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RABBIT MEAT AS A POTENTIAL SOURCE OF MULTIDRUG-RESISTANT AND ENTEROTOXIGENIC *STAPHYLOCOCCUS AUREUS* STRAINS

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Abstract
Staphylococcus aureus in rabbit meat is a consequence of insufficient hygienic handling and improper processing posing a major health hazard. This study was conducted to assess rabbit meat as a potential source of Staphylococcus species, particularly Staphylococcus aureus (S. aureus). Furthermore, the identified S. aureus isolates were tested for the detection of the mecA virulence gene of methicillin-resistant Staphylococcus aureus (MRSA) and enterotoxin encoding genes (Sea, Seb, Sec, and Sed). A total of 80 samples of different rabbit meat cuts represented by shoulder, ribs, loin, and thigh (20 of each) were collected from various markets of different sanitation levels. The results obtained revealed that the mean counts of Staphylococcus species were 7.40×10^5, 7.58×10^5, 7.60×10^5 and 8.29×10^5 CFU/g in the examined shoulder, ribs, loin and thigh samples, respectively. Out of 17 identified S. aureus isolates, 5 (29.4%) strains were characterized by the presence of the mecA gene. A large proportion of the isolates obtained were resistant to at least three antibiotics. Enterotoxins were evaluated by ELISA. The results showed that three strains isolated from shoulder produced Sea, Seb, and Sec enterotoxins, the strains isolated from ribs failed to produce enterotoxins, while two strains isolated from loin and thigh produced Sea enterotoxin. The presence of S. aureus, especially MRSA strains, in the examined rabbit meat indicates the necessity of enforced application of strict hygienic measurements.

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Introduction

Consumer awareness of and demand for efficient protein sources have increased in recent decades as consumer understanding of the relationship between nutrition and a healthy lifestyle has developed [1,2]. The rabbit industry has gained much more interest due to the fact that rabbit meat has various advantages, which qualify it as one of the most beneficial healthy foods.

Rabbit meat is a popular culinary product and one of the most consumed meats throughout the world. Its use has recently grown in a number of the Middle Eastern countries, notably Egypt [3–5]. It is recognized as an excellent source of easily digestible animal protein, polyunsaturated fatty acids (PUFAs), vitamins, and minerals (such as calcium, magnesium, and zinc), while being low in fat, sodium, and cholesterol [6]. Rabbit meat, on the other hand, is very prone to deterioration and food poisoning bacteria due to its high protein and moisture content. This has been related to the spread of microbial contamination that may have originated from the animal itself, personnel, equipment, or the environment throughout various stages of slaughter and processing [7,8].

Staphylococcus aureus is a spherical Gram-positive bacterium that is present in nearly one-third of the world population and causes staphylococcal food-borne intoxication, as some of its pathogenic strains are able to produce heat-stable enterotoxins [9]. Staphylococcal food poisoning (SFP) is one of the most prevalent food-borne illnesses in the world. It results from the ingestion of staphylococcal enterotoxins produced by enterotoxigenic strains of coagulase-positive staphylococci in food, mainly *S. aureus* and usually occurs within 30 minutes to 8 hours resulting in several symptoms that include vomiting, nausea, abdominal cramping, diarrhea, chills, and sweating [10]. Staphylococcal food poisoning (SFP) is generally self-limiting and resolves typically within 24–48 h after beginning based on the quantity of contaminated food consumed, the amount of the ingested toxin in food, and the general health of patients [11]. Occasionally, it can be serious enough to warrant hospitalization, especially when it comes to children, the elderly, or debilitated people.

Staphylococcus aureus produces numerous toxins including staphylococcal enterotoxins (SEs; SEA to SEE, SEG to SEI, and SER to SET) with the emetic activity. The detection of SE-encoding genes allows the identification of potentially enterotoxigenic *S. aureus*, regardless of whether a strain produces the toxin or not [12]. SEs are a significant contributor to food poisoning, which typically happens after ingestion of various foods, especially processed meat and dairy products that have been exposed to *S. aureus* through improper handling and subsequent storage at high temperatures [13].

Despite the low prevalence of MRSA in raw food, there is still a chance that it could spread through the food supply, particularly in uncooked meat. In fact, MRSA-related foodborne disease outbreaks have been documented [14]. Moreover, food handlers who handle contaminated food may also be at risk for health problems. Foods can be contaminated during processing by MRSA-colonized food handlers, and carcasses from MRSA-infected animals can become contaminated during slaughter [15].

The incidence of antibiotic resistance in food-associated pathogenic bacteria, including *S. aureus*, has been a growing issue over the last few decades due to the intensive use of antibiotics in public health and animal husbandry, and the risk of transfer of antibiotic resistance determinants [16]. Lack of adequate hygienic measures during food preparation is one of the main causes of contamination as food handlers themselves can carry the pathogenic bacterium. Besides that, *S. aureus* can withstand a broad variety of temperature, pH, and salinity [17]. In addition, most of the nosocomial *S. aureus* infections are triggered by methicillin-resistant *S. aureus* (MRSA) strains and have become a world-wide recognized cause of morbidity and mortality [18]. Methicillin-resistant *S. aureus* (MRSA) strains that are resistant to quinolones or multi-resistant to other antibiotics have emerged, which leaves a restricted option for their control [19]. Therefore, the current study was designed to determine the incidence of enterotoxigenic and methicillin-resistant *S. aureus* (MRSA) strains in rabbit meat cuts (shoulder, ribs, loin, and thigh) retailed in Zagazig city, Sharkia governorate, Egypt, as well as to investigate the antimicrobial susceptibility profile and the major staphylococcal enterotoxins (SEs) among the isolated *S. aureus* strains.

Objects and methods

Samples collection and preparation

The objects of the study were the rabbit meat samples from Zagazig City, Sharkia province, Egypt. Eighty random samples of rabbit meat (20 each of shoulder, ribs, loin, and thigh) were collected from various locations with different levels of sanitation. All collected samples were promptly transferred in an icebox under complete aseptic conditions to the laboratory for bacteriological examination without delay. Twenty-five grams of each rabbit meat cut were homogenized aseptically for 1 min with 225 mL of 0.1% sterile buffered peptone water (HIMEDIA, M614, Mumbai, India) in a stomacher (Colworth, model 400, UK) to prepare a homogenate of 10^{-1} (as an initial dilution) and allowed to stay for 5 min. Quantity of 1 ml of the homogenate was transferred into a sterile test tube containing 9 ml of 0.1% BPW and then serially diluted tenfold in the same diluent [20].

Determination of Staphylococcus count

The *Staphylococcus* species count in the samples was determined through bacteriological analysis using Baird Parker agar (BP) supplemented with egg yolk tellurite emulsion following ISO 6888–1:2021 with slight modifications [21]. Briefly, 0.1 mL from each prepared dilution was spread onto duplicate plates of Baird-Parker agar (HIMEDIA, M043–100G, Mumbai, India) supplemented with egg yolk tellurite emulsion (50 mL/L, Oxoid SR54, UK) and then incubated at 37 °C for 24–48 hours. After incubation, presumptive colonies (black, shiny, convex, 1–1.5 mm in diameter, surrounded by a clear halo zone) and/or atypical colonies (black colonies with no zones) were observed and counted. The grown colonies were subsequently confirmed as *Staphylococcus* and identified as belonging to the genus *Staphylococcus* through gram staining, as they typically appear as gram-positive cocci arranged in clusters.

Isolation and identification of Staphylococcus aureus

For *S. aureus* isolation, up to five suspected colonies were picked up and cultured on slope agar for further identification. Isolated purified strains were morphologically identified using Gram's stain and further confirmed as *S. aureus* by biochemical tests (catalase, mannitol fermentation, coagulase, and DNase tests) according to MacFaddin [22].

Genomic DNA Extraction and PCR Analysis

Genomic DNA extraction from each coagulase-positive *S. aureus* isolate was performed using the QIAamp DNA kit (Qiagen, Germany, GmbH) following the manufacturer's instructions. Identification of coagulase-positive isolates was carried out through a species-specific PCR assay. The PCR analysis included the detection of species-specific (nuc) and methicillin resistance (mecA) genes in *S. aureus* isolates. The oligonucleotide primer sequences (Applichem GmbH, Germany) used in PCR reactions for the amplification of the target genes of *S. aureus* and the sizes of amplified products are detailed in Table 1. The DNA amplification process was carried out using a Thermal Cycler (Master cycler, Eppendorf, Hamburg, Germany). PCR protocols for both (nuc and mecA) virulence genes were carried out according to Cho et al. [23]. Amplified DNA fragments were analyzed using 1.5% agarose gel electrophoresis in 1X TBE buffer stained with ethidium bromide (Applichem, Germany, GmbH), captured, and visualized using a UV transilluminator.

Detection and typing of staphylococcal enterotoxins

The incidence of enterotoxins was evaluated by ELISA. According to Shingaki et al. [24], the clear culture supernatant fluid was tested serologically by Reverse Passive Latex Agglutination technique (RPLA) using kits for the detection of staphylococcal enterotoxins A, B, C and D (SET-RPLA, Denka Sekeu LTD, Japan). The sensitivity of this test kit in detecting enterotoxins is 0.5 ng/ml of test extract. The test was conducted in a V-type microtiter plate, with each row containing 8 wells. Each test sample required the use of 5 rows of wells. Initially, 25 µl of diluent was dispensed into each well using a micropipette. Then, the sample was mixed simultaneously with 5 diluents (25 µl each). Two-fold dilutions of the test sample were performed across the 5 rows, with the last well in each row containing only 25 µl of diluent. Quantities of 25 µl of latex suspensions sensitized separately with anti-enterotoxin A, B, C, and D were added to the wells of the 1st, 2nd, 3rd, and 4th rows of the plate, respectively. Additionally, 25 µl of control latex was added to each well in the fifth row, followed by thorough mixing. The plate was covered and left undisturbed at room temperature for 24 hours. Subsequently, each well in every row was examined for agglutination.

Table 1. Oligonucleotide primers of PCR reactions for the amplification of the target genes of *S. aureus*

Target gene	Oligonucleotide sequence (5′ → 3′)	Product size (bp)	References
nuc (F)	5′ GCGATTGATGGTGATACGGTT ′3	270	[25]
nuc (R)	5′ AGCCAAGCCTTGACGAACTAAAGC ′3		
mecA (F)	5′ TAGAAATGACTGAAC GTCCG ′3	533	[26]
mecA (R)	5′ TTGCGATCA ATGTTACCGTAG ′3		

nuc: thermonuclease and mecA: methicillin-resistant *S. aureus* (MRSA) virulence genes.

Demonstration of antimicrobial susceptibility profile of S. aureus isolates

Antibiotic susceptibility testing of *S. aureus* strains was performed using a single diffusion assay against 16 antibiotic discs of varying concentrations [27]. The antimicrobial discs, such as kanamycin (K), cephalexin (CE), oxacillin (OX), penicillin G (P), tetracycline (T), nalidixic acid (NA), cephalothin (CN), ampicillin (AM), sulphamethoxazole (SXT), cefotaxime (CF), clindamycin (CL), erythromycin (E), ciprofloxacin (CP), gentamicin (G), linezolid (LZ), and amikacin (AK), were used to perform antibiogram analysis. Each strain was streaked on Mueller–Hinton agar (Himedia, Mumbai, India), and drug-impregnated discs were placed on the agar medium surface.

The Multiple Antibiotic Resistance (MAR) index was calculated using the formula: MAR index = a ÷ b, where (a) represents the number of antibiotics, to which the isolates were resistant, and (b) is the total number of tested antibiotics.

Statistical analysis

Data were analyzed with one-way ANOVA test using the Statistical Package for Social Sciences software for Windows (SPSS-14; Chicago, IL, USA) using post hoc tukey-kramer honestly correction to estimate the differences in microbial counts. *P*-value of <0.05 was considered statistically significant.

Results and discussion

Prevalence and count of Staphylococcus species in the examined rabbit meat samples

As shown in Table 2, the prevalence and mean *Staphylococcus* count in the examined samples were recorded. All investigated rabbit meat cuts (shoulder, ribs, loin, and thigh) were positive for *Staphylococcus* (100%). *Staphylococcus* species count of shoulder samples ranged from 2.3×10^4 to 2.0×10^6 with a mean value of $7.40\times10^5\pm1.21\times10^5$ CFU/g, while ribs samples recorded *Staphylococcus* count varied from 3.2×10^4 to 1.6×10^6 with a mean count of $7.58\times10^5\pm0.83\times10^5$ CFU/g. Besides, loin samples and thigh samples had *Staphylococcus* count of $7.60\times10^5\pm0.82\times10^5$ and $8.29\times10^5\pm0.85\times10^5$ CFU/g, respectively.

Table 2. *Staphylococcus* species count (CFU/g) and prevalence in the examined rabbit meat samples

Samples	Positive samples	*Staphylococcus* species count (CFU/g)		
	No (%)	Minimum	Maximum	Mean ± SE
Shoulder	20 (100%)	2.3×10^4	2.0×10^6	$7.40\times10^5\pm1.21\times10^5$
Ribs	20 (100%)	3.2×10^4	1.6×10^6	$7.58\times10^5\pm0.83\times10^5$
Loin	20 (100%)	2.5×10^4	1.3×10^6	$7.60\times10^5\pm0.82\times10^5$
Thigh	20 (100%)	3.2×10^4	1.4×10^6	$8.29\times10^5\pm0.85\times10^5$

Means are not significantly different at *P*> 0.05; No (%): number and percentage of positive samples; CFU/g: Colony Forming Units per gram.

Rabbit meat and offal are unique sources of high-quality animal protein that also have a high nutritive value for other nutrients. However, rabbit meat is also regarded as a potential source of spoilage and food poisoning organisms, which can cause a variety of negative health effects and shorten the shelf life of rabbit meat [4]. *Staphylococcus* contamination of food results from inadequate hygienic handling and processing, which could be hazardous to human health [28]. Regarding *Staphylococcus* count, Morshdy et al. [8] recorded an initially higher count of 1.34×10^4 CFU/g in freshly untreated rabbit meat samples from Egypt. However, lower results of *Staphylococcus* count were reported by Khalafalla [29] in freshly slaughtered and processed rabbit samples obtained from grocery stores in Beni-Suef city, Egypt, with mean values of 10^2 and 4×10^3 CFU/g, respectively. The high counts of staphylococci could be associated with improper personal hygiene of untrained employees and cross contamination from skin and utensils.

As concerns the incidence of *S. aureus*, 17 (85%) out of 20 *Staphylococcus* isolates in the present study were serologically identified as *S. aureus*. Lower results were obtained by

Kpodékon et al. [30] who detected staphylococci isolated from 30 frozen rabbit carcasses in Benin with a prevalence of 26%, while Kohler et al. [31] documented staphylococci prevalence in rabbit samples from Switzerland with a percentage of 30.6%. Furthermore, Rodriguez-Calleja et al. [32] investigated prevalence of *S. aureus* isolated from rabbit carcasses in Spain with a percentage of 52.9%. Additionally, Bello et al. [33] demonstrated *S. aureus* prevalence from rabbit meat in Nigeria with a percentage of 30.3%. Moreover, Khalafalla [29] isolated *S. aureus* from freshly slaughtered and processed rabbit samples obtained from grocery stores in Beni-Suef, Egypt with a prevalence of 5% and 10%, respectively. The variations of the results may be attributed to how the samples were handled and unsanitary practices observed during data collection. The sharing of environments, facilities, and equipment for the processing of rabbits and poultry, as well as the maintenance of such environments, facilities, and equipment, and the effectiveness of hygienic practices, are critical factors that may have a significant impact on the microbiological profile of the final product [29,34].

Detection of enterotoxigenic and methicillin-resistant S. aureus

The data presented in Table 3 indicate that only five strains were enterotoxigenic. Among six tested shoulder isolates, only one multitoxigenic strain carried three virulence genes (Sea, Seb, and Sec) with each gene accounting for 16.6% (1/6). Similarly, one strain out of five tested loin isolates carried only one gene (Sea gene) with a percentage of 20% (1/5). Furthermore, one strain out of four tested thigh isolates carried only one gene (Sea gene) with a percentage of 25% (1/4). However, *S. aureus* isolates from ribs did not produce any type of enterotoxins.

The results obtained in Figure 1 indicate that all isolates of *S. aureus* were positive for the species-specific (nuc) gene, while the methicillin resistance (mecA) gene was detected in only 5 strains. These strains were classified as methicillin-resistant *S. aureus* (MRSA), accounting for a percentage of 29.4%. This distribution included two isolates from the shoulder (2/6=33.3%), two isolates from the loin (2/5=40%), and one isolate from the thigh (1/4=25%), while the ribs tested negative (Table 3).

Table 3. Incidence of enterotoxins and *mecA* virulence genes among the isolated *S. aureus* strains

Samples	*S. aureus*	SEA	SEB	SEC	SED	mecA
Shoulder	6 (30%)	1 (16.6%)	1(16.6%)	1(16.6%)	0	2 (33.3%)
Ribs	2 (10%)	0	0	0	0	0
Loin	5 (25%)	1 (20%)	0	0	0	2 (40%)
Thigh	4 (20%)	1 (25%)	0	0	0	1 (25%)
Total	17 (85%)	3 (17.6%)	1 (5.8%)	1 (5.8%)	0	5 (29.4%)

SEA: *S. aureus* enterotoxin A; SEB: *S. aureus* enterotoxin B; SEC: *S. aureus* enterotoxin C; and SED: *S. aureus* enterotoxin D. mecA: MRSA gene.

Staphylococcus aureus produces an extracellular thermostable nuclease, which is encoded by the *nuc* gene and is one of the most distinctive and useful traits that could be used to differentiate *S. aureus* from other *Staphylococcus* species [35]. Similarly, Manukumar and Umesha [36] demonstrated the *nuc* gene in all *S. aureus* strains isolated from different food samples in India. Also, Maktabi et al. [37] detected the *nuc* gene in all 150 *S. aureus* isolates obtained from different raw meat samples in Iran.

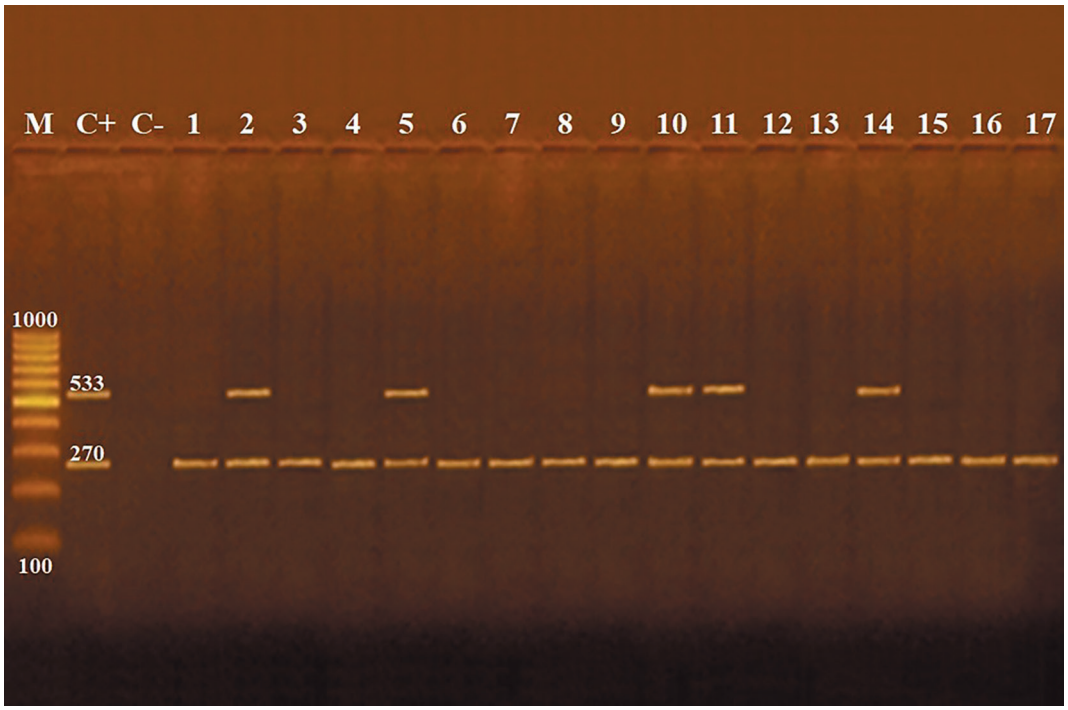

Figure 1. Agarose gel electrophoresis of PCR amplification products of species-specific (nuc) and methicillin resistance (mecA) genes in *S. aureus* isolates. **Lane M**: 100 bp ladder as a molecular size DNA marker. **Lane C+**: Positive control for nuc (270 bp) and mecA (533 bp) genes in *S. aureus* isolates. **Lane C-**: Negative control. **Lanes 2, 5, 10, 11, and 14**: Positive *S. aureus* strains for the mecA gene. **Lanes 1, 3, 4, 6, 7, 8, 9, 12, 13, 15, 16, and 17**: Negative *S. aureus* strains for the mecA gene. The nuc gene, specific to *S. aureus* with a molecular size of 270, was positive for all 17 isolates

The isolates of *S. aureus* were tested for demonstration of enterotoxins and the *mecA* virulence gene of MRSA. In a Spanish study, among 27 *S. aureus* isolates from rabbit samples, Rodriguez-Calleja et al. [32] detected two harbored genes for staphylococcal enterotoxin B (*Seb*), and two harbored genes for staphylococcal enterotoxin C (*Sec*), while the remaining isolates were negative for *Sea*, *Seb*, *Sec*, *Sed*, and *See*. Besides, Kohler et al. [31] identified 102 (67.5%) staphylococcal strains carrying enterotoxin genes from rabbit samples in Switzerland. On the other hand, all 281 *S. aureus* isolates from rabbit samples in Fujian, China, detected by Wang et al. [38] were negative for *Sea* and *Seb* virulence genes. *S. aureus* toxins were not detected in rabbit meat samples in Slovakia [39]. According to Le Loir et al. [40], most *S. aureus* strains isolated from food do not produce SEs. Moreover, other species of *Staphylococcus* can produce SEs, but are not looked for in routine testing.

The isolates were also tested for presence of the *mecA* gene. Moreno-Grúa et al. [41] identified 30 methicillin-resistant *S. aureus* with a percentage of 12.5%, while the methicillin-resistant *mec*A gene was detected in 27 isolates with a percentage of 11.25% in the studied isolates from commercial rabbitries in the Iberian Peninsula. Besides, MRSA was found in 48% (11/23) of the rabbits carrying *S. aureus* in Italy by Agnoletti et al. [42]. Furthermore, Lozano et al. [14] identified MRSA in 5 out of 318 (1.6%) food samples (pork, chicken, rabbit, veal, and wild boar) in Spain. On the contrary, Kohler et al. [31] failed to detect the *mec*A gene from the investigated *Staphylococcus* isolates obtained from rabbit samples in Switzerland.

The results of the present study highlight that rabbit meat may constitute a risk for consumers and especially for immunocompromised individuals. In immunocompromised persons, the specific and non-specific immune responses are not able to act as barriers to prevent colonization of the gastrointestinal tract, and ingestion of food contaminated by MRSA may sometimes lead to lethal diseases [43].

Antimicrobial susceptibility profile of S. aureus isolates

The isolates of *S. aureus* (n=17) were tested for antimicrobial susceptibility as depicted in Table 4. The highest resistance was recorded against kanamycin, cephalexin, oxacillin, penicillin G, tetracycline, and nalidixic acid with a percentage of 100%, 76.5%, 64.7%, 58.8%, 52.9%, and 47.1%, respectively, while the most effective antimicrobials were amikacin, linezolid, gentamicin, ciprofloxacin, clindamycin, cefotaxime, erythromycin with a percentage of 94.1%, 88.2%, 88.2%, 82.4%,76.5%, 70.6%, and 70.6%, respectively. The isolates' MAR index ranged from 0.063 to 1 with an average of 0.389 (Table 5).

Antibiotic susceptibility testing was performed on all 17 *S. aureus* isolates. A total of sixteen antimicrobial drugs from various antibiotic classes were employed. Some were chosen because research revealed that a substantial percentage of bacteria were resistant to them [44,45]. Antibiotics with veterinary and human health implications were also considered. The high prevalence of multidrug-resistant strains found in this study is consistent with previous findings in intensively raised rabbits in the Iberian Peninsula [38]. The obtained results were in parallel with Attili et al. [46] who documented high tetracycline resistance (95.8%), but low penicillin resistance (3.1%) of 96 *S. aureus* strains isolated from rabbit samples in central Italy was observed. Also, Wang et al. [37] detected resistance of *S. aureus* strains isolated from rabbit samples in Fujian Province, China, to kanamycin and penicillin with a percentage of 19.57% and 11.03%, respectively. In accordance with the results, Simonová et al. [39] revealed high resistance among *S. aureus* isolates obtained from rabbit meat samples in Slovakia to penicillin (100%). Also, high resistance to erythromycin and gentamycin (64% for each) was recorded. In agreement with the current study, Rodriguez-Calleja et al. [47] found high resistance of *S. aureus* strains isolated from rabbit meat in Spain to tetracycline (61.5%), but in difference with the detected results, low penicillin resistance (26.9%) was reported.

Table 4. Antimicrobial resistance profile of *S. aureus* isolates (n=17)

Antimicrobial agents	Sensitive		Intermediate		Resistant	
	No.	%	No.	%	No.	%
Kanamycin (K)	—	—	—	—	17	100
Cephalexin (CE)	3	17.6	1	5.9	13	76.5
Oxacillin (OX)	4	23.5	2	11.8	11	64.7
Penicillin G (P)	5	29.4	2	11.8	10	58.8
Tetracycline (T)	7	41.2	1	5.9	9	52.9
Nalidixic acid (NA)	6	35.3	3	17.6	8	47.1
Cephalothin (CN)	9	52.9	—	—	8	47.1
Ampicillin (AM)	9	52.9	1	5.9	7	41.2
Sulphamethoxazol (SXT)	10	58.8	1	5.9	6	35.3
Cefotaxime (CF)	12	70.6	1	5.9	4	23.5
Clindamycin (CL)	13	76.5	—	—	4	23.5
Erythromycin (E)	12	70.6	2	11.8	3	17.6
Ciprofloxacin (CP)	14	82.4	1	5.9	2	11.8
Gentamicin (G)	15	88.2	—	—	2	11.8
Linezolid (LZ)	15	88.2	1	5.9	1	5.9
Amikacin (AK)	16	94.1	—	—	1	5.9

n: Number of *S. aureus* isolates. No.: Number of sensitive, intermediate or resistant *S. aureus* isolates.%: Percentage of sensitive, intermediate or resistant *S. aureus*.

High penicillin resistance is not surprising because of its widespread use for treatment in humans and animals. Although the European Union regulates the use of antibiotics as growth promoters, the existence of resistant organisms is still found, confirming their intensive use in therapy [48]. High susceptibility to erythromycin in this study may be attributed to the fact that this antibiotic is not used in rabbits due to its toxicity [49]. More resistant strains are thought to have the best chances of survival; thus, their prevalence increased as they filled the space left by those who did not survive the antibiotic treatment. This finding

Table 5. Resistance profile of multi-drug resistant *S. aureus* isolates (n=17)

Pattern	Resistance profile	Number of antibiotics	Number of isolates (%)	MAR
I	K, CE, OX, P, T, NA, CN, AM, SXT, CF, CL, E, CP, G, LZ, AK	16	1 (5.88%)	1
II	K, CE, OX, P, T, NA, CN, AM, SXT, CF, CL, E, CP, G	14	1 (5.88%)	0.875
III	K, CE, OX, P, T, NA, CN, AM, SXT, CF, CL, E	12	1 (5.88%)	0.75
IV	K, CE, OX, P, T, NA, CN, AM, SXT, CF, CL	11	1 (5.88%)	0.688
V	K, CE, OX, P, T, NA, CN, AM, SXT	9	2 (11.76%)	0.563
VI	K, CE, OX, P, T, NA, CN, AM	8	1 (5.88%)	0.500
VII	K, CE, OX, P, T, NA, CN	7	1 (5.88%)	0.438
VIII	K, CE, OX, P, T	5	1 (5.88%)	0.313
XI	K, CE, OX, P	4	1 (5.88%)	0.250
X	K, CE, OX	3	1 (5.88%)	0.188
XI	K, CE	2	2 (11.76%)	0.125
XII	K	1	4 (23.5%)	0.063
Average				0.389

MAR: Multiple Antibiotic Resistance index
K: Kanamycin **CE: Cephalexin** **OX: Oxacillin** **P: Penicillin G**
T: Tetracycline **NA: Nalidixic acid** **CN: Cephalothin** **AM: Ampicillin**
SXT: Sulphamethoxazol **CF: Cefotaxime** **CL: Clindamycin** **E: Erythromycin**
CP: Ciprofloxacin **G: Gentamicin** **LZ: Linezolid** **AK: Amikacin**

suggests that long-living rabbits play an important role in maintaining resistant strains and spreading them to newly introduced and newborn individuals.

Conclusion

Generally, the current study identified multidrug-resistant and multitoxigenic *S. aureus* in rabbit meat, highlighting its potential as a source for transmitting foodborne pathogens. The data obtained confirms that rabbit meat can cause staphylococcal intoxication in consumers, with the majority of *Staphylococcus* isolates being *S. aureus*, and some testing positive for MRSA and enterotoxin virulence genes. The high *Staphylococcus* count in raw retail rabbit meat in the Egyptian market suggests a risk of common foodborne diseases. The assessment of antibiotic resistance and pathogenicity revealed severe issues for food industrial applications and quality control as many isolates showed resistance to at least three antibiotics. Thus, initiatives are needed to enhance sanitary standards in Egyptian markets, especially in traditional markets with higher contamination rates. Health agency regulations should be disseminated to all workers, and safety programs for slaughtering and meat preparation outlined by international organizations and national authorities must be followed. Effective preventive measures must be authorized and implemented to safeguard consumer health.

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