheep, goat and cattle were collected aseptically using sterile cotton buds and placed in test tube containing nutrient broth and transported to the Laboratory of the Department of Microbiology and Hygiene, Bangladesh Agricultural University maintaining 4°C temperature in an ice box.

## 2.2. Isolation of *Staphylococcus* spp. and *Pseudomonas* spp.

The collected skin swab and pus samples were enriched into nutrient broth by incubating at 37<sup>o</sup>C for 24 hours. To isolate and study the cultural properties of *Staphylococcus* spp. and *Pseudomonas* spp., enriched cultured were streaked onto different selective and differential culture media like Mannitol salt agar, MacConkey's agar, EMB agar and Blood agar according to the methods described by Cowan 1985 where all of the media were brought from the Indian company, Himedia.

## 2.3. Identification of Staphylococcus spp. and Pseudomonas spp. by conventional methods

For identification of isolated *Staphylococcus* spp. and *Pseudomonas* spp., Gram's staining and biochemical tests were performed. Gram's staining was performed according to the method described by Merchant and Packer, 1967 where all of the reagents like crystal violet, Gram's iodine, safranin, acetone alcohol, immersion oil were brought from the German company, Merck. Different types of biochemical tests like sugar fermentation test, MR-VP reaction, indole reaction, Oxidase and catalase test were performed according to the methods described by Douglas *et al.*, 1998 and OIE, 2012 where all of the reagents were brought from the German company, Merck.

# **2.4.** Molecular detection of *Staphylococcus* spp. and *Pseudomonas* spp. by PCR assay **2.4.1.** Preparation of DNA templates

Extraction of DNA from the *Staphylococcus* spp. and *Pseudomonas* spp. was carried out by conventional boiling and rapid cooling method (Medici *et al.*, 2003). In brief, 200  $\mu$ l deionized water was taken into an eppendorf tube, a pure bacterial colony from nutrient agar was mixed with the deionized water. The tube then transferred to boiling water and boiled for 10 minutes then immediately to the icebox for cold shock about 10 minutes and then centrifuged at 10,000 rpm for 10 minutes. Supernatant were collected and used as DNA template during PCR.

## 2.4.2. PCR amplification of toxA gene of Pseudomonas aeruginosa and nuc gene of Staphylococcus aureus

Details of the oligonucleotide primers used for the amplification of *toxA* gene of *Pseudomonas aeruginosa* and *nuc* gene of *Staphylococcus aureus* are summarized in Table 1. PCR reaction mixture (25µl) for *Pseudomonas aeruginosa* was prepared using 12.5µl master mixture (Promega, USA), 10 pmol primer (Bioneer, South Korea) of each, 5µl DNA template and 5.5µl nuclease free water.

PCR reaction mixture (25 µl) for *Staphylococcus aureus* was prepared using 5 µl genomic DNA, 12.5 µl PCR master mixture (Promega, USA), 1 µl primer each (1 µl forward and 1 µl reverse primer) and 5.5 µl of nuclease free water. For the amplification of of *toxA* gene of *Pseudomonas aeruginosa*, the cycling conditions consisted of initial denaturation for 2 minutes at 95°C, followed by 30 cycles of denaturation at 95°C for 40 seconds, annealing at 50°C for 1 minute, extension at 72°C for 2 minute and final extension at 72°C for 10 minutes where the cycling conditions for the amplification of *nuc* gene of *Staphylococcus aureus* consisted of initial denaturation at 95°C for 5 minutes followed by denaturation at 95°C for 1 minute, annealing at 55°C for 45 seconds and extension at 72°C for 1 minute. The final extension was conducted at 72°C for 10 minutes. The PCR reaction was performed for 30 cycles. Amplification was performed in a thermal cycler (Eppendorf, Germany). The amplified products were electrophoresed into 1.5% agarose (Sigma-Aldrich, USA) gel at 100 volt visualized under Gel doc/UV trans-illuminator (BioRad). 100 bp (for *Salmonella* spp.) and 1 kb (for *E. coli*) DNA size marker (Promega, USA) were used.

## 2.5. Antibiogram profiles

Antibiogram was performed by employing the Kirby-Bauer disc diffusion method (Bauer *et al.*, 1959) using eight different commercially available antibiotic discs (HiMedia, India and Oxoid Ltd., England) on Mueller-Hinton agar (HiMedia, India) to assess the susceptibility and resistance pattern of the isolates. The selected antibiotics used were ciprofloxacin (5  $\mu$ g/disc), amoxicillin (30  $\mu$ g/disc), gentamicin (10  $\mu$ g/disc), erythromycin (30  $\mu$ g/disc), and tetracycline (30  $\mu$ g/disc). The interpretation on susceptibility was done according to the guidelines of Clinical and Laboratory Standards Institute (CLSI, 2012).

## 3. Results

## 3.1. Isolation and cultural characterization of *Pseudomonas* spp. and *Staphylococcus* spp.

The growth of *Pseudomonas* spp. and *Staphylococcus* spp. was indicated by the presence of turbidity in the nutrient broth after overnight incubation at 37°C. On Nutrient agar, *Pseudomonas* produces circular, smooth, mucoid colony with characteristics of grape like odor. *Pseudomonas* produces pale, colorless, smooth colonies

on MacConkey's agar and Smooth, circular, pink color colony on EMB agar. On blood agar, Pseudomonas produces white colony with hemolysis was produced. Staphylococcus produces circular, small, smooth, yellowish colonies on mannitol salt agar with changing the color of the medium from from bright red into yellow. On blood agar, white to golden colony with beta hemolysis were produced by Staphylococcus.

#### 3.2. Identification of *Staphylococcus* spp. and *Pseudomonas* spp. by conventional methods

In Gram's staining, the isolated *Staphylococcus* spp. revealed Gram positive, violet colored, cocci shaped organisms arranged in grape like clusters and *Pseudomonas* spp. revealed Gram negative, pink colored, short rod shaped organisms with singly arranged.

Fermentation of five basic sugars with the production of acid indicated that the isolates were *Staphylococcus spp*. positive while *Pseudomonas* spp. fermented dextrose and mannitol with the production of only acid but did not ferment maltose, lactose and sucrose. *Staphylococcus spp*. were found as MR test and indole positive where *Pseudomonas* spp. were found as MR test and indole negative. Both organisms (*Staphylococcus* spp. and *Pseudomonas* spp.) were found as catalase positive, VP test negative. *Pseudomonas* spp. specifically found as oxidase test positive.

## 3.3. Molecular detection of Staphylococcus aureus and Pseudomonas aeruginosa by PCR assay

DNA extracted from all of the isolates (36 *Staphylococcus aureus* and 7 *Pseudomonas aeruginosa*) were used in PCR assay. Polymerase chain reaction with the primers *nuc* gene (for *Staphylococcus aureus*) and *toxA* gene (for *Pseudomonas aeruginosa*) identified all of those isolates as positive for *Staphylococcus aureus* and *Pseudomonas aeruginosa* showing amplification of 279 bp and 150 bp respectively as presented in Figures 1 and 2.

#### 3.4. Results of distribution of selected bacteria present in different samples of different rearing condition

Animals reared in semi-intensive was affected skin diseases more than intensive farm. *Staphylococcus aureus* infection occurred 66.67% in semi-intensive and 53.33% infection in intensive farm. *Pseudomonas aeruginosa* infection found 13.33% in backyard farm and 10% infection in good husbandry farm. Nonspecific bacteria were found to be 36.66% in intensive farm and 20% in backyard farm. The results of bacterial distribution are shown in Table 2 and Table 3.

## 3.5. Results of distribution of bacterial isolates from different animal species

*Staphylococcus aureus* was isolated more than the *Pseudomonas aeruginosa* from skin sample of different animal. *Staphylococcus aureus* 60% and *Pseudomonas aeruginosa* 15% of cattle skin sample. *Pseudomonas aeruginosa* was isolated from 15% goat skin sample and *Staphylococcus aureus* was isolated from 70% goat skin sample. *Staphylococcus aureus* was isolated from 15% sheep skin samples. This result is shown in Table 4.

## 3.6. Results of antibiogram study of the isolated bacteria

From all bacterial isolates, ten (10) isolates of *Staphylococcus aureus* were selected randomly and *Pseudomonas aeruginosa* positive seven (7) isolates were selected for the antibiotic sensitivity test and resistance pattern against commonly used antibiotics. The results of sensitivity against antibiotic discs (zone of inhibition) were categorized as sensitive (S), intermediate (I), resistant (R).

## 3.6.1. Antibiotic sensitivity pattern of Staphylococcus aureus

From the antibiotic sensitivity test of *Staphylococcus aureus*, it was found that 80% of the isolated *Staphylococcus aureus* were sensitive to erythromycin, 70% to gentamicin, 60% to ciprofloxacin, 50% to tetracycline. The organisms showed 30% intermediate sensitivity to ciprofloxacin, 20% ton amoxicillin 10% to erythromycin, tetracycline and gentamicin. The isolates showed 80% of the *Staphylococcus aureus* were resistant to amoxicillin, 40% to tetracycline, 20% to gentamicin and 10% to ciprofloxacin, and erythromycin.

## 3.6.2. Antibiotic sensitivity pattern of Pseudomonas aeruginosa

From the antibiotic sensitivity test of *Pseudomonas aeruginosa*, it was found that 85.72% of the isolated *Pseudomonas aeruginosa* were sensitive to ciprofloxacin, 71.43% to gentamicin and 14.29% to tetracycline. The organisms showed 57.14% intermediate sensitivity to tetracycline, 42.85% to erythromycin, 14.29% to amoxicillin and ciprofloxacin. The isolates showed 85.72% of the *Pseudomonas aeruginosa* were resistant to amoxicillin, 57.14% to erythromycin and 28.57% to tetracycline.

# Table 1. List of Primers used in this study.

	Sequence (5'-3')		Amplicon	Reference
Primer		Target	size (bp)	
toxA F	5'GGAGCGCAACTATCCCACT3'			
toxA R	5'TGGTAGCCGACGAACACATA3'	toxA gene of Pseudomonas aeruginosa	150 bp	Sabharwal <i>et</i> <i>al.</i> (2014)
nuc F	5'-CGA TTG ATG GTG ATA CGGTT-3'		270.1	
nuc R	5'-ACG CAA GCC TTG ACG AAC TAA AGC-3'	Nuc gene of Staphylococcus aureus	279 bp	Dewanand <i>et</i> <i>al.</i> (2007)

# Table 2. Frequency distribution of bacteria from collected samples of skin of animals reared in intensive farm.

Organism	Cattle	Goat	Sheep	Total no. of	Percentage (%)	
				Isolates		
Staphylococcus aureus	5	6	5	16	53.33	
Pseudomonas aeruginosa	2	0	1	3	10	
Nonspecific, mixed	3	4	4	11	36.66	
and contaminant						
Total	10	10	10	30	100	

# Table 3. Frequency distribution of bacteria from collected samples of skin of animals reared in semiintensive farm.

Organism	Cattle	Goat	Sheep	Total no. of	Percentage (%)
				Isolates	
Staphylococcus aureus	7	8	5	20	66.67
Pseudomonas aeruginosa	1	1	2	4	13.33
Nonspecific, mixed	2	1	3	6	36.66
and contaminant					
Total	10	10	10	30	100

# Table 4. Percentage of bacterial isolates from different animal species.

Organism	Ca	Cattle		Sheep	Goat	
	No. of	% of	No. of	% of isolate	No. of isolate	% of
	isolate	isolate	isolate			isolate
Staphylococcus aureus	12(20)	60	14(20)	70	10(20)	50
Pseudomonas aeruginosa	3(20)	15	1(20)	5	3(20)	15

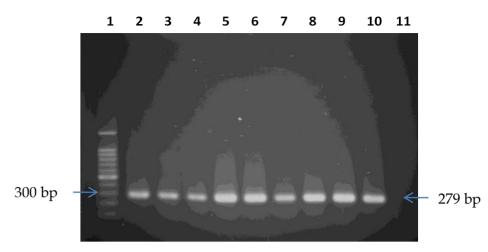


Figure 1. PCR of *Staphylococcus aureus* specific *nuc* gene (size = 279 bp). Here, Lane 1: 100 bp ladder, Lane 2-9: test samples positive for *nuc* gene, Lane 10: Positive control, Lane 11: Negative control.

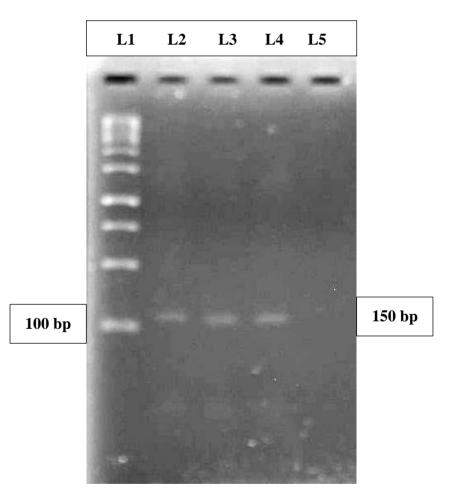


Figure 2. Results of PCR targeting *toxA* gene for identification of *Pseudomonas aeruginosa*. L1: 1 kb plus DNA ladder, L2, L3, L4: DNA of *Pseudomonas aeruginosa*, L5: Negative control.

# 4. Discussion

This is the study of isolation and identification of bacterial isolates of skin lesion from sheep, goat, and cattle. Other important focusing point of this study is rearing system. Intensive and semi-intensive rearing system is concerned in this study. Thirty samples were collected from sheep, goat and cattle of intensive rearing system and 30 samples were collected from semi-intensive rearing system. Skin wound swab and pus samples were collected for this study. Samples of intensive farms were collected from Dairy and Breed Development Farm

(BDBDF), Barisal Sadar, Barisal, Dairy Farm, BAU Mymensingh, Goat Farm, BAU, Mymensingh, sheep farm of Tangail sadar upazilla and samples of semi-intensive farm are collected from Narail sadar, Mymensingh sadar and Tangail sadar upazilla. In this study Gram positive Staphylococcus aerues and Gram negative Pseudomonas aeruginosa were isolated from skin lesions. The Gram positive Staphylococcus aerues characterized by morphology, cultural properties and biochemical test. In this study selective media were used for isolation of *Staphylococcus aureus*. The organism produced characteristic yellow color in mannitol salt agar and change the media color. Staphylococcus aureus produced yellow color in nutrient agar. Colony characteristics of *Staphylococcus aureus* in MS agar and morphological, cultural and biochemical characteristics of Staphylococcus aureus recorded in this study were similar to the findings of Das (2012), Chatterjee et al. (1990), and Jahan et al. (2014). They recorded yellowish colony on MS agar, cocci shaped in a grape like arrangement under the microscope. Catalase positive Staphylococcus aureus fermented five sugars and produced acid in this study. Das (2012) and Chatterjee et al. (1990) reported that fermentation of glucose, maltose, lactose, sucrose and mannitol fermentation with only acid production. In this experiment all isolates produced β-hemolysis in 5% bovine blood agar. Jahan et al. (2014) found 100% β-hemolysis production by 64 Staphylococci on 5% sheep blood agar. In this study 5% bovine blood was used because of some limitations of sheep blood collection. Gram negative Pseudomonas aeruginosa produced circular mucoid colony without pigment. Colorless colony produced in MacConkey agar and haemolysis in blood agar. The isolated organism fermented dextrose and mannitol and not fermented lactose. These characteristics colonies were similar with finding of Hossain et al. (2013) Haleem et al. (2011) and Wahba and Darrell (1965). The isolated Pseudomonas aeruginosa did not produce any characteristic pigment neither on nutrient agar nor on MacConkey agar and some atypical strain of *Pseudomonas aeruginosa* may not produce pigment on agar media and has been reported by some investigators (Wahba and Darrell, 1965 and Haynes, 1951). The isolated isolates of *Pseudomonas* aeruginosa were catalase and oxidase positive. This result was found to be similar with Ningthoujam and Shovarani (2008) and Gaby and Hadley (1957). Antibiotic susceptibility tests were performed against all Staphylococcal and Pseudomonas isolates according to instruction of National Committee on Clinical Laboratory Standard Institute (CLSI, 2012). From the antibiotic sensitivity test of Staphylococcus aureus, it was found that 80% of the isolated Staphylococcus aureus were sensitive to erythromycin, 70% to gentamicin, 60% to ciprofloxacin, 50% to tetracycline. This result almost similar with Jahan et al. (2014) and Thaker et al. (2013). Wei et al. (2014) described that Staphylococcus aureus resistant to penicillin 90.1%, erythromycin 48.6%, ciprofloxacin 36.9%, clindamycin 27.9%, gentamycin 18.9%, chloramphenicol 9.0% tetracycline 7.2% of these strains were observed. In this study it was found that the isolates showed 80% of the Staphylococcus aureus were resistant to amoxicillin, 40% to tetracycline, 20% to gentamicin and 10% to ciprofloxacin, and erythromycin which is almost similar with Wei et al. (2014). It was found that 85.72% of the isolated Pseudomonas aeruginosa were sensitive to ciprofloxacin, 71.43% to gentamicin and 14.29% to tetracycline. Pseudomonas aeruginosa showed highest sensitivity to Ciprofloxacin. This result was similar with Hossain et al. (2013). There were (n=60) samples collected from skin lesions of three species of animal. More number of samples from various location of Bangladesh was needed for successful accomplishment of this study. The study was performed only characterization of Staphylococcus aureus and Pseudomonas aeruginosa. Others bacteria involved skin infection were not possible to characterized in this study due to time constraint and lack of sufficient facilities. Only *Pseudomonas aeruginosa* and *Staphylococcus aureus* were identified by PCR.

#### 5. Conclusions

It may be concluded that *Staphylococcus aureus* and *Pseudomonas aeruginosa* may cause primary and secondary infection of skin in sheep, goat and cattle. Bacterial infections were more occurred in semi-intensive housing system. *Pseudomonas aeruginosa* and *Staphylococcus aureus* were successfully confirmed using PCR technique. Antibiogram profile of *Staphylococcus aureus* revealed that these isolates were highly sensitive to erythromycin and gentamycin, moderately sensitive to ciprofloxacin and resistant to amoxicillin. Antibiogram profile of *Pseudomonas aeruginosa* revealed that these isolates were highly sensitive to ciprofloxacin and gentamicin, moderately sensitive to tetracycline and resistant to erythromycin and amoxicillin. Erythromycin and gentamicin were more effective for the treatment of *Staphylococcus aureus* skin infection and ciprofloxacin and gentamicin were more effective for the treatment of *Pseudomonas aeruginosa* skin infection of sheep, goat, and cattle.

#### Acknowledgements

The authors are grateful to the Ministry of Science and Technology for financial support. Also thankful to the Head, Department of Microbiology and Hygiene of Bangladesh Agricultural University (BAU) for giving chance to use the laboratories during this research work.

#### **Conflict of interest**

None to declare.

## References

- Al-Harbi KB, 2011. Prevalence and etiology of abscess disease of sheep and goats at Qassim Region, Saudi Arabia. Vet. World, 4: 495-499.
- Chatterjee C, NC Nag and JP Ray, 1990. Studies on coagulase status and biochemical characters of animal strains of *Staphylococci*. Indian J. Anim. Health, 29: 157-161.
- CLSI, 2012. Performance standards for antimicrobial disk and dilution susceptibility tests for bacterial isolated form animals; approved standard third edition. Clinical and Laboratory Standards Institute (Formally NCCLS). 69-72. Communication 28 287-98.
- Cowan ST, 1985. Cowan and steel's Manual for Identification of Bacteria (2ndedn.). Cambridge University Press. Cambridge, London. pp. 158-160.
- Das S, 2012. Characteristics of methicillin-resistant and vancomycin sensitive staphylococcal isolated from laboratory animals and workers, MS thesis, Department of Microbiology and Hygiene, Bangladesh Agricultural University, Mymensingh.
- Gaby WL and C Hadley, 1957. Practical laboratory test for the identification of *Pseudomonas aeruginosa*. J. Bacteriol., 74: 356-358.
- Haleem H, J Kadhim, T Ilham and A Banyan, 2011. Isolation of *Pseudomonas aeruginosa* from clinical cases and environmental samples, and analysis of its 71 antibiotic resistant spectrum at Hilla Teaching Hospital. Med. J. Babylon, 8: 618-624.
- Haynes WC, 1951: *Pseudomonas aeruginosa* –its characterization and identification. J. Gen. Microbiol., 5: 939-950.
- Hide and Skin Merchants Association (HSMA), Survey report 2005, Dhaka, Bangladesh.
- Hossain MG, S Saha, MM Rahman, JK Singha and AA Mamun, 2013. Isolation, identification and antibiogram study of *Pseudomonas Aeruginosa* from cattle in Bangladesh. Journal of Veterinary Advances, 3: 180-185.
- Huq MM and MA Mollah, 1972. A survey on the prevalence of lice in sheep and goat of Dhaka and Mymensingh, Bangladesh. Bangladesh Vet. J., 6: 21-24.
- Jahan M, M Rahman, MS Parvej, SMZH Chowdhury, ME Haque, MAK Talukder and S Ahmed, 2014. Isolation and characterization of *Staphylococcus aureus* from raw cow milk in Bangladesh. J. Adv. Vet. Anim. Res., 2: 49-55.
- Mannan MA, MSR Khan, MM Rahman, F Begum and MZ Uddin, 2009. Isolation and identification of dermatophilus bacteria from the skin lesions of cattle. Bangl. J. Vet. Med., 7: 342-347.
- Mazur T and NN Sorokina, 2013. Epizootiological features of *Pseudomonas* Infection. Journal of Comparative Pathobiology, 6: 585-589.
- Merchant IA and RA Packer, 1967. Veteriary Bacteriology and Virology. Seventh Edition. The Iowa University Press, Ames, Iowa, USA. pp. 286-306.
- Ningthoujam DS and N Shovarani, 2008. Isolation and characterization of a *Pseudomonas aeruginosa* strain DN1 degrading p-nitrophenol. Res. J. Microbiol., 3: 345-351.
- Nooruddin M and AS Dey, 1990. Further study on the prevalence of skin disease in domiestic ruminants in Bangladesh. Bangladesh Veterinarian, 7: 75-81.
- OIE, 2012. Manual of standards for diagnostic test and vaccines.
- Paul HL, APM Antunes, AD Covington, P Evans and PS Phillips, 2013. Bangladeshi leather industry: an over view of recent sustainable developments. J. Soc. Leath. Tech. Ch., 97: 25-32.
- Peregudon TA, KB Mun and MT Isakov, 1985. *Staphylococcus* infection in goats. Veterinariya, Moscow, USSR. 2: 4-42.
- Quder SA, 1973. A survey on the prevalence of ticks of sheep and goat in Comilla Kotwali Pollice Station, Bangladesh. M.S thesis, BAU, Mymensingh.
- Saikh H, MM Huq, MJ Karim and MMM Khan, 1983. Parasites of zoonotic importance in domesticated ruminants. Pak. Vet. J., 3: 23-25.

- Saini NS, SN Sharma, MS Oberoy and KS Roy, 1992. Effect of operation theater environment on laparotomy wound infection in bovine. J. Vet. Med., 39: 258-263.
- Sanousi EI, AA Hamad and AA Gameel, 1989. Abscess disease in goats in the Sudan. Rev. Elev. Med. Vet. Pays. Trop., 42: 379-382.
- Stulberg D and M Penrod, 2002. Caring for common skin condition: common bacterial skin infections. Am. Fam. Physician, 66: 119-124.
- Swartz M, 2000. Cellulitis and Subcutaneous Tissue Infections. Mandell: Principles and Practice of Infectious Diseases, 5thed London: Churchill Livingtone, 1039-1040.
- Thaker HC, MN Brahmbhatt and JB Nayak, 2013. Isolation and identification of *Staphylococcus aureus* from milk and milk products and their drug resistance patterns in Anand, Gujarat. Vet. World, 6: 10-13.
- Wahba AH and JH Darrell, 1965. The identification of atypical strains of *Pseudomonas aeruginosa*. J. Gen. Microbiol., 38: 329-342.
- Wei W,Y Xiaojie,Y Xiaorong, M Lingling, G Wenying, M Guozhu, L Weiwei and G Yunchang, 2014. Antimicrobial resistance profiles and genetic diversity of bovine *Staphylococcus aureus* isolated in 5 provinces of China in 2013. Chi. J. Prev. Med., 48: 406-411.