Journal of Applied Biology & Biotechnology Vol. 13(6), pp. 134-141, November-December, 2025
Available online at http://www.jabonline.in
DOI: 10.7324/JABB.2025.v13.i6.16

A novel multidrug-resistant *Salmonella enterica* strain (A10) isolated from eggshells in wet markets of Taif, Saudi Arabia

Haitham Alharthi[1], Ghanem Al-Ghamdi[1], Tariq Alpakistany[2], Bandar Almiman[1], Abeer M. Alghamdi[1], Fawzyah Obeedallah Albaldi[1], Nourah M Almimoni[1], Mohammed Dosh[3], Mohammed A. Thabet[1], Khalid S Al-Zahrani[1], Thuraya Ahmed Mohammed[1,4], Mohammad Melebari[1]*, Khaled S. Gazi[1]

[1]Department of Biology, Faculty of Science, Al-Baha University, Al-Baha 65431-65779, Saudi Arabia.
[2]King Faisal Medical Complex Ministry of Health, , Taif, Saudi Arabia.
[3]Department of Public Health, College of Public Health and Health Informatics, University of Ha'il, Ha'il, Saudi Arabia.
[4]Department of Microbiology and Molecular Biology, Faculty of science and Technology, Al Neelain University, Khartoum, Sudan.

ARTICLE INFO

Article history:
Received on: 07/02/2025
Accepted on: 17/06/2025
Available online: 16/09/2025

Key words:
Antibiotic resistance,
Wet market,
Egg shells,
Sanitation of eggs,
Public health Laws,
Poultry,
Salmonella.

ABSTRACT

The fact that multidrug-resistant bacteria are common on eggshells sold in wet markets is very vulnerable for public health, especially since eggs are often handled poorly. As part of this study, chicken eggshells from different wet markets in Taif, Makkah area, Saudi Arabia, were collected and tested for bacterial pollution. Overall, a total of 150 eggs were randomly selected and examined for both external and internal microbial contamination. The VITEK 2 antimicrobial sensitivity showed significant multidrug resistance, especially in the *Salmonella enterica* strain. 16S r-RNA was used to identify five types of bacteria: *Escherichia hermannii*, *Escherichia coli*, *Staphylococcus aureus*, *Staphylococcus xylosus*, and a new strain of *S. enterica* (A10). Finding *S. enterica* A10 on eggshells in this area is the first narrative of its kind, showing that eggshells are an underrated way for antibiotic-resistant bacteria to spread. More protection, limits on drug use in chicken, and better cleanliness in local markets are all needed because of these results.

1. INTRODUCTION

Around the world, poultry eggs are an affordable and nutrient-dense source of protein that is widely consumed. However, eggs are now known to be one of the sources of outbreaks in the US [1-4]. Moreover, raw egg-based desserts, tiramisu, and milkshakes are becoming more popular among consumers, who may be putting themselves at greater risk of bacterial illnesses. The egg's protective shell prevents microbes from reaching the egg's interior. The shell, on the other hand, is constantly covered with muck, feathers, blood, nesting materials, and pathogens [5,6].

Antibiotics are effective in treating bacterial illnesses [7,8]. Nevertheless, the incorrect use of antibiotics might give rise to antibiotic resistance (AR), which occurs when microbes develop resistance to antibiotics [7,9-12]. When bacteria undergo genetic changes and develop resistance to antibiotics, the likelihood of disease transmission and mortality rates increase, while the effectiveness of therapy diminishes [8,12,13]. AR is a significant global health issue, causing numerous fatalities annually [14-18]. High rates of carbapenem resistance were detected in a study conducted in Medina between 2014 and 2018, with 38.4% for emipenem and 46.1% for meropenem, the selected drugs according to local standards [19]. This validates the necessity of increasing public consciousness and offering direction about the proper utilization of antibiotics to address the escalating problem of AR. There are widespread concerns regarding the ability of antibiotic-resistant bacteria to transmit to people through the food chain in various contexts such as food production systems [20-22]. Antibiotic-resistant bacteria present on eggshells can be ascribed to multiple factors, including cross-contamination during handling and processing, excessive antibiotic use in animal husbandry, and the persistence of resistant strains in the marketplace.

In addition, the secretion of bacteria such as *Escherichia coli* and *Salmonella* by chickens, the host, influences the prevalence of these bacteria on eggshells; *E. coli* typically contaminates eggs through fecal matter or during hatching, while *Salmonella* is intermittently excreted [23-27]. Antibiotic-resistant bacteria present on eggshells can be ascribed to multiple factors, including cross-contamination during handling and processing [28], excessive antibiotic use in

*Corresponding Author:
Mohammad Melebari,
Department of Biology, College of Science Al-Baha University,
Al-Baha 65799, Saudi Arabia.
E-mail: melebari@bu.edu.sa

animal husbandry [29], and the persistence of resistant strains in the marketplace [30-32].

It is important to take note of the prevalence and detection of antibiotic-resistant bacteria on eggshells obtained from wet markets. Wet markets serve as bustling hubs for the distribution and trade of food in numerous countries. They offer a diverse selection of perishable products, including eggs, which are a staple in numerous global cuisines [32,33]. They can contribute to contamination of foodstuffs, especially eggshells, with antibiotic-resistant bacteria due to often uncontrolled and inadequate sanitation measures [33]. Since animals are frequently killed on site, wet markets can also increase waste and pollution because vital waste must be managed appropriately.

Eggs that are cracked have a high susceptibility to contamination, which can lead to a decrease in egg quality and constitute a threat to consumer health [34]. Eggshell fractures provide spores with direct entry into the egg, facilitating swift bacterial proliferation during processing and storage, hence increasing the risk of food borne illnesses [35-37]. Microscopic fissures may be unnoticed by the unaided vision and may not manifest until several days after being stored, hence posing challenges in identifying fractures [38]. The quality of eggs can be considerably reduced as a result of many types of fractures, such as cracks of varying lengths, widths, and severity, which can occur due to the intricate composition of the eggshell [39-41]. Eggshell cracks can give microorganism's direct access to the egg, causing them to breed rapidly and potentially contaminate them with strains of antibiotic-resistant bacteria [34].

Understanding the prevalence and identification of antibiotic-resistant bacteria on eggshells sourced from wet markets is essential for evaluating the potential health risks associated with egg consumption. This knowledge is crucial for developing effective control measures aimed at ensuring food safety. By assessing the frequency of these resistant strains, and we can better inform public health strategies and regulatory policies to mitigate the risks of food-borne illnesses linked to AR in poultry products [41-46]. This research aims to investigate the prevalence and detection of antibiotic-resistant bacteria in eggshells obtained from the wet markets in Taif city, Makkah region.

2. MATERIALS AND METHODS

2.1. Study Area

Taif is a Saudi city located in the Makkah region, west of Saudi Arabia, on the eastern slope of the Sarawat Mountains, with a height of 1,700 m, and gradually increasing towards the west and south to 2,500 m with a population of 1,281,613.

2.2. Sample Collection and Isolation of Bacteria

To assess the microbiological quality of poultry eggs, various samples of organic eggs and commercial eggs (n = 150) were collected from various grocery shops and from different farms in Taif city of Makkah region.

The 150-egg sample size was selected because they were easy to find locally, could be processed in a lab, and were statistically large enough to find significant trends in bacterial contamination and resistance. The samples were collected in sterile polythene bags and stored on ice, then processed for isolation and purification.

2.3. Isolation and Bacterial Identification

All the experiments were conducted following the methods proposed by Melebari *et al*. [47], with some modifications. The egg samples were labeled, and then, the surfaces of the intact shell eggs were swabbed with sterile cotton swabs that had been moistened with tryptone soy broth (TSB) from Difco Laboratories in Detroit, USA. After collecting samples from the outside surface of the shell (n = 150), the same eggs (n = 150) were analyzed to assess the microbiological condition inside (total = 300). In order to prevent the spread of contaminants from the shell to the contents, the eggs were immersed in a solution of 70% alcohol for duration of 30 s. They were then rinsed with sterile water and left to dry in a biosafety cabinet. The samples were swabbed and then transferred to the Eppendorf tube containing TSB using sterile techniques, then incubated at 37°C for 24 h, then streaked onto different media including Eosin Methylene Blue Agar (EMb; Scharlau), Blood Agar Base (BA; Scharlau), Sorbitol MacConkey agar (SMA) (SMA; Neogen), *Staphylococcus* medium (SA; Oxoid) and *Salmonella shigella* Agar (SSA; Fluka Analytical) plates, and incubated at 37°C for 24 h. Visible bacteria colonies on the plates were inspected and then sub-cultured on fresh sterile Nutrient agar plates to get pure isolates. The pure isolates were transferred to sterile agar slants and kept at a temperature of 4□ for future use. Selected strains were described and identified using polymerase chain reaction (PCR) and their identity and genetic connection were confirmed.

Searches for similarities with nucleotide sequences in the Gen Bank were performed using BLAST search, and the sequence was deposited in the GeneBank under accession number (PP967976). The identified sequences were used to construct the phylogenetic tree.

2.4. Antimicrobial Susceptibility Testing

Antimicrobial susceptibility testing was performed using the automated VITEK 2 system (bioMérieux, France) following the manufacturer's instructions. Bacterial isolates were cultured on Mueller–Hinton agar and incubated at 37°C for 18–24 h. A bacterial suspension was prepared in sterile saline (0.45% NaCl) and adjusted to 0.5 McFarland standard using a DensiCHEK Plus system. The suspension was inoculated into VITEK AST cards and analyzed using the VITEK 2 Compact system. Minimum inhibitory concentrations were automatically determined and interpreted. *Staphylococcus aureus* ATCC 29213 and *E. coli* ATCC 25922 were used as normal reference strains for quality control and to prove that the VITEK 2 method worked properly in susceptibility testing.

2.5. Identification of the AR Strains via PCR

PCR reactions were done using universal bacterial primers 27F (5'-AGAGTTTGATCMTGGCTCAG-3') and 1492R (5'-TACGGYTACCTTGTTACGACTT-3'), which were selected because they can copy almost the whole *16S rRNA* gene [48]. Thermal cycling was done in 35 rounds of denaturation at 95°C for 30 s, annealing at 58°C for 30 s, extension at 72°C for 90 s, and final extension at 72°C for 7 min. With SafeView™ DNA dye on a 2% agarose gel in 1X TBE buffer, gel electrophoresis was carried out. Using a UV transilluminator, DNA bands were seen, and MoleculeOn's 100–1000 bp DNA ladder was utilised as a molecule size indicator.

2.6. Statistical Analysis

The Statistical Package for the Social Sciences version 25 was used for the statistical study. To find out how often bacterial isolates occurred descriptive statistics were used. Chi-square tests were used to see how often different types of eggs and bugs were contaminated and how they responded to antibiotics. A $P < 0.05$ was thought to be scientifically important.

3. RESULTS

3.1. Identification of Bacterial Isolates

In this study, 300 samples were studied, varied between commercial eggs and organic eggs from the shops in addition to farms in the city of Taif. Results of this study were obtained after bacterial isolates were isolated from chicken eggs on different media, and it was as the following: (62%) grown on the BA media, (7%) on the *S. shigella* agar media, (12%) on the EMb media, and 15% on the *Staphylococcus* agar media, as shown in [Figure 1].

Following the isolation of samples from chicken eggs, a total of 10 *S. aureus* isolates were identified in both commercial and organic eggs. In addition, five *Salmonella* isolates and 5 *E. coli* isolates were detected in organic chicken eggs. A Chi-square test revealed that organic eggs showed a higher prevalence of contamination by highly pathogenic strains when compared to commercial eggs ($P < 0.05$). The Chi-square test showed that bacterial contamination was significantly more frequent on the surface of the eggs compared to the interior ($P < 0.05$), and there is a higher likelihood of them contaminating the inside of the eggs if there are any fractures on the eggshell.

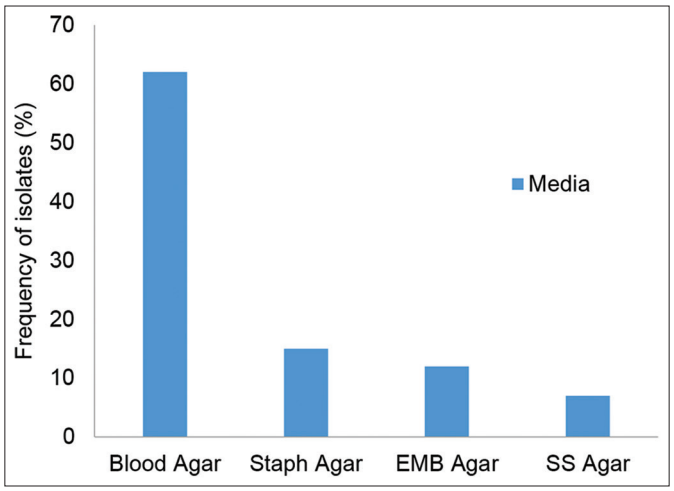

Figure 1: Frequency of incidence of bacteria isolated from retailed eggshells on different media.

3.2. Antimicrobial Sensitivity Test

Tests detected the sensitivity or resistance of bacterial isolates against the most common antibiotics via VITEK [Table 1]. Results have shown that *Escherichia hermannii* was resistant to (Amoxicillin [AMX] +, + Ampicillin, + Piperacillin, Trimethoprim/Sulfamethoxazole [SMZ]). While *Staphylococcus xylosus* is resistant to Cefixime +, + Ceftazidime, *E. coli* have shown their resistance to the following

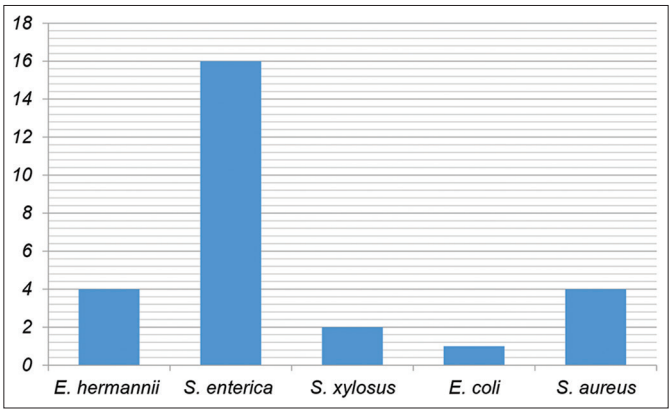

Figure 2: Antibiotic-resistant bacterial isolates against 19 antibiotics used in this study.

Table 1: Antibiotic susceptibility profile of the bacterial strains.

Antimicrobial	*Escherichia hermannii*	*Salmonella enterica*	*Staphylococcus xylosus*	*Escherichia coli*	*Staphylococcus aureus*
Amoxicillin+	+	+	-	-	-
Ampicillin+	+	+	-	-	-
Piperacillin+	+	+	-	-	+
Trimethoprim/sulfamethoxazole	+	+	-	+	-
Cefaclor+	-	+	-	-	-
Cefazolin	-	+	-	-	-
Cefuroxime	-	+	-	-	-
Cefuroxime axetil	-	+	-	-	-
Ceftriaxone	-	+	-	-	-
Amikacin	-	+	-	-	-
Gentamicin	-	+	-	-	-
Tobramycin+	-	+	-	-	-
Ciprofloxacin	-	+	-	-	-
Levofloxacin+	-	+	-	-	-
Ofloxacin+	-	+	-	-	-
Nitrofurantoin	-	+	-	-	-
Cefixime+	-	-	+	-	+
Ceftazidime+	-	-	+	-	+
Benzylpenicillin	-	-	-	-	+

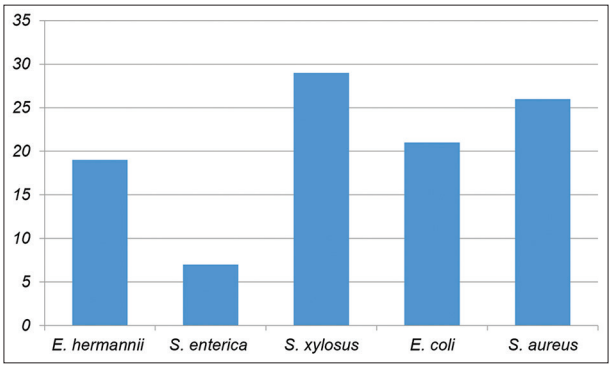

Figure 3: Antibiotic-sensitive bacterial isolates against 19 antibiotics used in this study.

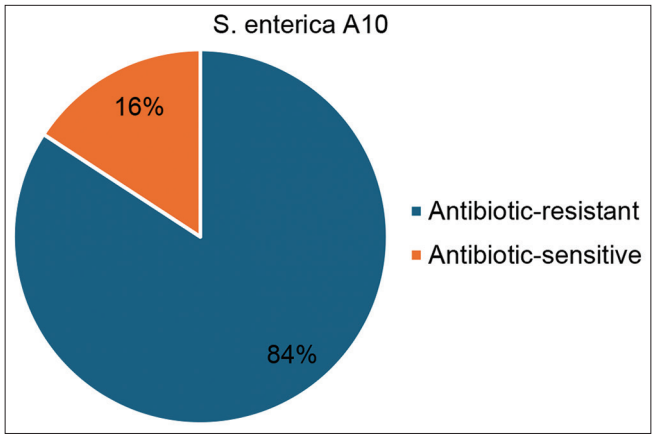

Figure 4: Strain A10 against the antibiotic agents used in this study.

antibiotics (Trimethoprim/SMZ), and *S. aureus* is resistant to (Benzylpenicillin, + Piperacillin, + Cefixime, + Ceftazidime). The results showed that *Salmonella enterica* has a high rate of resistance to antibiotics, as shown in [Figures 2-4].

Results revealed the presence of a novel strain, A10, that was identified as *S. enterica* that were isolated from the eggshell obtained from the wet markets in Taif city., The 16S rRNA sequencing method was used in order to accomplish the molecular characterization of the *S. enterica* A10 strain. After the genomic DNA was extracted, primers were used to amplify the region of 16S rRNA that was under investigation. The 16S rRNA partial sequencing that was obtained was then uploaded to the NCBI database of the GenBank with the accession number PP967976 (https://www.ncbi.nlm.nih.gov/Genbank). The obtained sequences were blasted with the highly similar sequences which were downloaded and imported in BIOEDIT version 7.1.11. Using molecular evolutionary genetics analysis 11 software, a neighbor-joining tree was constructed for the *Salmonella* isolates against 15 *Salmonella* spp. from the Gene bank database *S. enterica* A10 was in the same bootstrap with other *Salmonella* in order to determine whether or not it was closely related to other *Salmonella* species as can be seen in Figure 5.

4. DISCUSSION

The eggs used in this study were contaminated since they were obtained directly from farmers to retail establishments and then collected for the study. Five bacterial isolates, including *E. hermannii*, *E. coli*, *S. aureus*, *S. enterica*, and *S. xylosus*, were isolated from the eggshell and yolk layer, according to this study.

It is unclear whether these isolates originated from the reproductive system, the digestive system, or both. *Salmonella* was detected in the

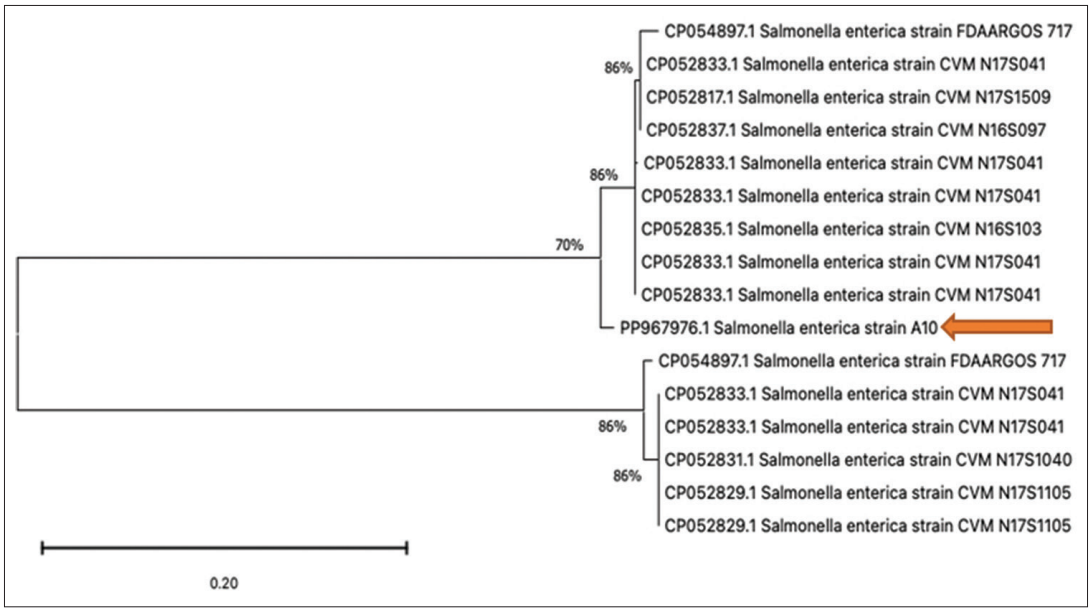

Figure 5: Phylogenetic tree made from Salmonella *enterica* A10 (*16S rRNA* genes) and 15 other *Salmonella* spp. strains found in GenBank. The Neighbor-Joining method in molecular evolutionary genetics analysis 11 software was used to build the tree. Bootstrap values (n = 1000 repeats) are shown at the ends of each branch. The red line points to the new A10 strain that was found in this study. It is closely related to other known multidrug-resistant *S. enterica* strains, which suggests that they share genes and may have evolved together.

egg samples, corroborating findings from another study conducted in Faisalabad city [49], which also reported contamination with *E. coli*, *Salmonella enteritidis*, *Bacillus subtilis*, and *S. aureus*. Notably, *S. enteritidis* was identified as the most prevalent isolate, accounting for 40.83% of the total bacterial isolates in the Faisalabad samples [49]. This high prevalence of *S. enteritidis* raises significant concerns regarding food safety, as it is well-known for its role in food-borne illnesses associated with egg consumption. The presence of multiple bacterial pathogens in the egg samples underscores the importance of implementing stringent hygiene and monitoring practices in poultry production and handling to mitigate the risk of contamination and ensure consumer safety [46].

Our findings differ from a study conducted on eggshells from three major markets in Minna, Nigeria, which identified *S. aureus* as the most prevalent isolate, comprising 36.4% of the samples [50]. In addition, research from Tanzanian markets revealed that *Bacillus* species were the most predominant, accounting for 39.7% of the analyzed samples [51]. These discrepancies underscore the regional variations in bacterial contamination patterns that are linked to different egg production and handling practices. The high prevalence of *S. aureus* in Nigeria may point to potential hygiene issues and cross-contamination during egg processing and distribution. Conversely, the dominance of *Bacillus* species in Tanzania could reflect differences in environmental conditions or microbial ecosystems that affect contamination rates [52].

Furthermore, our results are also at odds with findings from a study conducted in Zambia, which evaluated 216 pooled egg samples at two contamination levels: Eggshell and egg content [53]. That study reported that 2.31% of eggshell samples tested positive for *Salmonella* spp. (5 out of 216 samples), while 34.26% (74 out of 216) were positive for *E. coli*. Notably, the eggs tested negative for *Salmonella* and *E. coli* in the egg content, indicating that eggshells exhibited a higher level of contamination with *E. coli* than the egg contents ($\chi^2 = 20.95$, $P < 0.0001$). Against *E. coli* isolates, imipenem showed 100% effectiveness. However, *Salmonella* demonstrated a resistance of 80% to tetracycline (TET) and 60% to ampicillin, while *E. coli* exhibited resistance rates of 94.6% to colistin sulfate, 83.8% to TE, and 59.5% to ampicillin. Thus, the Zambian study highlighted a significantly higher contamination of egg surfaces with *E. coli* (34.3%) compared to *Salmonella* (2.31%) [53].

These variations in bacterial contamination across different regions emphasize the importance of localized studies to identify specific risk factors associated with egg safety. Factors such as farming practices, environmental conditions, and market handling protocols can significantly influence the microbial profile of eggs [51,54,55]. The predominance of *E. coli* in Zambia, along with the resistance patterns observed, raises concerns regarding public health and food safety, particularly in relation to the potential for antibiotic-resistant strains entering the food supply.

The VITEK 2 system's MIC values showed that *S. enterica* A10 was resistant to many β-lactam drugs, even third-generation cephalosporins. This is a trend of resistance that should be taken very seriously. A10 was more resistant to cefixime and ceftazidime than global isolates. This could be because of selective forces in that area or possible horizontal gene transfer. This fits with what other countries have found, but it shows a resistance profile that has not been seen before in Saudi Arabia.

This study is unique because it found *S. enterica* A10 on eggshells, which is an area that is often ignored as a source of contamination. It also sequenced the genome of this strain (GenBank Accession: PP967976), which will be used as a new standard for future epidemic monitoring [Figure 4].

This study presented findings that are inconsistent with previous research that identified the presence of *E. coli* and *Salmonella* species in table eggs sold for human consumption in Peshawar city [56], along with their resistance to various antimicrobial drugs commonly used in Pakistan for poultry and human health [56]. A total of 80 eggs were collected from various shops in Taif city for this analysis. *E. coli* and *Salmonella* spp. were isolated from 85.36% (70) and 14.63% (12) of the total collected egg samples (80), respectively. The shells of eggs showed significantly higher contaminations as compared to egg internal contents for both *E. coli* and *Salmonella* spp. Hence, it can be concluded from this study that, the table eggs sold in the markets of Peshawar Pakistan are infected with multidrug resistance (MDR) strains of *E. coli* and *Salmonella* spp. [57].

The results of our study on antibiotic sensitivity and resistance indicate that bacterial isolates exhibit varying levels of sensitivity to certain antibiotics while showing resistance to others. Notably, *Salmonella* demonstrated a significant level of AR, highlighting its serious pathogenic potential. Our findings align with those from a similar study conducted in China [58], which also analyzed isolates from both eggs and chicken intestines. The antibiotics SMZ, ceftriaxone, nalidixic acid, cefazolin, and AMX showed considerable resistance among the isolates. In addition, strains isolated from chicken intestines displayed relative resistance to ciprofloxacin (CIP) and tetracycline (TET), while strains from eggs exhibited resistance to gentamicin (GEN) and kanamycin. Among the eight *Salmonella* strains identified from eggs, six or more antibiotics were resisted, whereas most bacterial isolates were resistant to two to five antibiotics [58]. Using PCR amplification, 34 multidrug-resistant *Salmonella* strains were examined for 33 virulence genes and 15 types of drug resistance genes. Six isolates were found to carry more than 20 virulence genes, and ten possessed seven or more drug resistance genes [58].

In a study from Minna, Gram-positive bacteria exhibited the highest sensitivity to GEN (100%) but showed 100% resistance to cloxacillin, ceftazidime, and erythromycin. Similarly, while no Gram-negative bacteria were resistant to CIP, *E. coli*, *Salmonella*, and *Shigella* isolated from eggshells were resistant to augmentin and AMX [50]. Further analysis of the result revealed that all the isolated bacteria from eggshells were multidrug resistant except *Neisseria* spp. with multidrug-resistant index >0.2 [50].

The high prevalence and the detection of pathogenic and multidrug-resistant strains, in the current study highlight the poor management system in the poultry farm and different related sites, which is in agreement with other studies [59-65]. In this study, poultry has been reported as a source of nontyphoidal *Salmonella* which are resistant to clinically relevant antibiotics and remarkably pose a high risk to both animals and humans. Moreover, the indiscriminate use of antimicrobials in poultry animals for growth promotion and disease prevention is considered the key driver behind the surge. More attention should be focused on increasing antibiotic surveillance capacity to cope with the spread of emerging resistance and on the alternative therapeutic approaches.

5. CONCLUSION

Overall, this study has been able to demonstrate the presence of different isolates outside and inside eggs in Taif. It has also shown the

presence of resistant isolates outside and inside eggs, which poses a serious public health concern given the consumption patterns of these eggs.The eggshells of eggs sold in the Makkah province were found to be infected with antibiotic-resistant strains of *E. hermannii*, *E. coli*, *S. aureus*, *S. enterica*, and *S. xylosus*.

The study highlighted the significant incidence of multidrug-resistant nontyphoidal *Salmonella* in chicken farms, as well as inadequate management systems and the indiscriminate use of antimicrobials for growth promotion and disease prevention. It advocates for improved antibiotic surveillance and alternate therapy strategies.

Transferring tainted eggs to consumers poses a significant risk to public health. Antibiotic use in chicken farms should be decreased. Employing good hygiene when collecting, storing, and processing eggs.

The results of our study's antibiotic sensitivity and resistance test show that the bacterial isolates are sensitive to some antibiotics and resistant to others, and the strain *Salmonella entrica* A10 was a powerfull antibiotic resistant pathogen.

This work has some challenges, even though it does demonstrate that multidrug-resistant *S. enterica* A10 can be found on eggshells. First, the study only looked at Taif City, so it might not show the overall trends of disease in Saudi Arabia. Second, the molecular characterization was limited to 16S rRNA sequencing. Whole-genome sequencing would give us a better understanding of how resistance works. Finally, there was no direct examination of sources of pollution in fields or wet markets. This could have helped find the origin of resistant types more exactly.

6. FUTURE PROSPECTIVES

Based on this conclusion, it is necessary to give training programs on the best practices of improving the understanding of individuals regarding the proper handling of eggs for those responsible for their care. It is highly recommended to improve the hygienic conditions of poultry farms and regulate the use of antibiotics to prevent their misuse and overuse. Consumers should clean the eggshell before breaking it for cooking. Further research should be conducted to identify the source of contamination. The level of risk associated with consuming contaminated eggs should be evaluated. Education should include instructions on good sanitary practices for handling.

7. AUTHORS' CONTRIBUTIONS

All authors made substantial contributions to the conception and design, acquisition of data, or analysis and interpretation of data; took part in drafting the article or revising it critically for important intellectual content; agreed to submit to the current journal; gave final approval of the version to be published; and agreed to be accountable for all aspects of the work. All the authors are eligible to be author as per the International Committee of Medical Journal Editors (ICMJE) requirements/guidelines.

8. FUNDING

There is no funding to report.

9. CONFLICT OF INTEREST

The authors report no financial or any other conflicts of interest in this work.

10. ETHICAL APPROVAL

This study does not involve experiments on animals or human subjects.

11. DATA AVAILABILITY STATEMENTS

The contig that was obtained during the study was submitted to the NCBI database under accession number PP967976 and the experimental data that support the findings of this study are available from the corresponding author upon request.

12. PUBLISHER'S NOTE

All claims expressed in this article are solely those of the authors and do not necessarily represent those of the publisher, the editors and the reviewers. This journal remains neutral with regard to jurisdictional claims in published institutional affiliation.

13. USE OF ARTIFICIAL INTELLIGENCE (AI)-ASSISTED TECHNOLOGY

The authors declares that they have not used artificial intelligence (AI)-tools for writing and editing of the manuscript, and no images were manipulated using AI.

REFERENCES

1. Adak GK, Meakins SM, Yip H, Lopman BA, O'Brien SJ. Disease risks from foods England and wales 1996-2000. Emerg Infect Dis. 2005;11(3):365-72. https://doi.org/10.3201/eid1103.040191
2. Koutsoumanis K, Allende A, Alvarez□Ordóñez A, Bolton D, Bover□Cid S, Chemaly M, *et al. Salmonella* control in poultry flocks and its public health impact. EFSA J. 2019;17(2):e05596. https://doi.org/10.2903/j.efsa.2019.5596
3. Braden CR. *Salmonella enterica* serotype enteritidis and eggs: A national epidemic in the United States. Clin Infect Dis. 2006;43(4):512-7. https://doi.org/10.1086/505973
4. Guard-Petter J. The chicken, the egg and *Salmonella* enteritidis. Environ Microbiol. 2001;3(7):421-30. https://doi.org/10.1046/j.1462-2920.2001.00213.x
5. De Reu K, Grijspeerdt K, Messens W, Heyndrickx M, Uyttendaele M, Debevere J, *et al*. Eggshell factors influencing eggshell penetration and whole egg contamination by different bacteria, including *Salmonella* enteritidis. Int J Food Microbiol. 2006;112(3):253-60. https://doi.org/10.1016/j.ijfoodmicro.2006.04.010
6. Board RG, Tranter HS. The microbiology of eggs. In: Egg Science and Technology. Danville: Food Products Press; 1995. p. 81-104. https://doi.org/10.1201/9781003069439
7. Ventola CL. The antibiotic resistance crisis: Part 1: Causes and threats. Pharm Ther. 2015;40(4):277-83.
8. Llor C, Bjerrum L. Antibiotic prescribing in primary care: A review of the literature. Infect Drug Resist. 2014;7:207-15. https://doi.org/10.2147/IDR.S53649
9. World Health Organization. Antimicrobial Resistance. Source. Switzerland: WHO Antimicrobial Resistance; 2023.
10. Centers for Disease Control and Prevention. Antibiotic Resistance. Source. United States: CDC Antibiotic Resistance; 2022.
11. Llor C, Bjerrum L. Antimicrobial resistance: Risk associated with antibiotic overuse and initiatives to reduce the problem. Infect Adv Drug Safe. 2014;5:229-41. https://doi.org/10.1177/2042098614554919
12. Bush K, Courvalin P, Dantas G, Davies J, Eisenstein B, Huovinen P, *et al*. Tackling antibiotic resistance. Nat Rev Microbiol. 2011;9(12):894-6. https://doi.org/10.1038/nrmicro2693
13. Ranjbar R, Alam M, Antimicrobial Resistance Collaborators. Global

burden of bacterial antimicrobial resistance in 2019: A systematic analysis. Evid Based Nurs. 2022;399 (10325):629-655.
14. Melebari M. Shiga toxin-producing escherichia coli: A focused review of the middle east countries. Am J Sci Med Res. 2023;9(3):1-10. https://doi.org/10.5281/zenodo.8277996
15. Ha DR, Haste NM, Gluckstein DP. The role of antibiotic stewardship in promoting appropriate antibiotic use. Am J Lifestyle Med. 2019;13(4):376-83. https://doi.org/10.1177/1559827617700824
16. Murray CJ, Ikuta KS, Sharara F, Swetschinski L, Aguilar GR, Gray A, *et al*. Global burden of bacterial antimicrobial resistance in 2019: A systematic analysis. Lancet. 2022;399(10325):629-55. https://doi.org/10.1016/S0140-6736(21)02724-0
17. Laxminarayan R, Duse A, Wattal C, Zaidi AK, Wertheim HF, Sumpradit N, *et al*. Antibiotic resistance-the need for global solutions. Lancet Infect Dis. 2013;13(12):1057-98. https://doi.org/10.1016/S1473-3099(13)70318-9
18. Laxminarayan R, Heymann DL. Challenges of drug resistance in the developing world. BMJ. 2012;344:e1567. https://doi.org/10.1136/bmj.e1567
19. Al-Zalabani A, AlThobyane OA, Alshehri AH, Alrehaili AO, Namankani MO, Aljafri OH. Prevalence of *Klebsiella pneumoniae* antibiotic resistance in Medina, Saudi Arabia, 2014-2018. Cureus. 2020;12(8):e9714.
20. Centers for Disease Control and Prevention (CDC). Antimicrobial Resistance Food and Food Animals. Available from: https://www.cdc.gov/food-safety/foods/antimicrobial-resistance.html [Last accessed on 2024 Feb 20].
21. Fletcher DR, Moffitt LR. Antibiotic resistance in food animals: A global perspective. Int J Antimicrob Agents. 2019;54(4):383-92. https://doi.org/10.1089/fpd.2006.0066
22. Isaacson RE, Torrence ME. The Role of Antibiotics in Agriculture: This Report is based on a Colloquium Sponsored by the American Academy of Microbiology Held November 2-4 2001, in Santa Fe, New Mexico. Washington, DC: American Society for Microbiology; 2002. Available from: https://www.ncbi.nlm.nih.gov/books/nbk559438 [Last accessed on 2024 Jun 28].
23. Shaji S, Selvaraj RK, Shanmugasundaram R. *Salmonella* infection in poultry: A review on the pathogen and control strategies. Microorganisms. 2023;11(11):2814. https://doi.org/10.3390/microorganisms11112814
24. Castro-Vargas RE, Herrera-Sánchez MP, Rodríguez-Hernández R, Rondón-Barragán IS. Castro-Vargas RE, Herrera-Sánchez MP, *et al*. Antibiotic resistance in Salmonella spp. isolated from poultry: A global overview. Vet World. 2020;13:2070-84. doi: 10.14202/vetworld.2020.2070-2084
25. Cardoso MJ, Nicolau AI, Borda D, Nielsen L, Maia RL, Møretrø T, *et al*. *Salmonella* in eggs: From shopping to consumption-a review providing an evidence-based analysis of risk factors. Compr Rev Food Sci Food Saf. 2021;20:2716-41. https://doi.org/10.1111/1541-4337.12753
26. Kapena MS, Muma JB, Mubita CM, Munyeme M. Antimicrobial resistance of *Escherichia coli* and *Salmonella* in raw retail table eggs in Lusaka, Zambia. Vet World. 2020;13(11):2528-33. https://doi.org/10.14202/vetworld.2020.2528-2533
27. Sornplang P, Aieamsaard J, Saksangawong C, Suayroop N. Risk factors associated with *Salmonella* prevalence, its antibiotic resistance, and egg antibiotic residues in the layer farming environment. Vet World. 2022;15(3):543-50. https://doi.org/10.14202/vetworld 2022.543-550
28. Jan S. Scientific opinion on the public health risks of table eggs due to deterioration and development of pathogens. EFSA J. 2014;12(7):3782. https://doi.org/10.2903/j.efsa.2014.3782
29. Li Y, Zhang M, Luo J, Chen J, Wang Q, Lu S, *et al*. Antimicrobial resistance of *Escherichia coli* isolated from retail foods in northern Xinjiang, China. Food Sci Nutr. 2020;8(4):2035-51. https://doi.org/10.1002/fsn3.1491
30. Santhanam R, Luu VT, Weinhold A, Goldberg J, Oh Y, Baldwin IT. Native root-associated bacteria rescue a plant from a sudden-wilt disease that emerged during continuous cropping. Proc Natl Acad Sci U.S.A. 2015;112(36):E5013-20. https://doi.org/10.1073/pnas.1505765112
31. Lamichhane B, Mawad AM, Saleh M, Kelley WG, Harrington PJ 2nd, Lovestad CW, *et al*. Salmonellosis: An overview of epidemiology, pathogenesis, and innovative approaches to mitigate the antimicrobial resistant infections. Antibiotics (Basel). 2024;13(1):76. https://doi.org/10.3390/antibiotics13010076
32. Huang H, Naushad S, Ogunremi D, Kuria J. *Salmonella* - Perspectives for Low-Cost Prevention, Control and Treatment. London: IntechOpen; 2024. https://doi.org/10.5772/intechopen.102150
33. Lupia C, Castagna F, Bava R, Naturale MD, Zicarelli L, Marrelli M, *et al*. Use of essential oils to counteract the phenomena of antimicrobial resistance in livestock species. Antibiotics (Basel). 2024;13(2):163. https://doi.org/10.3390/antibiotics13020163
34. Liu YC, Chen TH, Wu YC, Tan FJ. Determination of the quality of stripe-marked and cracked eggs during storage. Asian-Australas J Anim Sci. 2017;30(7):1013.
35. Al-Gheethi A, Ma NL, Rupani PF, Sultana N, Yaakob MA, Mohamed RM, *et al*. Biowastes of slaughterhouses and wet markets: An overview of waste management for disease prevention. Environ Sci Pollut Res Int. 2021;30:71780-93. https://doi.org/10.1007/s11356-021-16629-w
36. Pesavento G, Calonico C, Runfola M, Nostro AL. Free-range and organic farming: Eggshell contamination by mesophilic bacteria and unusual pathogens. J Appl Poult Res. 2017;26(4):509-17. https://doi.org/10.3382/japr/pfx023
37. Edema MO, Atayese AO. Bacteriological quality of cracked eggs sold for consumption in Abeokuta, Nigeria. Int J Poult Sci. 2006;5(8):772-5. https://doi.org/10.3923/ijps.2006.772.775
38. Saelens G, Robertson L, Gabriël S. Diagnostic tools for the detection of taeniid eggs in different environmental matrices: A systematic review. Food Waterborne Parasitol. 2022;26:e00145. https://doi.org/10.1016/j.fawpar.2022.e00145
39. Washburn KW. Incidence, cause, and prevention of egg shell breakage in commercial production. Poult Sci. 1982;61(10):2005-12. https://doi.org/10.3382/ps.0612005
40. Kulshreshtha G, Benavides-Reyes C, Rodriguez-Navarro AB, Diep T, Hincke MT. Impact of different layer housing systems on eggshell cuticle quality and *Salmonella* adherence in table eggs. Foods. 2021;10:2559. https://doi.org/10.3390/foods10112559
41. Bain MM. Recent advances in the assessment of eggshell quality and their future application. World's Poult Sci J. 2005;61(2):268-77. https://doi.org/10.1079/WPS200459
42. Spitzer H. An Analysis of Bacterial Contamination of Chicken Eggs and Antimicrobial Resistance. Celebrating Scholarship Creativity Day (2011-2017); 2015. Available from: https://digitalcommons.csbsju.edu/elce-cscday/77 [Last accessed on 2024 Feb 20].
43. Lima É, Oliveira MB, Freitas A. Antibiotics in intensive egg production: Food safety tools to ensure regulatory compliance. Food Chem Adv. 2023;3:100548 https://doi.org/10.1016/j.focha.2023.100548
44. Moraes DM, Almeida AM, Andrade MA, Nascente ED, Duarte SC, Nunes IA, *et al*. Antibiotic resistance profile of *Salmonella* sp. Isolates from commercial laying hen farms in central-western brazil. Microorganisms. 2024;12:669. https://doi.org/10.3390/microorganisms12040669
45. Islam M, Sabrin MS, Kabir MH, Aftabuzzaman M. Antibiotic sensitivity and resistant pattern of bacteria isolated from table eggs of commercial layers considering food safety issue. Asian J Med Biol Res. 2018;4(4):323-29.

46. Chadha A, Bhardwaj P, Selvaraj SK, Arasu K, Praveena S, Pavan A, *et al*. Emerging trends in antimicrobial resistance in foodborne pathogens: An overview. J Food Prot. 2020;83(3):535-48. https://doi.org/10.1155/2022/2745416
47. Melebari M, Alpakistany T, Taha T, Alsalman AS. Prevalence of pathogenic bacteria on face masks from wet markets in Makkah duringthe COVID-19 pandemic. Sci J King Faisal University Basic Appl Sci. 2022;23(2):33-8. https://doi.org/10.37575/b/sci/220040
48. Dadar M, Milad A, Zorriehzahra MJ. Emerging of a New Antibiotic Resistance Bacteria Pathogen, Acinetobacter Lwoffii in Water Environments and their Pathogenic Effects. The 17th International and Iranian Congress of Microbiology. 2016.
49. Aghaalikhani E, Ahmadi H. *In vitro* antimicrobial susceptibility of bacterial isolates from chicken eggs in Faisalabad. Arch Anesthesiol Crit Care. 2018;4(4):527-34.
50. Adabara NU, Amarachi1 CE, Adedeji1 AS, Usman A, Maude M, Sadiq FU, *et al*. Study of antibiotics resistance in bacteria isolated from retailed eggshell in three major markets in Minna, Nigeria. Nig J Pure Appl Sci. 2020;33(2):3709-19. https://doi.org/10.48198/njpas/20.a03
51. Jones DR, Anderson KE, Musgrove MT. Comparison of environmental and egg microbiology associated with conventional and free-range laying hen management. Poult Sci. 2011;90(9):2063-8.
52. Sineque CI, Machado AC, Macuamule CJ, Ngowi HA. High levels of bacterial and antimicrobial drug residues contamination in chicken eggs for human consumption in Morogoro municipality, Tanzania. Food Res. 2022;6:124-35. https://doi.org/10.26656/fr.2017.6(6).725
53. Umali DV, Lapuz RR, Suzuki T, Shirota K, Katoh H. Transmission and shedding patterns of *Salmonella* in naturally infected captive wild roof rats (*Rattus rattus*) from a *Salmonella*-contaminated layer farm. Avian Dis. 2012;56(2):288-94. https://doi.org/10.1637/9911-090411-Reg.1
54. Hessel CT, De Oliveira Elias S, Pessoa JP, Zanin LM, Stedefeldt E, Tondo EC. Food safety behavior and handling practices during purchase, preparation, storage and consumption of chicken meat and eggs. Food Res Int. 2019;125:108631.
55. Jones DR, Anderson KE, Guard JY. Prevalence of coliforms, *Salmonella*, *Listeria*, and campylobacter associated with eggs and the environment of conventional cage and free-range egg production. Poult Sci. 2012;91(5):1195-202.
56. Ullah R, Afridi M, Rehman MU, Hasan TN, Tahir M, Khan HA, *et al*. Antimicrobial resistance in *Escherichia coli* and *salmonella* spp. Isolated from table eggs shells and its contents sold at different markets in Peshawar, Pakistan. Microbes Infect Dis. 2023;4:968-75.
57. Fardous J, Shamsuzzaman SM. Detection of potential pathogenic aerobic bacteria from egg shell and egg contents of hen collected from poultry. Bangladesh Med Res Counc Bull. 2015;41(2):67-72.
58. Hai D, Yin X, Lu Z, Lv F, Zhao H, Bie X. Occurrence, drug resistance, and virulence genes of *Salmonella* isolated from chicken and eggs. Food Control. 2020;113:107109.
59. Melebari M. Incidence of potential pathogenic bacteria at restaurants in Al-Mandaq City, Saudi Arabia: First study. J Pure Appl Microbiol. 2023;17(3):1916-25. https://doi.org/10.22207/JPAM.17.3.57
60. Melebari M. Detection of potentially pathogenic bacteria in butcher shops: First report from al Mandaq City, Saudi Arabia. Polish J Natl Sci. 2023;38(1):31-42. https://doi.org/10.31648/pjns.8607
61. Alpakistany T, Taha TM, Gazi KS, Thabet MA, Hroobi AA, Melebari M. β-lactamases-dependent antimicrobial resistance in enterobacteria isolated from commercial poultry farms in the Makkah province, Saudi Arabia. J App Biol Biotech. 2024;12(2):231-8. https://doi.org/10.7324/JABB.2024.150557
62. Dagnew B, Alemayehu H, Medhin G, Eguale T. Prevalence and antimicrobial susceptibility of *Salmonella* in poultry farms and in-contact humans in Adama and Modjo towns, Ethiopia. MicrobiologyOpen. 2020;9:e1067. https://doi.org/10.1002/mbo3.1067
63. Fagbamila IO, Barco L, Mancin M, Kwaga J, Ngulukun SS, Zavagnin P, *et al*. *Salmonella* serovars and their distribution in Nigerian commercial chicken layer farms. PLoS One. 2017;12(3):e0173097. https://doi.org/10.1371/journal.pone.0173097
64. Ketema L, Ketema Z, Kiflu B, Alemayehu H, Terefe Y, Ibrahim M, *et al*. Prevalence and antimicrobial susceptibility profile of *Salmonella* serovars isolated from slaughtered cattle in Addis Ababa, Ethiopia. Biomed Res Int. 2018;2018:9794869. https://doi.org/10.1155/2018/9794869
65. Ziyate N, Karraouan B, Kadiri A, Darkaoui S, Soulaymani A, Bouchrif B. Prevalence and antimicrobial resistance of *Salmonella* isolates in Moroccan laying hens farms. J Appl Poult Res. 2016;25(4):539-46. https://doi.org/10.3382/japr/pfw036

How to cite this article:
Alharthi H, Al-Ghamdi G, Alpakistany T, Almiman B, Alghamdi AM, Albaldi FO, Almimoni NM, Dosh M, Thabet MA, Al-Zahrani KS, Mohammed TA, Melebari M, Gazi KS. A novel multidrug-resistant *Salmonella enterica* strain (A10) isolated from eggshells in wet markets of Taif, Saudi Arabia. J Appl Biol Biotech 2025;13(6):134-141.
DOI: 10.7324/JABB.2025.v13.i6.16