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Original Research Article

Phenotypic and genotypic characterization of methicillin resistant *Staphylococcus aureus* isolated from different sources

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ABSTRACT

Food contaminated with multiple antibiotic-resistant *S. aureus* can be a major threat to the public health. The purpose of this study was to isolate *S. aureus* from different food sources, determine their antimicrobial susceptibility as well as detection of *mec*A gene among some resistant isolates. Out of 125 from food of animal origin samples, 19 *S. aureus* isolates were recovered, and the antimicrobial susceptibility testing showed a high resistance against kanamycin, penicillin G, oxacillin, erythromycin and tetracycline. All the tested isolates were multiple drug resistant (MDR). Eight out of 19 (15.2%) isolates were phenotypically resistant to oxacillin as well as they were carriers for *mec*A gene.

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1. Introduction

Antibiotics were extensively used in animals and poultry production for prophylaxis, therapeutic and growth promoter purposes, the correlation between extensive use of antimicrobials and development of resistant bacteria is well authenticated for pathogenic bacteria (Hawkey and Jones, 2009). Food of animal origin contaminated with antibiotic-resistant bacteria can be an important threat to public health, the antibiotic resistance determinants can be transferred from resistant bacteria to other bacteria of human public health significance. The prevalence of antimicrobial resistance among food-borne pathogens has elevated during recent decades (Threlfall et al., 2000; Chiu et al., 2002).

Molecular analysis of antibiotic-resistant genes has shown that identical elements founded in bacteria that affect both animals and humans, which explain the role of raw foods in the dissemination of resistance genes and resistant bacteria to humans via the food chain (O'Brien et al., 1982; Teuber, 2001). Recently, many studies were undertaken to assess the antibiotic resistance of bacteria in food of animal origin such as raw milk (Munsch-Alatossava and Alatossava, 2007) and meat products (White et al., 2001). These studies reported that a significant proportion of the isolates from the food products demonstrated extensive resistance to antibiotics. The resistance genes can be transfered from resistant bacteria to the intestinal flora of humans through food products and the commensal flora could act as a reservoir for resistance genes for pathogenic bacteria (Aarestrup et al., 2008).

Multiple studies have discovered the high prevalence of MDR *S. aureus*, including methicillin-resistant *S. aureus* (MRSA) from food of animal origin in Europe, Canada, and United States (Khanna et al., 2008; Smith et al., 2010), which represents a huge problem in public health (Morosini et al., 2006). Most of MRSA isolates carried mecA gene which is responsible for methicillin resistance (El-Jakee et al., 2011)

Therefore, the aim of this study was to detect *S.aureus* from different food sources, determine their antimicrobial susceptibility as well as detection of *mec*A gene among some MDR isolates.

2. Materials and methods

Samples

One hundred and twenty five samples were randomly collected from milk, meat and their products from Giza and Beni-Suef Governorates markets (Table 1). Samples were aseptically collected in sterile plastic bags separately and transferred immediately under hygienic measures in ice box to the laboratory to be examined for presence of *S. aureus*.

Isolation and identification of *S.aureus*

One loopfull from the incubated samples was plated onto (Difco) mannitol salt agar (Difco), incubated for 18-24 hrs at 37°C and examined for bacterial growth. The suspected colonies were identified morphologically and biochemically according to Cruickshank et al. (1975) and Quinn et al. (2002).

Antimicrobial sensitivity testing of the identified isolates

S. aureus isolates were tested against 10 antimicrobial agents representing different classes of antimicrobials by the disk diffusion method and evaluated according to Clinical and Laboratory Standards Institute (CLSI, 2013) (Table 2).

Phenotypic characterization of methicillin resistant *S. aureus*

Disc diffusion sensitivity testing of *S. aureus* isolates was performed with 1µg oxacillin discs. On Mueller Hinton Agar, according to CLSI recommendations, oxacillin complete inhibition zone diameter of ≤12 mm was considered resistant and those with inhibition zone of ≥13 mm were susceptible.

Genotypic characterization of MRSA

S. aureus isolates were inoculated on Triptycase Soya Agar. After incubation period, fresh colonies were suspended in 500 µl sterile saline. DNA was extracted from the suspension using a QIAamp DNA Mini Kit according to the manufacturer's instructions (Qiagen).

Detection of *mec*A gene using PCR

The Polymerase chain reaction was performed for detection of *mec*A gene using primers previously described by McClure et al. (2006). Briefly, 310 bp gene was amplified using primers mecA-F (5'-GTA GAA ATG ACT GAA CGT CCG ATA A-3') and mecA-R (5'-CCA ATT CCA CAT TGT TTC GGT CTA A-3'). Polymerase chain reaction contained 6 µl of DNA template, 12.5 µl Emerald Amp GT PCR

master mix (2x premix), 1 µl from each primer (20pmol) and finally 4.5 µl PCR grade water.

The following temperature profile was used for DNA amplification: initial denaturation at 94°C for 5 min followed by 35 cycles of amplification (denaturation at 94°C for 30 sec, annealing at 50°C for 45 sec and extension at 72°C for 45 sec) and final extension at 72°C for 10 min. PCR amplifications were performed using T3 Thermal cycler (Biometra). The PCR products were visualized by 1 × TBE electrophoresis in ethidium bromide-stained, 1% agarose gel.

Positive control

Confirmed positive sample in RLQP (Reference laboratory for veterinary quality control on poultry production, Dokki, Giza).

3. Results

Results of *S.aureus* isolation

Seventy two Gram positive cocci were recovered out of 125 food samples. Out of them, 19 isolates (15.2%) were coagulase positive also fermented maltose, trehalose, mannitol and sucrose so; they were characterized biochemically as *S. aureus* (Table 3).

Results of antimicrobial sensitivity

The results revealed that all *S. aureus* isolates were sensitive (100%) to apramycin, trimethoprim-sulfamethoxazole, gentamicin and cefazolin. On the other hand, they were resistant to amoxicillin, kanamycin erythromycin, tetracycline, penicillin G and oxacillin, with a percentage of 31.6%, 42.1%, 36.3%, 57.8% 73.6% and 84.2%, respectively (Table 4). All tested isolates are multiple drug resistant (MDR).

Phenotypic detection of MRSA among the tested isolates

Out of 19 *S. aureus* isolates 16 isolates were phenotypically resistant to oxacillin disk so they were characterized phenotypically as MRSA with a percentage of 84.2%.

Genotypic characterization MRSA among the tested isolates

Eight oxacillin resistant isolates were selected for investigation using PCR to amplify *mec*A gene. All the selected isolates (100%) were positive at an amplicon size of 310 pb (Fig. 1) and confirmed as MRSA.

Table 1. Samples collected from sale markets.

Product	Milk	Yoghurt	Kareesh	Minced meat	Burger	luncheon
Numbers	28	18	19	20	20	20

Table 2. Antimicrobial agents used against isolated *S. aureus*.

Antimicrobial Agent	Code	Disk Concentration
Apramycin	APR	15 µg
Cefazoline	KZ	30µcg
Amoxicillin	AML	10 µg
Erythromycin	E	15µg
Trimethoprim + sulfamethoxazole	SXT	25 µg
Gentamicin	CN	10 µg
Kanamycin	K	30 µg
Vancomycin	VA	30 µg
Penicillin	P	10units
Oxacillin	OX	1 µg
Tetracycline	TE	30 µg

Table 3. Prevalence of the isolated *S. aureus* among different food products.

Source of samples	Total number of samples examined	Recovered *S. aureus* isolates	
		Number	Percentage
Milk	28	8	28.5
Yogurt	19	1	5.2
Kareish cheese	18	0	0
Total milk and milk products	65	9	13.8
Minced meat	20	5	25
Burger	20	2	10
Luncheon	20	3	15
Total meat and meat products	60	10	16.6
Grand total	125	19	15.2

Table 4. Antimicrobial sensitivity of 19 *S.aureus* isolates recovered from raw milk, meat and their products.

Antibacterial agent	Milk and milk products (*n*=9)						Meat and meat products (*n*=10)					
	Sensitive		Intermediate		Resistant		Sensitive		Intermediate		Resistant	
	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
Penicillin groups												
Penicillin	2	22.2	0	-	7	77.7	3	30	0	-	7	70
Oxacillin	1	11	0	-	8	88.8	1	10	1	10	8	80
Amoxicillin	3	33.3	2	22.2	4	44.5	7	70	1	10	2	20
Glycopeptides group												
Vancomycin	8	88.8	0	-	1	11	9	90	0	-	1	10
Aminoglycoside group												
Gentamicin	9	100	0	-	0	0	10	100	0	-	0	-
Kanamycin	2	22.2	2	22.2	5	55.5	4	40	3	30	3	30
Apramycin	6	66.6	3	33.3	0	0	9	90	1	10	0	-
Macrolide group												
Erythromycin	6	66.6	1	11	2	22.2	5	50	2	20	3	30
Tetracycline group												
Tetracycline	2	22.2	0	-	7	77.7	3	30	3	30	4	40
Folate pathway inhibitor												
Sulfamethaxazole+Trimethoprime	9	100	0	-	0	0	10	100	0	-	0	-
Cephalosporin group												
Cefazolin (first generation)	9	100	0	-	0	0	10	100	0	-	0	-

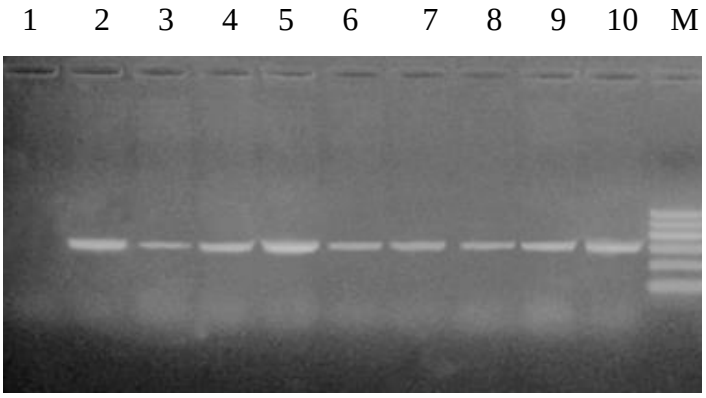

Fig. 1. Amplification of *mec* A gene at amplicon size of 310 pb. Lane 1: Negative control. Lane 2-9: Positive amplification of *mec*A gene at 310 bp of the tested of MRSA isolates. Lane 10: Positive control. Lane M : DNA ladder (100-600 bp).

4. Discussion

Food of animal origin is an ideal culture medium for growth of many organisms (Hill, 1996). They are liable to harbor different types of microorganisms through processing, handling, distribution and storage as well as preparation. They are considered as serious sources of food borne diseases and have been linked to major outbreaks of food poisoning worldwide (Hassanien, 2004).

It has been revealed that the isolation of *S. aureus* from raw milk and milk products (Kareish cheese and yoghurt) and meat and meat products (burger and luncheon) with a percentage of 13.8% and 16.6% respectively (Table 3). Nearer percentage was recorded by El-Jakee et al. (2008) and Song et al. (2014). Higher results for *S.aureus* contamination in raw milk was previously reported (El-jakee et al., 2013; Gwida and El Ghary, 2013). Occurrence of *S. aureus* in milk is variable and this may be due to variation in season, geographical location, number of animals in the farm, farm size, hygienic measures, sampling, farm management practices, and differences in the detection methods. El Sayed et al. (2011) stated that the difference in white soft cheese in Egypt due to acidity as Domiati or Kareish acid coagulation, enzyme coagulation , different salt concentrations, keeping temperatures, ripening in brine solutions are factors affecting the microbiological quality of these varieties .

In the current study, the incidence of *S. aureus* in meat products (16.6%) was in agreement with Pesavento et al. (2007), Hanson et al. (2011) and Abdaslam et al. (2014). Ali et al. (2010) isolated *S. aureus* from meat sample with a percentage of 7% lower than our results. Hassanin (2007), Song et al. (2014) and Li et al. (2015) found *S. aureus* with a percentage higher than our results

Antibiotic-resistant *S. aureus* strains can be transmitted by contaminated foods with resistant bacteria and spread between animals and humans (Gundogan et al., 2006). There is a relationship between the prevalence of antibiotic resistance and the consumption of antibiotic agents. Some researchers reported a primary factor for antibiotic resistant bacteria in food is related to using of antibiotics for therapeutic purposes in animals (Al-Zu'Bi et al., 2004). The studied *S. aureus* isolates were sensitive (100%) to apramycin, trimethoprime+sulfamethoxazole, gentamicin and cefazolin. On the other hand, they were resistant to amoxicillin, erythromycin, kanamycin, tetracycline, penicillin G, oxacillin with a percentage of 31.6%, 36.3%, 42.1%, 57.8%, 73.6% and 84.2%, respectively (Table 4) as well as a high prevalence of multidrug resistance. Jamali et al. (2015) stated that *S. aureus* were resistant to tetracycline (56.1%) and gentamicin (2.1%) but lower incidence for penicillin G, erythromycin, streptomycin, kanamycin and oxacillin. The high prevalence of MDR *S. aureus* isolates in our study was in agreement with those

given by Albuquerque et al. (2007), Haran et al. (2012) and Tan et al. (2014). The same results obtained by Argudín et al. (2011) detected resistant *S. aureus* resistant to oxacillin (95%) but differ in erythromycin (70%), tetracycline (100%), gentamicin (14%) and kanamycin (29%). Gundogan et al. (2011) reported that 10.5% of *S.aureus* isolates were resistant to vancomycin. Hanson et al. (2011) reported the same results but with lower incidence in oxaciliin resistant in *S. aureus* isolated from meat.

The accurate and rapid detection of antibiotic resistance genes is extremely important in preventing the spread of infections. PCR-based molecular methods are preferred for determination of antibiotic resistance genes. During the last 10 years, many studies have demonstrated the extremely high capacity of molecular methods such as PCR and pulsed-field gel electrophoresis (PFGE), this methods were increasingly used for their rapid, specific, reliable and accurate detection of bacteria and genes of interest (Laplana et al., 2007). Recently, the detection of antibiotic resistance genes was accomplished by PCR methods directed to *mec*A gene (Al-Zu'Bi et al., 2004).

MRSA as one of the most common pathogens that cause nosocomial infection worldwide (Al-Zu'Bi et al., 2004). Since the first identification of an MRSA isolate in 1960 in UK (Donnio et al., 2004). MRSA described the first foodborne outbreak of that caused death of five out of twenty-one patients (Kluytmans et al., 1995). MRSA has recently emerged as a health concern and currently causes approximately 94,000 invasive infections yearly in the United States of America, leading to an estimated 18,650 deaths (Klevens et al., 2007).

The present results revealed that all isolates of *S. aureus* which were resistant to oxacillin were carrier for mecA gene. The results agree with those reported by Shahraz et al. (2012) and Jamali et al. (2015). A lower prevalence of *mec*A gene in food of animal origin was detected by Normanno et al. (2007). Lee (2003) stated that contaminated foods of animal origin may represent a source of MRSA infection for humans.

It can be concluded that *S. aureus* contaminated milk and meat under study as well as their products in noticeable percentage. Worrisome most of the recovered isolates are MDR and MRSA and they could be transmitted to human being representing public health hazard.

References

Albuquerque W, Macrae A, Sousa O, Vieira G, Vieira R (2007). Multiple drug resistant *Staphylococcus aureus* strains isolated from a fish market and from fish handlers. Braz. J. Microbiol., 38: 131–134.

Argudín MA, Mendoza MC, González-Hevia MA, Bances M, Guerra B, Rodicioa MR (2012). Genotypes, exotoxin gene content, and antimicrobial resistance of *Staphylococcus aureus* strains recovered from foods and food handlers. Appl. Environ. Microbiol., 78(8):2930–2935.

Aarestrup FM, Wegener HC, Collignon P (2008). Resistance in bacteria of the food chain: Epidemiology and control strategies. Expert. Rev. Anti. Infect. Ther., 6(5):733–750.

Abdaslam SA, Hassan MA, Kaheel HH, Abobaker TM, AlnourainTH, Hamdan HA, Shankar SG, Thambirajah JJ (2014). Isolation of *Escherichia coli* O157 and other food borne pathogens from meat products and their susceptibility to different antimicrobial agents. Curr. Res. Microbiol. Biotechnol., 2(3): 391–397.

Ali NH, Farooqui A, Khan A, Khan AY, Kazmi SU (2010). Microbial contamination of raw meat and its environment in retail shops in Karachi, Pakistan. J. Infect. Dev. Ctries., 4(6): 382–388.

Al-Zu'Bi E, Bdour S, Shehabi AA (2004). Antibiotic resistance patterns of mecA positive *Staphylococcus aureus* isolates from clinical specimens and nasal carriage. Microb. Drug Resist., 10(4), 321–324.

Chiu CH, Wu TL, Su LH, Chu C, Chia JH, Kuo AJ, Chien MS, Lin TY (2002). The emergence in Taiwan of fluoroquinolone resistance in *Salmonella enterica* serotype Choleraesuis. N. Engl. J. Med., 346: 413–419.

Cruickshank R, Mar Mien BP, Swain RHA (1975). Medical Microbiology. 12th ed. Churchill. Livingstone. Edinburgh, London, New York.

Clinical and Laboratory Standards Institute (CLSI) (2013): Approved standards: Performance Standards for Antimicrobial Disk Susceptibility Tests; Approved Standard—Eleventh Edition.

Donnio PY, Preney L, Gautier-Lerestif AL, Avril JL, Lafforgue N (2004). Changes in staphylococcal cassette chromosome type and antibiotic resistance profile in methicillin-resistant *Staphylococcus aureus* isolates from a

French hospital over an 11 year period. J. Antimicrob. Chemother., 53(5): 808–813.

Duran N, Ozer B, Duran GGl, Onlen Y, Demir C (2012). Antibiotic resistance genes & susceptibility patterns in staphylococci. Indian J. Med. Res., 135(3): 389–396.

El-Jakee J, Ata SN, Bakry M, Zouelfakar SA, Elgabry, E, Gad El-Said WA (2008). Characteristics of Staphylococcus aureus Strains Isolated from Human and Animal Sources. American-Eurasian J. Agric. Environ. Sci., 4 (2): 221–229.

El-Jakee JK, Ata SN, Samy AA, Bakry MA, Elgabry EA, Mai MK, Gad El-Said WA (2011). Antimicrobial resistance in clinical isolates of *Staphylococcus aureus* from bovine and human sources in Egypt .Global Vet., 7: 581–586.

EL-Jakee J, Marouf SA, Ata SN, Abdel-Rahman HE, Abd ELMoez S, Samy AA, ELSayed E, Walaa E (2013). Rapid method for detection of *Staphylococcus aureus* enterotoxins in food. Global Vet., 11(3): 335–341.

El Sayed MA, Hosny IM, El Kholy WI, El Dairouty RK, Mohamed SHS (2011). Microbiological evaluation of Egyptian white soft cheeses style. J. Am. Sci., 7(5): 517–526.

Gundogan N, Citak S, Turan E (2006). Slime production, DNase activity and antibiotic resistance of *Staphylococcus aureus* isolated from raw milk, pasteurized milk and ice cream samples. Food Control J. 17(5): 389–392.

Gwida MM, EL-Gohary FA (2013). Zoonotic bacterial pathogens isolated from raw milk with special reference to *Escherichia coli* and *Staphylococcus aureus* in Dakahlia Governorate, Egypt. Open Access Sci. Reports. 2(4): 705–708.

Hawkey PM, Jones AM (2009). The changing epidemiology of resistance. J. Antimicrob. Chemother., 64 (Suppl 1): i3–i10.

Hassanien FS (2004). Bacterial hazards associated with consumption of some meat products. Benha Vet. Med. J., 15: 41–54.

Hassanin ZH (2007). Studies on food poisoning microorganisms in some meat products. M. V. Sc. Thesis, Fac. Vet. Med., Menoufia University, Sadat branch.

Hanson BM, Dressler AE, Harper AL, Scheibel RP, Wardyn SE, Roberts LK, Kroeger JS, Smith TC (2011). Prevalence of *Staphylococcus aureus* and methicillin-resistant *Staphylococcus aureus* (MRSA) on retail meat in Iowa. J. Infect. Public Health 4: 169–174.

Haran K, Godden S, Boxrud D, Jawahir S, Bender J, Sreevatsan S (2012). Prevalence and characterization of *Staphylococcus aureus*, including methicillin resistant *Staphylococcus aureus*, isolated from bulk tank milk from Minnesota dairy farms. J. Clin. Microbiol., 50(3): 688–695.

Hill WE (1996). The polymerase chain reaction: applications for the detection of foodborne pathogens. Crit. Rev. Food Sci. Nutr., 36(1-2):123–173.

Jamali H, Paydar M, Radmehr B, Ismail S, Dadrasnia A (2015). Prevalence and antimicrobial resistance of *Staphylococcus aureus* isolated from raw milk and dairy products. Food Control J., 54: 383–388.

Klevens RM, Morrison MA, Nadle J, Petit S, Gershman K, Ray S, Harrison LH, Lynfield R, Dumyati G, Townes JM, Craig AS, Zell ER, Fosheim GE, McDougal LK, Carey RB, Fridkin SK, Active Bacterial Core surveillance (ABCs) MRSA Investigators (2007). Invasive methicillin-resistant *Staphylococcus aureus* infections in the United States. JAMA, 298:1763–1771.

Khanna T, Friendship R, Dewey C, Weese JS (2008). Methicillin resistant *Staphylococcus aureus* colonization in pigs and pig farmers. Vet. Microbiol., 128: 298–303.

Kluytmans J, van Leeuwen W, Goessens W, Hollis R, Messer S, Herwarld L, Bruining H, Heck M, Rost J, van Leeuwen N, van Belkum A, Verbrugh H (1995): Food-initiated outbreak of methicillin resistant *Staphylococcus aureus* analyzed by pheno- and genotyping. J. Clin. Microbiol., 33: 1121–1128.

Laplana LM, Cepero MA, Ruiz J, Zolezzi PC, Calvo MA, Erazo MC, Gómez-Lus R **(2007):** Molecular typing of *Staphylococcus aureus* clinical isolates by pulsed-field gel electrophoresis, staphylococcal cassette chromosome mec type determination and dissemination of antibiotic resistance genes. Int. J. Antimicrob. Agents 30(6): 505–513.

Lee JH (2003). Methicillin (oxacillin)-resistant *Staphylococcus aureus* strains isolated from major food animals and their potential transmission to humans. Appl. Environ. Microbiol., 69(11): 6489–6494.

Li G, Wu C, Wang X, Meng J (2015). Prevalence and characterization of methicillin susceptible Staphylococcus aureus ST398 isolates from retail foods. Int. J. Food Microbiol., 196: 94–97.

McClure JA, Conly JM, Lau V, Elsayed S, Louie T, Hutchins W, Zhang K (2006). Novel multiplex PCR assay for detection of the staphylococcal virulence marker Panton-Valentine leukocidin genes and simultaneous discrimination of methicillin-susceptible from -resistant staphylococci. J. Clin. Microbiol., 44: 1141–1144.

Morosini M-I, García-Castillo M, Coque TM, Valverde A, Novais Â, Loza E, Baquero F, Cantón R (2007). Antibiotic coresistance in extended-spectrum-β-lactamase-producing Enterobacteriaceae and *in vitro* activity of tigecycline. Antimicrob Agents Chemother., 50(8): 2695–2699.

Munsch-Alatossava P, Alatossava T (2007). Antibiotic resistance of raw-milk-associated psychrotrophic bacteria. Microbiol. Res., 162(2):115–123.

Normanno G, Corrente M, La Salandra G, Dambrosio A, Quaglia NC, Parisi A, Greco GA, Bellacicco AL, Virgilio S, Celano GV (2007). Methicillin-resistant *Staphylococcus aureus* (MRSA) in foods of animal origin product in Italy. Int. J. Food Microbiol., 117: 219–222.

O'Brien AM, Hanson BM, Farina SA, Wu JY, Simmering JE, Wardyn SE, Forshey BM, Kulick ME, Wallinga DB, Smith TC (2012). MRSA in conventional and alternative retail pork products. PLoS One. 7(1):e30092. doi: 10.1371/journal.pone.0030092

Pesavento G, Ducci B, Comodo N, Lo Nostro A (2007). Antimicrobial resistance profile of *Staphylococcus aureus* isolated from raw meat: A research for methicillin resistant *Staphylococcus aureus* (MRSA). Food Control 18: 196–200.

Quinn PJ, Markey BK, Carter ME, Donnelly WJC, Leonard FC, Maguire D (2002). Veterinary Microbiology and Microbial Diseases.1st published Blackwell Science.

Sambrook X, Fritsch EF, Maniatis T (1989). Molecular cloning a laboratory manual.2nd ed. Cold Spring Harbor Laboratory Press.

Schlegelova J, Vlkova H, Babak V, Holasova M, Jaglic Z, Stosova T, Sauer P (2008). Resistance to erythromycin of *Staphylococcus* spp. isolates from the food chain. Vet. Med., 53(6): 307–314.

Song M, Bai Y, Xu J, Carter QM, Shi C, Shi X (2014). Genetic diversity and virulence potential of *Staphylococcus aureus* isolates from raw and processed food commodities in Shanghai. Int. J. Food Microbiol., 195: 1–8.

Smith TC, Pearson N (2010). The emergence of *Staphylococcus aureus* ST398. Vector Borne Zoonotic Dis., 11(4): 327–339.

Shahraz F, Dadkhah H, Khaksar R, Mahmoudzadeh M, Hossein H, Kamran M, Bourke P (2012). Analysis of antibiotic resistance patterns and detection of mecA gene *Staphylococcus aureus* isolated from packaged hamburger. Meat Sci., 90: 759–763.

Tan SL, Lee HY, Mahyudin NA (2014). Antimicrobial resistance of *Escherichia coli* and *Staphylococcus aureus* isolated from food handler's hands. Food Control J., 44: 203–207.

Threlfall EJ, Ward LR, Frost JA, Willshaw GA (2000). The emergence and spread of antibiotic resistance in food-borne bacteria. Int. J. Food Microbiol., 62:1–5.

Teuber M (2001). Veterinary use and antibiotic resistance. Curr. Opin. Microbiol., 4: 493–499.

White DG, Zhao S, Sudler R, Ayers S, Friedman S, Chen S, McDermott PF, McDermott S, Wagner DD, Meng J (2001). The isolation of antibiotic-resistant *Salmonella* from retail ground meats. N. Engl. J. Med., 345(16):1147–1154.