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MULTI-DRUG RESISTANT *Staphylococcus aureus* ISOLATED FROM MILK, CHICKEN MEAT, BEEF AND EGG IN BANGLADESH

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ABSTRACT

Staphylococcal infection is one of the most common food-borne diseases in the world. Moreover, antimicrobial resistance of pathogenic bacteria, including *Staphylococcus* (*S.*) *aureus* is an emerging problem of food safety. This study was conducted to investigate the prevalence of *S. aureus* in milk, chicken meat, egg and beef; and to determine the multi-drug resistance (MDR) profile of *S. aureus* in Mymensingh and Gazipur districts, Bangladesh. A total of 189 samples of milk (n=108), chicken meat (n=51), egg (n=20) and beef (n=10) were collected from Bangladesh Agricultural University dairy farm, American dairy farm, Gazipur and different dairy farms of municipal area and retail shops during July 2016 to June 2018. *S. aureus* were isolated and identified by conventional methods and polymerase chain reaction (PCR). Antimicrobial susceptibility tests were done through disc diffusion test using 10 commonly used antibiotics. The overall prevalence of *S. aureus* in all food samples was 43.39%. A total of 39 (76.47%) chicken meat, 25 (23.15%) milk, 11(55%) egg and 07 (70%) beef samples were *S. aureus* positive through conventional method. Among 82 culture positive samples only 39 samples (47.56%) were confirmed by PCR. Antibiogram study showed that *S. aureus* isolated from chicken meat were mostly resistant to oxytetracycline (71.79%); and highly sensitive to amikacin (100%) and neomycin (100%). *S. aureus* isolated from milk samples were highly sensitive to neomycin (100%), and resistant to amikacin (56%). Only 28.57% isolates of *S. aureus* originated from beef samples were resistant to oxytetracycline and 100 % isolates were sensitive to ciprofloxacin, gentamicin, erythromycin, azithromycin, doxycycline. Similarly, *S. aureus* isolated from egg samples were resistant to erythromycin (81.82%) and 100% sensitive to amikacin. Out of 41.46% MDR isolates 12%, 53.85%, 90.91% and 0% of the *S. aureus* originated from milk, chicken meat, egg and beef respectively. The higher prevalence of *S. aureus* in chicken meat, beef, egg and milk indicates unhygienic production, marketing and processing of these foods. Presence of MDR *S. aureus* in these foods might pose serious public health threats. Rational use of antibiotics with higher sensitivity should be prescribed in managing poultry diseases to reduce re-emerging MDR in Bangladesh.

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INTRODUCTION

S. aureus (*S. aureus*) is an opportunistic pathogen in human and other different animal species. The pathogen is mainly related to food poisoning and is the third largest cause of food related illness throughout the world (Sasidharan et al., 2011; Achi and. Madubuike 2007). A number of infectious diseases such as dermatitis, pneumonia, meningitis, osteomyelitis in human, bovine mastitis in cattle and bumble foot disease in poultry (Ali et al., 2017) are produced by enterotoxigenic strains of coagulase-positive staphylococci, mainly *S. aureus* (Hennekinne et al., 2012). Among the food borne outbreaks bacterial toxin were 9.8% of which 55.4% cases were due to staphylococcal enterotoxins (Anonymous, 2010). There are some reports on the prevalence of Staphylococcus organism in poultry (Rahman and Samad, 2003, Ali et al, 2017), in milk (Mueena et al., 2015), in restaurant (49.3%) and household (Islam et al., 2016) cockroachs (26.7%), in pet (Habibullah et al., 2017) animal (40.86%) , and in frozen meat (95.83%) rinse (Islam et al.2014) in Bangladesh.

The use of antimicrobial compounds in food animal production provides improved animal health and reduces foodborne diseases in humans. However, use of antibiotics has been shown to contribute to the increased prevalence of antibiotic-resistant bacteria of human significance (Mathew et al., 2007). Multidrug resistant *S. aureus* in poultry meat have been reported in the USA (Andrew et al., 2011) and very limited research on antibacterial resistant in foods of animal origin (Ashrafudoulla et al. 2017) and no report was found on staphylococcal infection in beef in Bangladesh. Hence, this study was carried out to determine the prevalence of *S. aureus* along with multiple-drug resistance profile in in chicken meat, egg, beef and milk in Bangladesh.

MATERIALS AND METHODS

Study area and Collection of samples

The samples were collected randomly from farms and local markets situated in Mymensingh and Gazipur district of Bangladesh. A total of 189 (51 poultry meat, 20 egg, 10 beef and 108 milk) samples have been tested from July 2016 to June 2018. Milk samples have been collected from BAU dairy farm, American dairy farm, Gazipur, surrounding other local small dairy farm. Aseptically 8-10 ml of milk was collected in test tube directly from teat of lactating cow and local market and send to the Medicine laboratory using icebox. Ten to twenty grams of chicken breast meat or beef were collected aseptically in sterile zipper bag and send to the Medicine laboratory using ice box. Egg aseptically collected with sterilize zipper bag from different retail shop of municipal market, Mymensingh.

Sample preparation

10 to 20 grams of meat were mixed with 45 ml of peptone (0.1%) water then homogenized suspension was prepared using sterilized pestle and mortar. Egg surface were washed with peptone water (0.1%) within zipper bag.

Isolation and identification of *S. aureus*

The homogenized suspensions (4 to 5ml) and egg washing water were then transferred into nutrient broths (5 ml/test tube) separately, nutrient agar and other selective media (MSA). In every step, samples were incubated at 37^0C for 22-24 hours. The positive samples were then subculture several times to obtain pure *S. aureus*. Gram staining and Biochemical test (five sugar fermentation test) were done to be confirmed (Cheesbrough, 1985). Antibiotic disc diffusion test were done for *S. aureus* in Muller- Hinton agar (Hi-Media, India) according to CLSI, 2012.

Polymerase chain reaction (PCR)

DNA extractions were performed through boiling method (Goering *et al.*, 2008) with some modification. One or two single colonies of *Staphylococcus* spp. were suspended in 200 µl of distilled water and boiled for 10 minutes in a heat block, then placed on ice for 10 minutes. After centrifugation at 10,000 rpm for 10 minutes at 4^0 C, the supernatant (150 µl) was placed in an eppendorf tube and kept at -20^0 C until used as a DNA template. PCR assay were applied in all the 82 isolates to confirm the *Staphylococcus sp.* and *S. aureus* based on *16S rRNA* and *nuc* gene (Table 1) respectively. PCR reaction mixture were 25 µl consisting of RNAse free water 5.5 µl, PCR master mixture (Thermo Scientific, EU) 12.5 µl, genomic DNA 5 µl and primer (Reverse and Forward) 2 µl. The PCR protocol consisted of the following steps : i) Denaturation- 94^0C for 1 min, ii) 30 cycles of 94^0 C for 30 second, 55^0C for 30 second, 72^0C for 1 min, iii) Final Elongation 72^0C for 05 minute. PCR amplify products were subjected to gel (1.5% agarose, Takara, Japan) electrophoresis with ethidium bromide fluorescence (100 v for 30 minutes) and visualized in gel documentation system (Biosciences, Germany) via UV transilluminator (302 nm).

Table 1. Primer sequences and sizes of PCR-amplified targets

Primers	Sequence(5'-3')	Amplicon size(bp)	Reference
16S rRNA gene	F5'- AAC TCT GTT ATT AGG GAA GAA CA -3' R5'- CCA CCT TCC TCC GGT TTG TCA CC -3'	756	Islam et al. 2014
Nuc gene	F5'-GCGATTGATGGTGATACGGTT-3' R5'-AGCCAAGCCTTGACGAACTAAAGC-3'	297	Brakstad et al. 1992 Mueena et al., 2015

Detection of multi-drug resistant *S. aureus*,

Antimicrobial sensitivity test

Antimicrobial susceptibility of *S. aureus,* was performed by the disc diffusion test applied on Muller-Hinton agar (Hi-media, India) *in vitro* using 10 commercially available antibiotics (Oxoid, UK) e. g., oxytetracycline (30µg), ciprofloxacin (5µg), gentamicin (10µg), erythromycin, (15µg), azithromycin (15µg), sulphonamide-trimethoprim (25µg), neomycin (10µg), amoxicillin (10µg), doxycycline (10µg) and amikacin (30µg) according to the guidelines of the CLS1 (2012).

RESULT

Prevalence

The overall prevalence of *S. aureus* in all food samples was 43.39%. A total of 39 (76.47%) chicken meat, 25 (23.15%) milk, 11 egg (55%) and 07 (70%) beef samples were *S. aureus* positive (Table-2).

Cultural, staining and biochemical characteristics

The cultural characteristics showed that *S. aureus* produce turbid growth on nutrient broth and smooth white to grayish white colony on nutrient agar, golden-yellow color on manitol salt agar. On Gram staining, *S. aureus,* were found as Gram positive coccus and arranged as grape like. All five basic sugars like dextrose, maltose, lactose, sucrose and mannitol were fermented with the production of acid without gas. Coagulase positive *Staphylococcus* was detected by observing clamp or precipitation in coagulase test. The positive catalase test of *Staphylococcus* spp was determined by producing gas bubbles.

Molecular detection

The result of PCR is presented in Figure 1 and Figure 2. The amplicon size of PCR product was 756 bp (16*SrRNA* gene) and 297 bp (*nuc* gene) reconfirmed as *Staphylococcus sp.* and *S. aureus,* respectively.

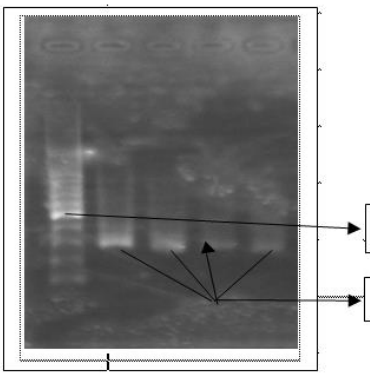

Figure 1. PCR product of *Staphylococcus aureus*
Showing Lane: 1 100 bp ladder
Lane: 2-5: *S. aureus* +ve (297 bp)

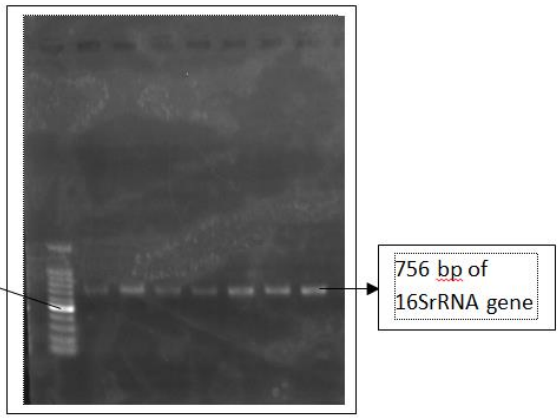

Figure 2. PCR product of *Staphylococcus sp*
Showing Lane: 1 100 bp ladder
Lane: 2-8: *Staphylococcus* +ve (756 bp)

Antibiogram study

The results of antibiotic sensitivity test have been shown in Table 3 and Fig. 3 and Fig. 4. *S. aureus* isolated from chicken meat were resistant to oxytetracycline (71.79%), azithromycin (64.10%) and erythromycin (58.97%); and sensitive to amikacin (100%), neomycin (100%), amoxicillin (94.87%), gentamicin (82.05%) and ciprofloxacin (71.79%). *S. aureus* isolated from milk samples were sensitive to neomycin (100%), gentamicin (92%) ciprofloxacin (92%), amoxicillin (88%), doxycycline (88%) and azithromycin (84%), and resistant to amikacin (56%) and erythromycin (44%). *S. aureus* isolated from beef samples were resistant to oxytetracycline (28.57%) and 100 % sensitive to ciprofloxacin, gentamicin, erythromycin, azithromycin, doxycycline, and 85.71 % sensitive to sulphonamide-trimethoprim, neomycin, amoxicillin and amikacin. *S. aureus* isolated from egg samples were 81.82% resistant to erythromycin, 72.73% resistant to ciprofloxacin, amoxicillin and sulphonamide-trimethoprim; and sensitive to amikacin (100%); 90.91% sensitive to gentamicin, amoxicillin and neomycin. Overall 41.46% of *S. aureus* isolates were found multi-drug resistant (MDR includes 3 or more antibiotic resistant). About 12%, 53.85% 90.91% and 0% of the *S. aureus* isolates originated from milk, chicken meat, egg and beef respectively were multi-drug resistant (Table 4).

Table 2. Prevalence of *Staphylococcus spp.* and *S. aureus*

Food Samples	Tested	Culture positive (%)	PCR +ve (%)	
			Staphylococcus sp.	***S. aureus***
Milk	108	25(23.15)	20 (80)	15
Chicken meat	51	39 (76.47)	19 (48.72)	01
Beef	10	07 (70.0)	0 (0)	0
Egg	20	11(55)	0 (0)	0
Total (%)	189	82(43.39%)	39 (47.56)	16 (19.5)

Table 3. Antibiogram study of *Staphylococcus aureus* isolates from different food samples

Antibiotics used	No. of Chicken meat (n=39)		No. of Milk (n=25)		No. of Beef (n=07)		No. of Egg (n=11)	
	R (%)	S (%)	R (%)	S (%)	R (%)	S (%)	R (%)	S (%)
Oxytetracycline	28 (71.79)	11 (28.94)	02 (20)	8 (80%), n*= 10	02 (28.57)	5 (71.43%)	7 (63.64)	4 (36.36)
Ciprofloxacin	11 (28.21)	28 (71.79)	02 (8)	23 (92%)	0	7 (100)	8 (72.73)	3 (27.27)
Gentamicin	07 (17.95)	32 (82.05)	02 (8)	23 (92%)	0	7 (100)	1 (9.09)	10 (90.91)
Erythromycin	23 (58.97)	16 (41.02)	11 (44)	14 (56%)	0	7 (100)	9 (81.82)	2 (18.18)
Azithromycin	25 (64.10)	14 (35.89)	04 (16)	21 (84%)	0	7 (100)	1* (9.09)	10* (90.91)
Sulphonamide and Trimethoprim	19 (48.71)	20 (51.28)	02 (8)	23 (92%)	01(14.29)	6 (85.71)	8 (72.73)	3 (27.27)
Neomycin	0	39 (100)	0	25 (100%)	01(14.29)	6 (85.71)	1 (9.09)	10 (90.91)
Amoxicillin	02 (5.12)	37 (94.87)	03 (12)	22 (88%)	01(14.29)	6 (85.71)	8 (72.73)	3 (27.27)
Doxycycline	16 (41.03)	23 (58.97)	03 (12)	22 (88%)	0	7 (100)	4 (36.36)	7 (63.64)
Amikacin	0	23 (100), n*=23	14 (56)	11 (44%)	01 (14.29)	6 (85.71)	0	11 (100)

R=Resistant, S= Sensitive, n* indicate No. of isolates used. *Indicate streptomycin (10µg/disc) were being used instead of azithromycin

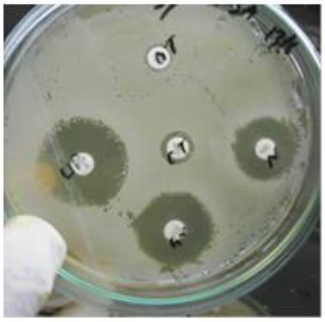

Figure 3. Antibacterial susceptibility test of *Staphylococcus aureus* on Muller Hinton Agar resistant to Oxytetracycline (OT), colistin Sulphate (CT), and Susceptible to Ciprofloxacin (CIP), Gentamicin (CN) and neomycin

Figure 4. Antimicrobial susceptibility test of *S. aureus* on Muller Hinton agar (beef sample). OT = Oxytetracycline, N = neomycin, N = Gentamicin, AK = Amikacin, ST = Sulphonamide and trimethoprim shows sensitive

Table 4. Detection of multi-drug resistant *S. aureus*

Antibiogram applied	Milk	Chicken meat	Beef	Egg	Total
No. of *S. aureus* isolates	25	39	07	11	82
Multi-drug resistant (%)	03 (12.00)	21 (53.85)	0 (0)	10 (90.91)	34 (41.46)

DISCUSSION

The overall prevalence of *S. aureus* in all food samples was 43.39%. We isolated *S. aureus* from 76.47% of chicken meat. Previous studies conducted in Bangladesh reported a wide range of *S. aureus* prevalence in chicken meat which varied from 24.56-95.83% (Islam et al., 2014; Datta et al., 2012; Sultana et al., 2014 and Andrew et al, 2011). This variation in prevalence with our findings might be due to the variation in the type of sample (Andrew et al, 2011) like raw and frozen sample. About 23% of our milk samples contained *S. aureus.* Mueena *et al.* (2015) also reported similar prevalence in milk sample (25.57%) sold from different open markets in Bangladesh. However, some authors reported higher prevalence (35.29-41.03%) of *S. aureus* in raw milk samples in Bangladesh (Tanzin et al., 2016); and in Tanzania (Jibril et al., 2018). We collected sample directly from teat which might reduce the chance of contamination in our samples. Around 70% of our beef samples were found to be contaminated with *S. aureus.* No inland literature was found about the prevalence of *S. aureus* in beef samples in Bangladesh. However, 39.7%, 9.4 % and 3% of the beef samples were reported to be contaminated with *S. aureus* in Sudan (Goja et al. 2013) , Ethiopia (Adugna et al. 2018) and Egypt (Osman et al. 2017) respectively. We detected *S. aureus* in 55% egg shell. However, other authors from Bangladesh reported higher prevalence of *S. aureus* from hen egg (66.67%) and duck egg (75%) in open market (Fardows et al. 2016; Fateha et al. 2018). These variations might be due to source of sample collection, hygienic condition of the farm and personal hygiene of the processors and handlers. Among the 82 culture positive samples only 39 (47.56%) samples were *Staphylococcus spp*. and 16 (19.51%) samples were *S. aureus* confirmed through PCR technique (Table 2). However, only 5.83% *S. aureus* in chicken and 15% *S. aureus* in beef were PCR positive out of conventional positive samples reported by Islam et al. (2014) and Ziad et al. (2014), respectively.

S. aureus isolated from our chicken meat samples were resistant to oxytetracycline (71.79%), azithromycin (64.10%) and erythromycin (58.97%); and sensitive to amikacin (100%), neomycin (100%), amoxicillin (94.87%), gentamicin (82.05%) and ciprofloxacin (71.79%). Similar findings also reported by other authors (Islam et al. 2016; Islam et al. 2014; Ali et al. 2017). *S. aureus* isolated from our milk sample were sensitive to neomycin (100%), gentamicin (92%) ciprofloxacin (92%), amoxicillin (88%), doxycycline (88%) azithromycin (84%) and oxytetracycline(80%), and resistant to amikacin (56%) and erythromycin (44%). Our results were supported by the findings of other authors (Islam et al. 2016; Tanzin et al., 2016; Hoque et al. 2018) from Bangladesh. *S. aureus* isolated from our beef sample were resistant to oxytetracycline (28.57%) and 100 % sensitive to ciprofloxacin, gentamicin, erythromycin, azithromycin and doxycycline; and 85.71 % sensitive to sulphonamide-trimethoprim, neomycin, amoxicillin and amikacin. These results were in contrast with the report of Sultana et al. (2014) who reported *S. aureus* as resistant to ciprofloxacin. However, Ashrafudoulla et al. (2017) reported *S. aureus* as sensitive to gentamicin and ciprofloxacin in human patients.

S. aureus isolated from our egg samples were 81.82% resistant to erythromycin, 72.73% resistant to ciprofloxacin, amoxicillin and sulphonamide-trimethoprim; and 100% sensitive to amikacin; 90.91% sensitive to gentamicin, azithromycin and neomycin. Similar to our report *S. aureus* were reported to be sensitive to gentamicin (Fateha et al. 2018; Ferdows et al., 2016). Ferdows et al. (2016) reported *S. aureus* as sensitive to ciprofloxacin which contradicts with our result. Similar to our result, Sangeda et al. (2017) reported *S. aureus* as resistant to ciprofloxacin and sulphonamide-trimethoprim in humans.

We observed 41.46% of *S. aureus* isolates as MDR (includes 3 or more antibiotic resistant) in which 12%, 53.85%, 90.91% and 0% of the *S. aureus* isolates originated from milk, chicken meat, egg and beef respectively. Similar to our findings, Andrew et al. (2011) reported 52 % MDR in poultry meat. Other authors also reported 8.3 to 63% of *S. aureus* isolates as MDR in different food samples (Datta et al. 2012; Sultana et al. 2014; Sangeda et al. 2017; Hoque et al. 2018; Fateha et al. 2018). Ashrafudoulla et al. (2017) also found MDR *S. aureus* in human clinical samples. Indiscriminate and recurrent use of different antibiotic agents in veterinary clinical practices could be the cause of high level of MDR.

CONCLUSION

The higher prevalence of *S. aureus* in milk, chicken meat, egg and beef indicates unhygienic production and processing of these foods. Presence of multi-drug resistant *S. aureus* in these foods may pose serious public health threats and this study may help the clinician as well as physician to take proper therapeutic decision to treat the cattle and poultry affected by *Staphylococcus* spp. in Bangladesh.

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REFERENCES

1. Achi OK and CN Madubuike, 2007. Prevalence and antimicrobial resistance of *S. aureus* isolated from retail ready to eat foods in Nigeria. Research Journal of Microbiology, 2 : 516-523.
2. Adugna F, M Pal, and G Girmay, 2018. Prevalence and Antibiogram Assessment of *S. aureus* in Beef at Municipal Abattoir and Butcher Shops in Addis Ababa, Ethiopia. Biomed Res Int. 2018; ID 5017685.
3. Ahmed T, S Baidya, BC Sharma, M Malek, kk as, M Acharjee, Sk munshi and R Noor, 2013. Identification of drug-resistant bacteria among.export quality shrimp samples in bangladesh. Asian Journal of Microbiology, Biotechnology and Environmental Science, 15(4): 31-36.
4. Ali Y, MA Islam,NH Muzahid MOF Sikder MA Hossain and LW Marzan, 2017. Characterization, prevalence and antibiogram study of *S. aureus* in poultry. Asian Pacific Journal of Tropical Biomedicine, 7(3): 253-256.

5. Andrew EW, T Contente-Cuomo, J Buchhagen, CM Liu, L Watson, K Pearce, JT Foster, J Bowers, EM Driebe, DM Engelthaler, PS Keim and LB Price, 2011. Multidrug-Resistant *Staphylococcus aureus* in US Meat and Poultry. Clinical Infectious Diseases, 52(10): 1227–1230.
6. Anonymous, 2010. The Community Summary Report on Trends and Sources of Zoonoses, Zoonotic Agents and food-borne outbreaks in the European Union in 2008. EFSA Journal, 1496: 1-376.
7. Ashrafudoulla M, M Rahman, MFR Mizan, MA Begum and SD Ha, 2017. Prevalence of multidrug resistant *S. aureus* in GonoShastho Nagar Hospital, Dhaka, Bangladesh. African Journal of Microbiology Research, 11(31): 1223-1229.
8. Brakstad OG, K Aasbakk and JA Maeland, 1992. Detection of Staphylococcus aureus by polymerase chain reaction amplification of the nuc gene. Journal of Clinical Microbiology, 30: 1654–1660.
9. CLSI (Clinical Laboratory Standard Institute), 2012. Performance standards for antimicrobial disk susceptibility tests: approved standard. 11th ed. Wayne: CLSI Publication M2-A11.
10. Datta, S, A Akter, IG Shah et al., 2012. Microbiological Quality Assessment of Raw Meat and Meat Products, and antibiotic susceptibility of Isolated *S. aureus.* Agriculture, Food and Analytical Bacteriology, 2 (3): 187-194.
11. Deraelle S, F Dilasser, M Duquenne and V Deperrois, 2009. Differential temporal expression of the staphylococcal enterotoxins genes during cell growth. Food Microbiology, 26: 896-904.
12. Fateha AE, M Arif, MA Islam and MM Khatun, 2018.Isolation and identification of duck egg-borne bacteria and their antibiogram profile. Journal of Advanced Veterinary and Animal Research, 5(2): 110-116.
13. Fardows J, AB Siddique, A Moureen, TAB Islam, N Farhana, C Rafia Naheen, 2016. Isolation and Identification of Pathogenic Gram-Positive Bacteria from Egg Shell of Hen and to See Their Antimicrobial Susceptibility Pattern. Journal of Enam Medical College, 6(1): 15-18.
14. Goering RV, Shawar RM, Scangarella NE, O'Hara FP, Amerine-Madsen H, West JM, et al. 2008. Molecular epidemiology of methicillin-resistant and methicillin-susceptible Staphylococcus aureus isolates from global clinical trials. Journal of Clinical Microbiology, 46:2842e7.
15. Goja AM**,** TAA Ahmed**,** SAM Saeed and HA Dirar**,** 2013**.** Isolation and Identification of *Staphylococcus* spp. in Fresh Beef. Pakistan Journal of Nutrition, 12 (2): 114-120.
16. Habibullah A, AMMT Rahman, MR Haydar, KHMNH Nazir and MT Rahman, 2017. Prevalence and molecular detection of methicillin-resistant staphylococcus aureus from dogs and cats in Dhaka city. Bangladesh Journal of Veterinary Medicine, 15 (1): 51-57
17. Hennekinne JA, Buyser MLD, and Dragacci S, 2012. *S. aureus* and its food poisoning toxins: characterization and outbreak investigation. *FEMS Microbiology Review* 36:815-836
18. Islam Al, D Nath A, K Islam, S Islam, S Chakma, MB Hossain, A Al-Faruq and MM Hassan, 2016. Isolation, identification and antimicrobial resistance profile of Staphylococcus aureus in Cockroaches (Periplaneta americana). Journal of Advanced Veterinary and Animal Research, 3 (3): 221-228.
19. Hoque MN, ZC Das, ANMA Rahman, MG Haider, MA Islam, 2018. Molecular characterization of *S. aureus* strains in bovine mastitis milk in Bangladesh. International Journal of Veterinary Science and Medicine, 6(1): 53-60.
20. Islam NN, M Akter, Z Farzana, AJB Kader, I Uddin, AMAMZ Siddiki and KM Kamaruddin, 2014. Detection of *S. aureus* in Frozen Chicken Rinse through Bacteriological and *Nuc* Gene Specific PCR Methods and their Drug Resistance Patterns in Southern Chittagong, Bangladesh. Research Journal of Microbiology, 9: 251-264.
21. Jibril M, MH Ziwa,YMG Hounmanou, A Kisanga, and HN Tuntufye, 2018. Molecular Typing and Antimicrobial Susceptibility of Methicillin-Resistant Staphylococcus aureus Isolated from Bovine Milk in Tanzania. International Journal of Microbiology. 2018: ID 4287431, 6
22. Osman K, A Alvarez-Ordóñez, L Ruiz, J Badr, F ElHofy, KS Al-Maary, IMI Moussa, AM Hessain, A Orabi, A Saad, and M Elhadidy, 2017. Antimicrobial resistance and virulence characterization of *S. aureus* and coagulase-negative staphylococci from imported beef meat. Annals Clinical Microbiology and Antimicrobials, 16: 35.

23. Mathew AG, R Cissell and S Liamthong, 2007. Antibiotic Resistance in Bacteria Associated with Food Animals: A United States Perspective of Livestock Production. Foodborne pathogens and Diseases, 4(2): https://doi.org/10.1089/fpd.2006.0066.
24. Mueena J, M Rahman, MS Parvej, SMZH Chowdhury, ME Haque, MAK Talukder and S Ahmed 2015. Isolation and characterization of Staphylococcus aureus from raw cow milk in Bangladesh. Journal of Advance Veterinary and Animal Research, 2(1): 49-55.
25. Rahman MA and Samad MA, 2003. Pattern of occurrence of single and concurrent diseases associated with mortality in commercial chickens in Bangladesh. Bangladesh Journal of Veterinary Medicine, 1(1): 15-20.
26. Rahn K, De Grandis SA, RC Clarke, R Curtiss, CL Gyles, et al., 1992. Amplification of an invA gene sequence of Salmonallatyphimurium by polymerase chain reaction as a specific method of detection of Salmonella. Molecular and Cellular Probes, 6: 271-279.
27. Samad MA, 2011. Public health threat caused by zoonotic diseases in Bangladesh. Bangladesh Journal of Veterinary Medicine, 9 (2): 95 – 120.
28. Sangeda RZ, S Lifumo, AWT Muigai and S Kariuki, 2017. Antimicrobial susceptibility patterns of Staphylococcus aureus and coagulase negative staphylococci isolated from humans in Nairobi, Kenya. African Journal of Microbiology Research 11(18): 705-711
29. Sasidharan S, B PrFateha, LL Yoga, 2011. Antimicrobial drug resistance of *S. aureus* in dairy products**.** Asian Pacific Journal of Tropical Biomedicine, 1 (2): 130-132.
30. Sultana F, Kamrunnahar, H Afroz, A Jahan, M Fakruddin, and S Datta, 2014. Multi-antibiotic resistant bacteria in frozen food (ready to cook food) of animal origin sold in Dhaka, Bangladesh. Asian Pacific Journal of Tropical Biomedicine, 4(1): 268-271.
31. Tanzin T, KHMNH Nazir, MN Zahan, MS Parvej, K Zesmin and MT Rahman, 2016. Antibiotic resistance profile of bacteria isolated from raw milk samples of cattle and buffaloes. Journal of Advanced Veterinary and Animal Research. 3 (1): 62-67.
32. Ziad W Jaradat, Yaser H. Tarazi and QO Ababneh, 2014. Molecular characterization of *S. aureus* isolated from meat and their antibiotic resistance profiles. Pakistan Veterinary Journal, 34(1): 58-62.