European Journal of Veterinary Medicine
www.ej-vetmed.org

Molecular Detection of Methicillin Resistant *Staphylococcus aureus* (MRSA) in Poultry in Bangladesh: Having Public Health Significance

Md. Jannat Hossain, M. Sohidullah, Muhammad Ashiqul Alam, Md. Shahin Al Mamun, Yassien Badr, Hend Altaib, and Md. Matiur Rahman*

ABSTRACT

Methicillin-resistant *Staphylococcus aureus* (MRSA) has a major public health concern. It can be identified throughout the chain of production for poultry, which raises questions regarding potential transmission from farm to consumer. MRSA has zoonotic significance and can be transmitted to humans and poultry. Several studies have been carried out on MRSA on poultry, but this study was conducted to find out the whole scenario of MRSA at the farm level. A total of 100 samples were collected randomly from different poultry farms and retail shops in Khulna city to investigate this study. MRSA was isolated and identified by culturing antibiotic susceptibility testing, and polymerase chain reaction (PCR). Among the 100 samples, 57% were positive for *S. aureus* and 80.70% of the isolated *S. aureus* showed hemolysis on blood agar. Among the 57 isolates, 78.94% were MRSA (oxacillin) and 19.29% were vancomycin-resistant *Staphylococcus aureus* (VRSA) phenotypically. Surprisingly, 59.64% of *S. aureus* results showed a positive *mecA* gene. It is also concerning that 60% of broiler meat and 53.84% of farm personnel were infected with MRSA. The present study revealed that MRSA could be transmitted from poultry to humans.

Keywords: MRSA, Public health, Poultry, VRSA.

Published Online: December 4, 2022

ISSN: 2736-6596

DOI :10.24018/ejvetmed.2022.2.6.69

M. J. Hossain
Department of Microbiology and Public Health, Faculty of Veterinary, Animal and Biomedical Sciences, Khulna Agricultural University, Bangladesh.
(e-mail: md.jannat.hossain@gmail.com)
M. Sohidullah
Department of Microbiology and Public Health, Faculty of Veterinary, Animal and Biomedical Sciences, Khulna Agricultural University, Bangladesh.
(e-mail: sohidullah038@gmail.com)
M. A. Alam
Department of Microbiology and Public Health, Faculty of Veterinary, Animal and Biomedical Sciences, Khulna Agricultural University, Bangladesh.
(e-mail: ashiq.alam@gmail.com)
M. S. A. Mamun
Satosho Co., Ltd. Gifu-Shi, Japan.
(e-mail: shahinfmrt83@yahoo.com)
Y. Badr
Department of Animal Medicine (Branch of Infectious Diseases), Faculty of Veterinary Medicine, Damanhour University, Egypt.
(e-mail: yassienbadr1@gmail.com)
H. Altaib
Middle East for Veterinary Vaccines (MEVAC®), Egypt.
(e-mail: hend.eltayeb@me-vac.com)
M. M. Rahman*
Department of Medicine, Faculty of Veterinary, Animal and Biomedical Sciences, Sylhet Agricultural University, Bangladesh.
(e-mail: matiur.vetmed @ au.ac.bd; matiur.vetmed@ gmail.com)

Corresponding Author

I. Introduction

Staphylococcus aureus is an opportunistic pathogen that causes numerous skin and food-borne illnesses in animals, poultry, and humans [1]. It is well established that *S. aureus* has a potential zoonotic significance and poses a serious foodborne illness risk to public health on a global scale [2], [3]. *S. aureus* could be isolated as a normal microflora from chicken skin, feathers, respiratory tracts, and intestinal tracts, just like in humans and other animals [4], [5]. However, various clinical problems, including dermatitis, arthritis, osteomyelitis, synovitis, tenosynovitis, femoral head necrosis, bumble-foot, and omphalitis, may be linked to this pathogen [6].

Methicillin-resistant *Staphylococcus aureus* (MRSA) is a common human and animal pathogen that has been frequently identified in clinical studies [7]. MRSA first appeared in the world in 1961 in England [8]. MRSA transmission from animals to humans through workplace livestock interaction. The 78 KDa protein known as PBP2a,

which binds to penicillin, is the mediator of MRSA [9]. This protein is encoded by the *mecA* gene, which is located on a substantial mobile genetic material known as the staphylococcal chromosomal cassette *mec* (SCC*mec*) [10].

Antibiotic-resistant bacteria in animals are of great concern due to their potential impact on human transmission [11]. Several types of meat, including raw chicken meat, have been linked to MRSA [12]. *S. aureus* can acquire many resistance genes and grow more virulent due to the widespread use of antimicrobial agents in the poultry sector [13]. As a result, the efficacy of preventative and control measures could be restrained by the transmission of these resistance genes and the resistant bacteria to humans through poultry [14]. Very few studies have been carried out on MRSA in poultry in Bangladesh focusing only on meat samples. In this study, we investigated MRSA in different poultry samples, including the poultry farm samples, and its potential transmission to humans.

II. Materials and Methods

A. Collection of Samples

From July 2021 to June 2022, 100 samples (broiler meat = 20, cloacal swab = 20, litter = 20, feces = 20, and hand washings of farm personnel = 20) were collected hygienically from different retail markets and poultry farms in Khulna city, Bangladesh. Then it was transferred properly with bacteriological transport media to the Bacteriology laboratory, Department of Microbiology and Public Health, Khulna Agricultural University for bacteriological analysis. Fig. 1 shows the study area of Khulna city.

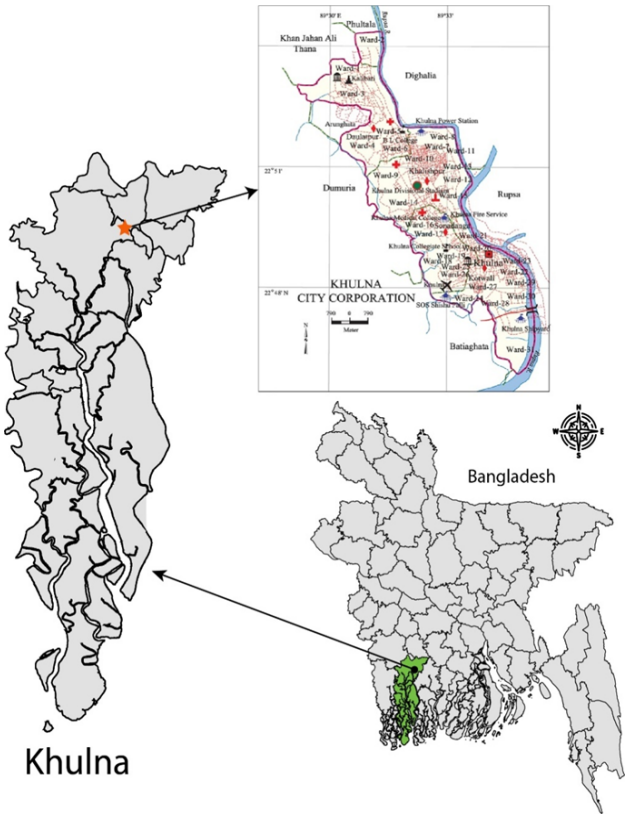

Fig. 1. Sample collection area (Khulna city) [15].

B. Isolation and Identification of Staphylococcus aureus

Firstly, *Staphylococcus* spp. was separated by using culture media containing Mannitol Salt agar (MSA). The yellow-colored colonies on MSA were Gram's stained, and the Gram-positive cocci bacteria having a clustered arrangement were considered *S. aureus* [16]. The hemolytic activity of the isolated bacteria was detected following growth on blood agar [16].

C. Antibiotic Sensitivity Test

An antibiotic sensitivity test was performed by disk diffusion on Mueller-Hinton agar (Hi Media, India) plates having a concentration of bacteria equivalent to 0.5 McFarland standards [17]. The plates were incubated at 37 °C aerobically for 18–24 hours to observe the results. Results of the antibiotic sensitivity tests were recorded as sensitive, intermediately sensitive, or resistant, and the zone of growth inhibition was compared with the zone size interpretative tables provided by the Clinical and Laboratory Standards Institute [18]. The antibiotic disc was used to determine the antibiogram of oxacillin (methicillin) at a dose of 1µg/disc. In addition, ciprofloxacin (5µg), ceftriaxone (30µg), amoxicillin (30µg), trimethoprim-sulfamethoxazole (25 µg), and vancomycin (30µg) also used in the antibiogram study. Any isolate showing resistance to oxacillin will be considered MRSA and resistance to vancomycin as VRSA.

D. Molecular Detection of Methicillin-resistant S. aureus (MRSA)

Boiling was used to recover the genomic DNA from *S. aureus* [19]. *S. aureus* detection was confirmed using polymerase chain reaction (PCR) with the *nuc* gene as the target. [20]. A final 25 µl reaction including, 12.5 µl of master mixture 2X (Promega, USA), 2 µl of genomic DNA (about 30 ng), 1 µl (100 pmol) of individual primer, and 8.5 µl nuclease-free water, was used for PCR. PCR Products were examined by electrophoresis on 1.5% agarose gel. The gel was stained with ethidium bromide and examined with an ultraviolet transilluminator (Biometra, Germany). A 100 bp DNA ladder (Promega, USA) was used as a molecular weight marker.

TABLE I: Primers used in PCR for *nuc* and *mecA* gene

Primers	Primer sequence (5'-3')	Product size (bp)	Reference
nuc F	5'-GCG ATT GAT GGTGAT ACG GTD-3'	279	[21]
nuc R	5'-AGC CAA GCC TTG ACG AAC TAA AGC-3'		
mecA F	5'-AAA ATC GAT GGT AAA GGT TGGC-3'	533	
mecA R	5'-AGT TCT GGC ACT ACC GGA TTT TGC-3'		

E. Statistical Analysis

To calculate the frequencies of MRSA, all the obtained data were entered into an excel spreadsheet (MS-2013) and descriptive statistics were done using SPSS software (SPSS-26.0, IBM, USA).

III. Result

The prevalence of MRSA in poultry farms was identified by phenotypic and genotypic observations. The yellow color colony characteristics of *S. aureus* were observed on MSA (Fig. 2). These colonies showed Gram-positive clustered coccus shape by Gram staining. Additionally, a zone of hemolysis was observed on blood agar. These results indicated the existence of *S. aureus*.

Table II showed that among 100 samples, 57% were positive for *S. aureus*, with the highest prevalence in poultry litter (70%). Among 57 isolates of *S. aureus*, 80.70% produced hemolysis on blood agar. Poultry litter isolates showed the highest degree (85.71%) of hemolysis. Table II also showed various degrees of MRSA in broiler meat (60%), cloacal swabs (66.67%), feces (50%), and hand washing of farm personnel (53.84%).

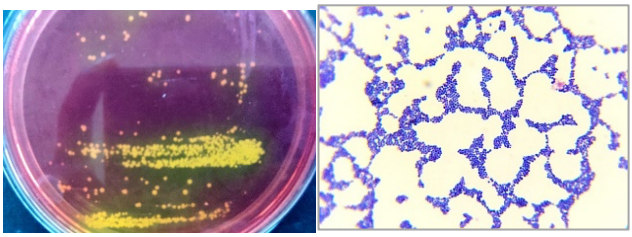

Fig. 2. Yellow colonies of *S. aureus* in Mannitol salt agar and Gram-positive cocci with cluster arrangement of *S. aureus*.

TABLE II: Prevalence of Methicillin Resistant *S. aureus* in Poultry

Sample (n)	Positive for *S. aureus* (*nuc*) (%)	Prevalence of MRSA (*mecA*) (%) among the *S. aureus*	Hemolysis on blood agar (%) among the *S. aureus*
Broiler meat (20)	10 (50.00)	6 (60.00)	8 (80.00)
Cloacal swab (20)	12 (60.00)	8 (66.67)	10 (83.33)
Feces (20)	8 (40.00)	4 (50.00)	6 (75.00)
Litter (20)	14 (70.00)	9 (64.28)	12 (85.71)
Hand washing (20)	13 (65.00)	7 (53.84)	10 (76.92)
Total (100)	57 (57.00)	34 (59.64)	46 (80.70)

Fig. 3 shows the phenotypic results of MRSA in different poultry samples. Among the 57 isolates, 78.94% were MRSA and 19.29% were VRSA phenotypically (Table III). The various degrees of resistance to ciprofloxacin (36.84%), ceftriaxone (10.52%), amoxicillin (26.31%), and trimethoprim-sulfamethoxazole (35.08%) (Table III).

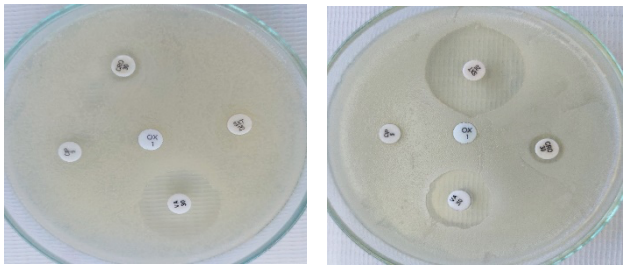

Fig. 3. Antibiotic susceptibility of the isolated *S. aureus* from poultry samples on Mueller Hinton agar plates each with five antibiotic discs.

Molecular detection of *S. aureus* was confirmed by PCR (Fig. 4 & 5). Overall, 59.64% *mecA* genes were found in isolated poultry samples, with the highest percentage (66.67%) found in cloacal swabs (Table II). Table II also confirmed MRSA in various poultry samples such as broiler meat (60%), feces (50%), and litter (66.28%). MRSA was also detected in human samples at 53.84% (Table II).

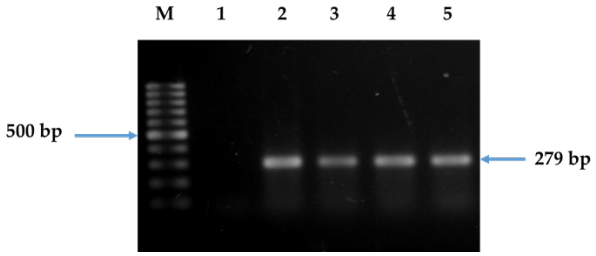

Fig. 4. Molecular detection of *nuc* gene of *S. aureus*. PCR amplification of *nuc* gene of *Staphylococcus aureus*. M=100 bp size DNA marker, Lane 1: negative control, Lane 2: positive control, Lane 3 to 5: representative samples of *S. aureus* isolated from different poultry samples.

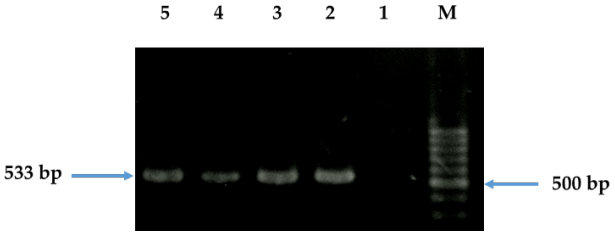

Fig. 5. Molecular detection of *mecA* gene of methicillin-resistant *S. aureus*. PCR amplification of *mecA* gene of methicillin-resistant *Staphylococcus aureus* (MRSA). M=100 bp size DNA marker, Lane 1: negative control, Lane 2: positive control, Lane 3 to 5: representative samples of MRSA isolated from different poultry samples.

IV. Discussion

MRSA has the potential to be harmful to both human and animal health. MRSA is frequently found in poultry farms [1]. In this study, we observed the overall prevalence of MRSA in various poultry samples was 59.64%. Several studies on MRSA in poultry have been carried out around the world. Previously, El-Tawab *et al.* [6] detected 66.6% MRSA from chicken samples in Giza, Egypt. Recently, Benrabia *et al.* [22] reported 33.5% MRSA in laying hens and 15.9% MRSA in broilers in Algeria. Earlier, Olayinka *et al.* [5] also reported 39.8% MRSA from poultry farm isolates in Zaria, Nigeria. In the current study, we found 60% MRSA from broiler meat samples. Earlier, Parvin *et al.* [23] reported 43.5% MRSA from frozen meat samples in Bangladesh. Similarly, 43% of MRSA was reported from raw chicken meat in Malaysia [24]. During this study, 66.67% of the cloacal swabs were infected with MRSA, whereas 32.60% were found in Nigeria [25]. Surprisingly, 53.84% of the poultry farmers' hand wash samples were contaminated with MRSA, whereas 83.3% were reported to health workers in Brazil [26].

TABLE III: Resistant Pattern of *S. aureus* Isolated from Poultry

Sources (n)	Resistant pattern to applied antibiotics (%)					
	OX	CIP	CRO	AX	SXT	VA
Broiler meat (10)	7 (70.00)	5 (50.00)	2 (20.00)	3 (30.00)	1 (10.00)	0 (00.00)
Cloacal swab (12)	10 (83.33)	3 (25.00)	0 (00.00)	2 (16.66)	5 (41.66)	2 (16.66)
Feces (8)	6 (75.00)	3 (37.50)	1 (12.50)	4 (50.00)	2 (25.00)	3 (37.50)
Litter (14)	12 (85.71)	6 (42.85)	3 (21.42)	5 (35.71)	7 (50.00)	4 (28.57)
Hand washing (13)	10 (76.92)	4 (30.76)	0 (00.00)	1 (7.69)	5 (38.46)	2 (15.38)
Total (57)	45 (78.94)	21 (36.84)	6 (10.52)	15 (26.31)	20 (35.08)	11 (19.29)

Here, n= Number of *S. aureus*, OX = Oxacillin, CIP = Ciprofloxacin, CRO= Ceftriaxone, AX = Amoxicillin, SXT = Trimethoprim-sulfamethoxazole, VA=Vancomycin.

Different classes of antibiotics are frequently used for the treatment of poultry in Bangladesh [27]. Most of them are β-lactam antibiotics, fluoroquinolones, and trimethoprim-sulfonamides. In this study, we observed 78.94% methicillin (oxacillin) resistance to *S. aureus* from poultry isolates, where the highest resistance was found in the case of poultry litter (85.71%). Previously, Parvin *et al.* [23] and Hossain *et al.* [28] detected 42.1% to 97.7% oxacillin-resistant *S. aureus* in poultry samples in Bangladesh. Recently, Benrabia *et al.* [22] reported 100% oxacillin-resistant *S. aureus* from poultry isolates in Algeria. Earlier, EL-Adaway *et al.* [29] observed 100% resistance to oxacillin in *S. aureus* isolates from broilers in Germany. Ciprofloxacin is widely used for the cure of different poultry diseases in Bangladesh [27]. Overall, 36.84% resistance was found in *S. aureus* isolates against ciprofloxacin during this study, while 50% resistance was observed in the case of broiler meat samples, which is very alarming for consumers. On the other hand, 50% resistance to ciprofloxacin against *S. aureus* was also observed by Benrabia *et al.* [22] in poultry species in Algeria. Earlier, EL-Adaway *et al.* [29] also observed 55.6% resistance to ciprofloxacin in *S. aureus* isolates from broilers in Germany.

Ceftriaxone is a semi-synthetic third-generation antibiotic that is effective in treating many septicemic diseases of poultry [27]. In this study, we observed 10.52% ceftriaxone resistance against *S. aureus* isolated from poultry samples. Previously, Ugwu *et al.* [30] reported 65.91% ceftriaxone resistance in *S. aureus* from poultry in Nigeria. We also found 26.31% amoxicillin-resistant *S. aureus* from poultry samples where 50% was identified from feces. Parvin *et al.* [23] reported 87% amoxicillin resistance to *S. aureus* from frozen chicken meat samples in Bangladesh. Previously, Otalu *et al.* [13] and Sallam *et al.* [31] reported 23% and 77.8% amoxicillin-resistant *S. aureus* from retail chicken in Nigeria and Egypt, respectively. Overall, 35.08% of trimethoprim-sulfamethoxazole resistance against *S. aureus* was observed during this study, while 50% was detected from poultry litter. Previously, 27.8% to 38.55% of trimethoprim-sulfamethoxazole resistance to *S. aureus* was documented in Nigeria and Algeria [13], [22].

Vancomycin is not commonly used for the treatment of poultry infections. But various degrees of vancomycin resistance were found during antibiotic susceptibility testing of poultry samples. For decades, vancomycin has been frequently used where MRSA is found [32], [33]. In this study, we found 19.29% VRSA from poultry, with the highest (37.5%) resistance observed from feces. Recently, El-Ghany [1] reported 54% VRSA from poultry in northwest Algeria. Earlier, Otalu *et al.* [13] also reported 46.2% VRSA from poultry samples in Nigeria.

The present study also confirmed the molecular detection of *S. aureus*, where 57% were found positive for the *nuc* gene. Recently, Shahid *et al.* [34] also detected 50.62% *nuc* gene of *S. aureus* from food processing environments in Bangladesh. Earlier, Akilu *et al.* [24] reported 77% *nuc* gene of *S. aureus* from raw chicken meat samples in Malaysia. We also found 59.64% positive *mecA* gene of methicillin-resistant *S. aureus* from poultry samples. Previously, Parvin *et al.* [23] detected the 43.5% *mecA* gene of MRSA from frozen broiler meat samples in Bangladesh. Earlier, varying degrees (37.2%-100%) of the *mecA* gene of MRSA were found positive in poultry samples in Egypt and Germany [6], [27], [29], [35].

Multidrug-resistant genes of MRSA found in poultry farming in Bangladesh result in the burden of medical costs and great health concerns for humans and animals. More host-adaptive evolutionary alterations may occur in animal-adapted clones, which might lead to the epidemic spread of new and more virulent MRSA strains among humans. Additional investigations are still required to determine the sources of MRSA infections in humans and the validity of animal-to-human transmission findings.

V. Conclusion

MRSA found in poultry and poultry farm personnel is a major health issue that indicates the zoonotic significance of MRSA. In this study, we found MRSA and VRSA from broiler meat samples, which reveals the direct threat to the consumer's health. The findings underscore the necessity of mitigating actions based on the farm-to-fork principle at all points in the poultry production chain to prevent or eradicate MRSA transmission in humans, poultry, and farms.

Acknowledgment

We sincerely acknowledge the Ministry of Science and Technology (MoST), Government of the People's Republic of Bangladesh for providing the research grant to carry out the research.

Funding

The research work was funded by R & D project grant 2021-2022 (grant number: 42, Agriculture and Environment Group), Ministry of Science and Technology, Bangladesh.

Conflict of Interest

The authors declare that there is no conflict of interest regarding the publication of this paper.

References

[1] Abd El-Ghany WA. *Staphylococcus aureus* in poultry, with special emphasis on methicillin-resistant strain infection: A comprehensive review from one health perspective. *Int J One Health* 2021; 7(2): 257-267.

[2] Shahid AH, Nazir KNH, El Zowalaty ME, Kabir A, Sarker SA, Siddique MP, *et al.* Molecular detection of vancomycin and methicillin resistance in *Staphylococcus aureus* isolated from food processing environments. *One Health* 2021; 13: 100276.

[3] Zaman SB, Sobur MA, Hossain MJ, Pondit A, Khatun MM, Choudhury MA, *et al.* Molecular detection of methicillin-resistant *Staphylococcus aureus* (MRSA) in ornamental birds having public health significance. *J Bangla Agri Univ.* 2020; 18(2): 415–420.

[4] Casey AL, Lambert PA, Elliott, TSJ. 2007. Staphylococci. *Int J Anti Agen.* 2007; 29: S23-S32.

[5] Olayinka BO, Bala HK, Ehinmidu JO, and Onaolapo, J.A. Multidrug resistant *Staphylococcus aureus* isolates from poultry farms in Zaria, Nigeria. *14th International Symposium on Staphylococci and Staphylococcal Infections*; 2010 Sep 6-9; Bath, UK.

[6] El-Tawab A, Ashraf A, Hofy FI, Mohamed SR, Amin SH. Characterization of Methicillin Resistance *Staphylococcus aureus* isolated from chicken and human. *Ben Vet Med J.* 2017; 32(1):132-137.

[7] Persoons D, Van Hoorebeke S, Hermans K, Butaye P, De Kruif A, Haesebrouck F, *et al*. Methicillin-resistant *Staphylococcus aureus* in poultry. *Emer Infect Dis.* 2009; 15(3): 452.
[8] Fluit AC. Livestock-associated *Staphylococcus aureus*. *Clin Microb Infect.* 2012; 18(8): 735-744.
[9] Ali Y, Islam MA, Muzahid NH, Sikder MOF, Hossain MA, Marzan LW. Characterization, prevalence and antibiogram study of *Staphylococcus aureus* in poultry. *Asian Pac J Trop Biomed.* 2017; 7(3): 253-256.
[10] Van Duijkeren E, Wolfhagen MJ, Box AT, Heck ME, Wannet WJ, Fluit AC. Human-to-dog transmission of methicillin-resistant *Staphylococcus aureus*. *Emer Infect Dis*. 2004; 10(12): 2235.
[11] Witte W. Medical consequences of antibiotic use in agriculture. *Science*, 1998; 279(5353): 996-997.
[12] Kitai S, Shimizu A, Kawano J, Sato E, Nakano C, Uji T, *et al.* Characterization of methicillin-resistant Staphylococcus aureus isolated from retail raw chicken meat in Japan. *J Vet Med Sci.* 2005; 67(1): 107-110.
[13] Otalu OJ, Junaidu K, Chukwudi OE, Jarlath, UV. Multi-drug resistant coagulase positive *Staphylococcus aureus* from live and slaughtered chickens in Zaria, Nigeria. *Int J Poult Sci.* 2011; 10(11): 871-875.
[14] Darwish WS, Atia AS, Reda LM, Elhelaly AE, Thompson LA, Saad Eldin WF. Chicken giblets and wastewater samples as possible sources of methicillin-resistant *Staphylococcus aureus*: Prevalence, enterotoxin production, and antibiotic susceptibility. *J Food Saf.* 2018; 38(4): e12478.
[15] Hossain MJ, Saha SM, Sohidullah M, Alam MA, Kundu SK. Comparative study on food safety knowledge, attitude and practice among street food vendors and consumers. *Int J Nat Sci.* 2021; 11 (2): 40-60.
[16] Habibullah A, Rahman A, Haydar M, Nazir KNH, Rahman MT. Occurrence and molecular detection of methicillin-resistant *Staphylococcus aureus* from dogs and cats in Dhaka City. *Bangla J Vet Med.* 2017; 15: 51-57.
[17] Bauer A, Kirby W, Sherris JC, Turck M. Antibiotic susceptibility testing by a standardized single disk method. *American J Clin Patho.* 1996; 45: 493-496.
[18] Clinical and Laboratory Standards Institute. *Performance standards for antimicrobial susceptibility testing*; 2017. CLSI supplement M100.
[19] Begum F, Islam M, Sohidullah M; Kabir SML, Islam M, Rahman MT. Molecular identification and antibiogram profiles *of Escherichia coli* isolated from apparently healthy and diarrheic goats. *Bangla J Vet Med.* 2016; 14: 203-208.
[20] Bakheet AA, Darwish SF. Prevelence of Methicillin-Resistant *Staphylococcus aureus* (MRSA) in Layer Chickens with Reference to its Ability to Form Biofilm and Enterotoxins. *Alexa J Vet Sci.* 2019; 63(2): 23-32.
[21] Kalorey DR, Shanmugam Y, Kurkure NV, Chousalkar KK, Barbuddhe SB. PCR-based detection of genes encoding virulence determinants in *Staphylococcus aureus* from bovine subclinical mastitis cases. *J Vet Sci.* 2007; 8(2): 151-154. DOI: 10.4142/jvs.2007.8.2.151.
[22] Benrabia I, Hamdi TM, Shehata AA, Neubauer H, Wareth G. Methicillin-resistant *Staphylococcus aureus* (MRSA) in poultry species in Algeria: Long-term study on prevalence and antimicrobial resistance. *Vet Sci.* 2020; 7(2): 54.
[23] Parvin MS, Ali MY, Talukder S, Nahar A, Chowdhury EH, Rahman MT, *et al.* Prevalence and multidrug resistance pattern of methicillin resistant *S. aureus* isolated from frozen chicken meat in Bangladesh. *Microorganisms* 2021; 9(3): 636.
[24] Aklilu E, Nurhardy AD, Mokhtar A, Zahirul IK, Rokiah AS. Molecular detection of methicillin-resistant *Staphylococcus aureus* (MRSA) and methicillin-resistant *Staphylococcus epidermidis* (MRSE) isolates in raw chicken meat. *Int Food Res J.* 2016; 23(1): 322.
[25] Kwoji ID, Jauro S, Musa JA, Lekko YM, Salihu SI, Danchuwa, HA. Phenotypic detection of methicillin-resistant *Staphylococcus aureus* in village chickens from poultry markets in Maiduguri, Nigeria. *J Adv Vet Ani Res.* 2019; 6(2): 163.
[26] da Silva LS, Andrade YM, Oliveira AC, Cunha BC, Oliveira EG, Cunha TS, *et al.* Prevalence of methicillin-resistant *Staphylococcus aureus* colonization among healthcare workers at a tertiary care hospital in northeastern Brazil. *Infec Pre Prac.* 2020; 2(4): 100084.
[27] Hossain MJ, Attia Y, Ballah FM, Islam MS, Sobur MA, Islam MA, *et al.* Zoonotic significance and antimicrobial resistance in Salmonella in poultry in Bangladesh for the period of 2011–2021. *Zoonotic Dis*. 2021; 1(1): 3-24.
[28] Hossain MJ, Islam MS, Sobur MA, Zaman SB, Nahar A, Rahman M, *et al.* Exploring Poultry Farm Environment for Antibiotic Resistant *Escherichia coli*, *Salmonella* spp., and *Staphylococcus* spp. Having Public Health Significance. *J Bangla Agri Univ.* 2020; 18(3): 615–622.
[29] El-Adawy H, Ahmed M, Hotzel H, Monecke S, Schulz J, Hartung J, *et al.* Characterization of methicillin-resistant *Staphylococcus aureus* isolated from healthy turkeys and broilers using DNA microarrays. *Front Microb.* 2016; 7: 2019.
[30] Ugwu MC, Omanukwue C, Chimezie C, Okezie U, Ejikeugwu CP, Nnnabuife-Iloh E, *et al.* Poultry farm and poultry products as sources of multiple antimicrobial-resistant *Salmonella* and *S. aureus*. *J Trop Dis.* 2019; 7(3): 1-23.
[31] Sallam KI, Abd-Elghany SM, Elhadidy M, Tamura T. Molecular characterization and antimicrobial resistance profile of methicillin-resistant *Staphylococcus aureus* in retail chicken. *J Food Prot.* 2015; 78(10):1879-1884.
[32] Tang J, Hu J, Kang L, Deng Z, Wu J, Pan J. The use of vancomycin in the treatment of adult patients with methicillin-resistant *Staphylococcus aureus* (MRSA) infection: a survey in a tertiary hospital in China. *Int J Clin Experi Med.* 2015; 8(10): 19436.
[33] Cong Y, Yang S, Rao, X. Vancomycin resistant *Staphylococcus aureus* infections: A review of case updating and clinical features. *J Adv Res.* 2020; 21: 169-176. DOI: 10.1016/j.jare.2019.10.005.
[34] Shahid AH, Nazir KNH, El Zowalaty ME, Kabir A, Sarker SA, Siddique MP, *et al.* Molecular detection of vancomycin and methicillin resistance in *Staphylococcus aureus* isolated from food processing environments. *One Health* 2021; 13: 100276.
[35] Feßler AT, Kadlec K, Hassel M, Hauschild T, Eidam C, Ehricht R, *et al.* Characterization of methicillin-resistant *Staphylococcus aureus* isolates from food and food products of poultry origin in Germany. *Appl Environ Microb.* 2010; 77(20): 7151-7157.