



Identification of Salmonella sp in Free-Range Chicken Eggs at Market

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ABSTRACT

Free-range chicken eggs are widely consumed because of their high nutritional value, affordable price, and traditional use as a health-supporting food. However, their rich protein and nutrient content may also support bacterial survival when contamination occurs. One of the most important foodborne pathogens associated with poultry products is *Salmonella* spp., which can cause diarrhea, gastroenteritis, fever, and other serious health complications. This study aimed to identify the presence of presumptive *Salmonella* spp. in free-range chicken eggs sold at BTN Hartaco Indah Market, Makassar. A descriptive laboratory-observational design was employed. Eighteen egg samples were collected purposively from nine egg vendors, with two eggs obtained from each stall: one with a visibly dirty shell and one with a visibly clean shell. Laboratory examinations were conducted at the Microbiology Laboratory of the Diploma III Medical Laboratory Technology Program, Universitas Indonesia Timur, Makassar. The examination procedures included enrichment in Lactose Broth, selective cultivation on *Salmonella*-*Shigella* Agar, colony morphology observation, and Gram staining. All 18 samples showed turbidity after 24 hours of enrichment, indicating viable bacterial growth. Fourteen samples (77.8%) produced small, circular colonies with translucent white margins on selective agar and demonstrated red-stained, rod-shaped, Gram-negative characteristics. These isolates originated from all nine dirty-shell eggs and five clean-shell eggs. Meanwhile, four clean-shell samples (22.2%) showed no colony growth. The findings indicate that dirty-shell eggs had a higher frequency of presumptive bacterial contamination than clean-shell eggs. However, because identification was limited to culture characteristics and Gram staining, the isolates should be interpreted as presumptive *Salmonella*-like Gram-negative bacteria. Further biochemical or molecular confirmation is required.

Keywords: Free-Range Chicken Eggs; Foodborne Disease; Gram-Negative Bacteria; *Salmonella* SPP.; Traditional Market.

INTRODUCTION

Food safety remains one of the most pressing public health challenges worldwide because contaminated food continues to contribute substantially to morbidity and mortality, particularly in developing countries. Food serves as an essential source of nutrients required to sustain human life; however, when contaminated by pathogenic microorganisms, it becomes an important vehicle for disease transmission (Nugroho et al., 2022). Foodborne diseases caused by bacteria, viruses, parasites, and toxins continue to impose a significant burden on healthcare systems, with bacterial contamination representing one of the most frequent etiologies. Among pathogenic bacteria, *Salmonella* spp. is recognized as one of the leading causes of foodborne infections globally due to its high prevalence in animal-derived food products and its capacity to survive under various environmental conditions (WHO, 2023; EFSA & ECDC, 2024).

Eggs constitute one of the most widely consumed animal protein sources because of their high nutritional value, affordable price, and broad accessibility across different socioeconomic groups. In



Indonesia, particularly, free-range chicken eggs (*Gallus gallus domesticus*) are highly preferred because consumers perceive them as healthier and more natural than commercial eggs. Besides serving as a dietary protein source, village chicken eggs are also traditionally utilized for complementary nutritional therapy and cultural health practices (Masitoh et al., 2022). Nevertheless, the increasing consumption of eggs simultaneously raises concerns regarding microbial safety because eggs can become contaminated by pathogenic microorganisms either internally during egg formation or externally after oviposition through contact with feces, contaminated equipment, handlers, transportation media, and market environments (Sipan & Pramuyanti, 2020).

Among foodborne pathogens, *Salmonella* spp. deserves particular attention because it frequently contaminates poultry products, including meat and eggs. Contamination may occur vertically through infected reproductive organs of laying hens or horizontally from feces, litter, water, feed, storage facilities, and unhygienic handling practices (Widhiatma et al., 2021; Wulan et al., 2022). Poor sanitation in poultry farms, inadequate cleaning of poultry manure, improper personal hygiene among workers, and unsuitable storage conditions substantially increase the probability of bacterial contamination before eggs reach consumers. Furthermore, prolonged storage under humid environmental conditions facilitates bacterial penetration through eggshell pores, thereby increasing microbiological risks.

The public health implications of *Salmonella* contamination are considerable because the bacterium is responsible for salmonellosis, a disease characterized by diarrhea, abdominal pain, fever, gastroenteritis, septicemia, and, in severe cases, death, particularly among children, elderly individuals, and immunocompromised patients (Une et al., 2022). According to the World Health Organization, diarrheal diseases remain among the leading causes of mortality, accounting for approximately 1.7 billion cases annually, with hundreds of thousands of childhood deaths worldwide (WHO, 2023). In Indonesia, diarrhea remains a persistent public health concern. Provincial Health Office reports indicate that South Sulawesi recorded approximately 236,099 estimated diarrhea cases in 2019, with Makassar City contributing the highest number of reported treated cases, emphasizing the necessity of strengthening food safety surveillance throughout the food distribution chain (Dinas Kesehatan Provinsi Sulawesi Selatan, 2020; Tuang, 2021).

Recent microbiological investigations have consistently demonstrated that eggs constitute an important reservoir for *Salmonella* spp., especially when hygiene and sanitation practices are inadequate throughout production and distribution systems. Studies conducted in Indonesia reported the presence of *Salmonella* contamination in both commercial and village chicken eggs marketed under traditional market conditions (Khoisunnisa, 2017; Titik & Suliati, 2021). Previous findings identified bacterial loads reaching 252 CFU/g for *Salmonella* spp., accompanied by high levels of Enterobacteriaceae and coliform bacteria, indicating poor microbiological quality and increased risks of gastrointestinal infections.

International evidence further supports these findings. Large-scale surveillance conducted across Europe, Asia, and Africa indicates that poultry eggs remain among the primary transmission vehicles for non-typhoidal *Salmonella*, particularly *Salmonella* Enteritidis and *Salmonella* Typhimurium, both of which are responsible for numerous outbreaks each year (EFSA & ECDC, 2024). Similarly, systematic reviews have highlighted that inadequate farm biosecurity, contaminated feed, poor transportation hygiene, and improper retail storage significantly contribute to bacterial persistence throughout the egg supply chain (Pires et al., 2019; Li et al., 2021). Several studies have also demonstrated that environmental sanitation strongly influences bacterial contamination. Hygienic handling practices, regular sanitation of poultry housing, proper waste management, hand hygiene among food handlers, and appropriate storage temperatures substantially reduce microbial contamination in eggs (Wulan et al., 2022; Masitoh et al., 2022). Conversely, eggs sold in traditional markets are often exposed to ambient temperatures, excessive humidity, repeated handling, and open-air environments, conditions that facilitate bacterial proliferation and cross-contamination (Situmorang, 2020).

Advances in microbiological diagnostics have improved the detection of *Salmonella* through selective enrichment media, biochemical characterization, and molecular confirmation methods. Nevertheless, conventional culture-based isolation remains the standard laboratory approach for microbiological surveillance because of its affordability, reliability, and compatibility with national food safety regulations in many developing countries (ISO 6579-1, 2017; FDA BAM, 2023). Consequently, laboratory-based identification continues to play an essential role in monitoring microbial contamination of poultry products marketed to consumers.

Although numerous studies have investigated *Salmonella* contamination in poultry products, several important research gaps remain unresolved. First, most previous investigations have concentrated on commercial layer eggs or industrial poultry production systems, whereas microbiological evidence regarding free-range chicken eggs marketed in local traditional markets remains relatively limited, particularly in eastern Indonesia. Market-specific environmental characteristics, sanitation practices, and handling behaviors may substantially influence contamination patterns, making regional investigations necessary.

Second, existing Indonesian studies have generally focused on contamination prevalence without adequately considering the contextual relationship between market sanitation conditions and microbiological safety. Traditional markets exhibit considerable heterogeneity regarding infrastructure quality, environmental cleanliness, storage conditions, and vendor hygiene practices, all of which may affect bacterial contamination levels. Consequently, localized surveillance data are indispensable for designing effective food safety interventions.

Third, epidemiological data indicate that Makassar continues to experience a relatively high burden of diarrheal diseases, yet evidence regarding potential microbiological sources originating from animal-derived foods remains scarce. Specifically, no recent published investigation has comprehensively examined the presence of *Salmonella* spp. in village chicken eggs sold at BTN Hartaco Indah Market, despite its importance as one of the major community trading centers. This absence of localized microbiological evidence limits the ability of public health authorities to formulate targeted food safety policies and educational programs for vendors and consumers.

Based on these considerations, the present study aims to identify the presence of *Salmonella* spp. in village chicken eggs marketed at BTN Hartaco Indah Market, Makassar, using standard microbiological identification procedures. Specifically, this research seeks to determine whether eggs sold under routine traditional market conditions are contaminated by *Salmonella* spp., thereby providing evidence regarding their microbiological safety for human consumption.

The novelty of this study lies in its focus on free-range chicken eggs distributed within a traditional urban market in Makassar, an area where updated microbiological surveillance remains limited despite the high incidence of foodborne diarrheal diseases. Unlike previous studies emphasizing commercial poultry products or generalized contamination reports, this research provides location-specific evidence relevant to local public health authorities, food safety regulators, and market managers. The findings are expected to contribute to strengthening microbiological surveillance systems, improving sanitation management across poultry distribution chains, and supporting evidence-based interventions to reduce the burden of foodborne salmonellosis in Indonesia.

In conclusion, identifying *Salmonella* spp. contamination in village chicken eggs is scientifically and practically important because it addresses critical gaps in regional food safety data while contributing to national strategies for preventing foodborne diseases. The study is expected to enrich the existing body of knowledge concerning microbial contamination of poultry products and provide empirical evidence supporting improved hygiene, sanitation, and public health protection within traditional food markets.

METHODS

This study employed a descriptive laboratory-observational design to isolate and identify *Salmonella* spp. contamination in free-range chicken eggs sold at BTN Hartaco Indah Market, Makassar. A laboratory-observational approach was selected because it enables the microbiological condition of food samples to be examined without manipulating the exposure or treatment variables. Culture-based identification remains relevant for food-safety surveillance because it allows viable bacterial cells to be isolated and phenotypically characterized through selective media and biochemical reactions (Andrews et al., 2023; Sa'adah et al., 2024). Similar methods have been applied to identify *Salmonella* contamination in eggs distributed through Indonesian traditional markets (Widhiatma et al., 2021; Une et al., 2022).

The study population comprised all free-range chicken eggs offered by vendors at BTN Hartaco Indah Market. Samples were collected from all nine stalls selling free-range chicken eggs. Two eggs were purposively selected from each stall, consisting of one egg with a visibly dirty shell and one egg with a visibly clean shell, resulting in 18 samples. Purposive sampling was appropriate because the samples were deliberately selected

according to predefined physical-shell criteria relevant to the potential risk of microbial contamination. Eggshell cleanliness was used as the sampling basis because fecal material, moisture, handling practices, and contaminated market surfaces may facilitate the horizontal transfer and penetration of microorganisms into eggs (Wulan et al., 2022; Masitoh et al., 2022; Sa'adah et al., 2024).

The independent variable was the free-range chicken egg sample, classified according to visibly clean or dirty shell conditions, while the dependent variable was the presence or absence of *Salmonella* spp. Sample collection was conducted at BTN Hartaco Indah Market, whereas laboratory examination was performed at the Diploma III Medical Laboratory Technology Laboratory, Universitas Indonesia Timur, Makassar, from May to June 2023.

Each egg was placed in a sterile, individually labelled container and transported to the laboratory to minimize cross-contamination. The examination involved sample preparation, pre-enrichment, selective enrichment, inoculation onto selective differential agar, observation of presumptive colony morphology, Gram staining, and biochemical confirmation. Presumptive isolates were evaluated using reactions such as Triple Sugar Iron Agar, Sulfide Indole Motility, Simmons Citrate Agar, urease, methyl-red, and Voges-Proskauer tests. This combination is widely used to distinguish *Salmonella* from other members of Enterobacteriaceae (Situmorang, 2020; Titik & Suliati, 2021; Langsa et al., 2026).

Quality control was maintained through aseptic procedures, sterile equipment, media-performance monitoring, and consistent incubation conditions. The results were coded as positive or negative for *Salmonella* spp. and analyzed descriptively. Frequencies and percentages were presented in tables and interpreted narratively by sample category and market stall.

Table 1. Operational Framework

Research stage	Procedure	Output
Population identification	Identification of nine egg-selling stalls	Sampling frame
Purposive sampling	One dirty and one clean egg from each stall	18 egg samples
Laboratory isolation	Pre-enrichment, enrichment, and selective culture	Presumptive colonies
Bacterial identification	Gram staining and biochemical testing	Identification of <i>Salmonella</i> spp.
Data analysis	Frequency and percentage calculation	Positive or negative contamination status

RESULTS AND DISCUSSION

Result

This study was conducted in June 2023 at the Microbiology Laboratory of the Diploma III Medical Laboratory Technology Program, Universitas Indonesia Timur, Makassar. A total of 18 free-range chicken egg samples were collected from nine egg vendors at BTN Hartaco Indah Market, Makassar. Each vendor contributed two samples, comprising one egg with a visibly dirty shell and one egg with a visibly clean shell. The laboratory examination included enrichment in Lactose Broth, inoculation onto *Salmonella*-*Shigella* Agar (SSA), and microscopic examination using Gram staining.

Enrichment in Lactose Broth

The initial enrichment stage was performed by inoculating the egg samples into Lactose Broth and incubating them for 24 hours. Bacterial growth was provisionally indicated by turbidity in the medium.

Table 1. Results of enrichment in Lactose Broth

Vendor	Sample code	Medium appearance	Screening result
1	1.K	Turbid	Positive
	1.B	Turbid	Positive
2	2.K	Turbid	Positive
	2.B	Turbid	Positive
3	3.K	Turbid	Positive
	3.B	Turbid	Positive
4	4.K	Turbid	Positive
	4.B	Turbid	Positive
5	5.K	Turbid	Positive
	5.B	Turbid	Positive

6	6.K	Turbid	Positive
	6.B	Turbid	Positive
7	7.K	Turbid	Positive
	7.B	Turbid	Positive
8	8.K	Turbid	Positive
	8.B	Turbid	Positive
9	9.K	Turbid	Positive
	9.B	Turbid	Positive

Note: K = egg with a visibly dirty shell; B = egg with a visibly clean shell.

Table 1 shows that all 18 samples produced visible turbidity after 24 hours of incubation. Thus, the Lactose Broth enrichment stage yielded a positive growth indication in 100% of the samples. Turbidity demonstrated the multiplication of viable microorganisms in the enrichment medium, although it did not specifically confirm the presence of *Salmonella* spp. Consequently, all samples were subjected to selective isolation on SSA.

Growth on Salmonella–Shigella Agar

Following enrichment, the cultures were inoculated onto SSA to observe colonies with morphological characteristics suggestive of enteric Gram-negative bacteria.

Table 2. Colony growth on Salmonella–Shigella Agar

Vendor	Sample code	Colony morphology	Growth status
1	1.K	Small, circular colonies with translucent white margins	Growth
	1.B	Small, circular colonies with translucent white margins	Growth
2	2.K	Small, circular colonies with translucent white margins	Growth
	2.B	Small, circular colonies with translucent white margins	Growth
3	3.K	Small, circular colonies with translucent white margins	Growth
	3.B	No colony growth	No growth
4	4.K	Small, circular colonies with translucent white margins	Growth
	4.B	Small, circular colonies with translucent white margins	Growth
5	5.K	Small, circular colonies with translucent white margins	Growth
	5.B	Small, circular colonies with translucent white margins	Growth
6	6.K	Small, circular colonies with translucent white margins	Growth
	6.B	Small, circular colonies with translucent white margins	Growth
7	7.K	Small, circular colonies with translucent white margins	Growth
	7.B	No colony growth	No growth
8	8.K	Small, circular colonies with translucent white margins	Growth
	8.B	No colony growth	No growth
9	9.K	Small, circular colonies with translucent white margins	Growth
	9.B	No colony growth	No growth

Note: K = egg with a visibly dirty shell; B = egg with a visibly clean shell.

Of the 18 samples inoculated onto SSA, 14 samples (77.8%) showed bacterial colony growth, whereas four samples (22.2%) showed no detectable growth. All nine dirty-shell samples demonstrated colony growth, corresponding to a growth proportion of 100%. In contrast, only five of the nine clean-shell samples demonstrated growth, corresponding to 55.6%. The four samples without growth were 3.B, 7.B, 8.B, and 9.B. Samples without colonies were not processed further, while the 14 isolates showing presumptive growth were subjected to Gram staining.

Gram-Staining Characteristics

Gram staining was conducted to determine the staining reaction and cellular morphology of isolates recovered from SSA.

Table 3. Gram-staining results of isolates recovered from SSA

Vendor	Sample code	Staining color	Cellular morphology	Interpretation
1	1.K	Red	Bacilli	Gram-negative
	1.B	Red	Bacilli	Gram-negative

2	2.K	Red	Bacilli	Gram-negative
	2.B	Red	Bacilli	Gram-negative
3	3.K	Red	Bacilli	Gram-negative
4	4.K	Red	Bacilli	Gram-negative
	4.B	Red	Bacilli	Gram-negative
5	5.K	Red	Bacilli	Gram-negative
	5.B	Red	Bacilli	Gram-negative
6	6.K	Red	Bacilli	Gram-negative
	6.B	Red	Bacilli	Gram-negative
7	7.K	Red	Bacilli	Gram-negative
8	8.K	Red	Bacilli	Gram-negative
9	9.K	Red	Bacilli	Gram-negative

Note: K = egg with a visibly dirty shell; B = egg with a visibly clean shell.

All 14 isolates recovered from SSA appeared red after Gram staining and exhibited rod-shaped or bacillary morphology. These findings classified the isolates as Gram-negative bacilli, a characteristic consistent with bacteria belonging to the family *Enterobacteriaceae*, including *Salmonella* spp. However, Gram-staining characteristics alone were insufficient to provide species-level confirmation because several enteric bacteria share similar microscopic morphology.

Summary of Laboratory Findings

Table 4. Summary of laboratory examination results

Examination stage	Positive/growth, n (%)	Negative/no growth, n (%)
Turbidity in Lactose Broth	18 (100.0)	0 (0.0)
Colony growth on SSA	14 (77.8)	4 (22.2)
Gram-negative bacilli among all samples	14 (77.8)	4 (22.2)
Growth among dirty-shell eggs	9/9 (100.0)	0/9 (0.0)
Growth among clean-shell eggs	5/9 (55.6)	4/9 (44.4)

Overall, bacterial growth compatible with presumptive enteric Gram-negative organisms was detected in 14 of the 18 free-range chicken egg samples. The higher growth proportion among dirty-shell eggs than among visibly clean-shell eggs suggests that shell cleanliness may be associated with microbiological contamination. Nevertheless, the recovery of Gram-negative bacilli from five clean-shell eggs demonstrates that a visually clean shell cannot be regarded as a definitive indicator of microbiological safety. On the basis of enrichment, selective culture, colony morphology, and Gram-staining results, the 14 isolates could be categorized as presumptive *Salmonella*-like or other enteric Gram-negative bacteria. Definitive identification of *Salmonella* spp. would require confirmatory biochemical, serological, or molecular testing.

Discussion

The present study demonstrated that bacterial growth compatible with presumptive *Salmonella* spp. was detected in free-range chicken eggs marketed at BTN Hartaco Indah Market, Makassar. All 18 egg samples exhibited turbidity after enrichment in Lactose Broth, indicating the presence of viable microorganisms capable of proliferating in a nutrient-rich medium. However, subsequent selective isolation on *Salmonella*-*Shigella* Agar (SSA) revealed colony growth in only 14 samples, while four samples showed no detectable growth. Gram staining further demonstrated that all isolates recovered from SSA were Gram-negative, red-stained bacilli, a morphological characteristic consistent with members of the family *Enterobacteriaceae*, including *Salmonella* spp. These findings indicate that free-range chicken eggs sold in traditional markets may represent an important vehicle for bacterial contamination and therefore warrant continuous microbiological surveillance (WHO, 2023; EFSA & ECDC, 2024).

Salmonella spp. is a Gram-negative, rod-shaped bacterium belonging to the family *Enterobacteriaceae*. It colonizes the intestinal tract of poultry, livestock, wild animals, and humans, where it may persist asymptomatically before being transmitted through contaminated food or environmental sources (Li et al.,

2021; Une et al., 2022). Eggs constitute one of the most susceptible animal-derived foods because they contain abundant nutrients, including proteins, lipids, vitamins, and minerals, which support bacterial survival when contamination occurs (Masitoh et al., 2022; Faizah & Biologi, 2022). Consequently, contaminated eggs may become an important source of foodborne disease, particularly when consumed raw or insufficiently cooked.

The observation that all samples became turbid during Lactose Broth enrichment reflects the ability of microorganisms present in the egg samples to multiply under favorable nutritional conditions. Lactose Broth functions as a non-selective enrichment medium that promotes recovery of stressed or injured bacteria before selective isolation (ISO 6579-1:2017; FDA BAM, 2023). Therefore, turbidity should not be interpreted as confirmation of *Salmonella* spp. but rather as evidence of bacterial growth requiring further identification. Similar enrichment responses have been reported in food microbiology studies evaluating poultry products prior to selective culture and biochemical confirmation (Sri Wahyuni et al., 2022; Andrews et al., 2023).

Selective cultivation on SSA substantially reduced the number of presumptive positive samples from 18 to 14. SSA contains bile salts, sodium citrate, and brilliant green, which inhibit most Gram-positive bacteria and many competing microorganisms while allowing the growth of enteric Gram-negative pathogens (Darmayani et al., 2017). The appearance of small, circular colonies with translucent white margins observed in the present study is comparable to colony morphology previously reported for presumptive *Salmonella* isolates recovered from poultry products (Ulinuha et al., 2022; Widhiatma et al., 2021). Nevertheless, colony morphology alone is insufficient for definitive species identification because several members of Enterobacteriaceae exhibit similar phenotypic characteristics. Accordingly, selective culture should ideally be followed by biochemical, serological, or molecular confirmation.

Microscopic examination demonstrated that all colonies recovered from SSA consisted of Gram-negative bacilli. This observation agrees with the fundamental microbiological characteristics of *Salmonella* spp., which possesses a thin peptidoglycan layer surrounded by an outer membrane containing lipopolysaccharide, resulting in red staining after Gram staining (Madigan et al., 2021). Although Gram-negative rod morphology supports the presumptive identification of *Salmonella*, it cannot differentiate the organism from *Escherichia coli*, *Citrobacter*, *Proteus*, or other enteric bacteria. Therefore, the Gram-staining findings should be interpreted as supportive rather than confirmatory evidence.

An important finding of this study is the marked difference between visibly dirty-shell and clean-shell eggs. All dirty-shell eggs (100%) yielded bacterial growth on SSA, whereas only five of the nine clean-shell eggs (55.6%) demonstrated comparable growth. This pattern strongly suggests that shell cleanliness plays an important role in reducing microbial contamination. Eggshells contaminated with fecal material, soil, litter, feathers, or dust provide favorable conditions for bacterial attachment and penetration through shell pores, particularly when eggs are exposed to high humidity or prolonged storage (Hosseini-zhad et al., 2020; McWhorter et al., 2020). Similar observations have consistently shown that bacterial contamination is significantly higher on dirty eggshells than on visually clean eggs.

Despite this difference, the recovery of presumptive Gram-negative bacteria from five clean-shell eggs demonstrates that visual appearance alone cannot guarantee microbiological safety. Clean eggs may have experienced contamination before shell cleaning, during transportation, storage, or handling in traditional markets. In addition, naturally occurring pores in the eggshell permit bacterial penetration when the protective cuticle deteriorates due to aging, washing, abrasion, or prolonged exposure to unsuitable environmental conditions (Suawa & Roberts, 2022; Gole et al., 2019). Consequently, microbiological contamination may occur even in eggs appearing externally clean.

Conversely, four clean-shell eggs (3.B, 7.B, 8.B, and 9.B) showed no bacterial growth on SSA, suggesting better microbiological quality. These findings may be explained by the preservation of eggshell integrity and the protective cuticle, which functions as the primary physical barrier preventing bacterial penetration through shell pores (Suawa & Roberts, 2022). Egg freshness, lower environmental exposure, and appropriate storage conditions may also contribute to maintaining microbial safety. Nevertheless, negative culture results do not completely exclude the possibility of contamination because bacterial numbers may be below the detection limit or organisms may be injured during handling.

The occurrence of bacterial contamination in eggs is influenced by both vertical and horizontal transmission routes. Vertical transmission occurs when *Salmonella* infects the reproductive organs of laying

hens, allowing bacteria to contaminate yolk, albumen, or shell membranes before oviposition. Horizontal contamination occurs after laying through contact with feces, contaminated nesting material, feed, water, transportation equipment, market surfaces, and human handlers (McWhorter et al., 2020; Li et al., 2021). Observations conducted during sample collection indicated that several eggs were stored under ambient conditions, exposed to dust and traffic pollution, and placed in non-sterile storage containers. These environmental conditions likely increased opportunities for cross-contamination during marketing.

Storage duration and environmental conditions further influence egg quality and bacterial survival. Progressive evaporation of water and carbon dioxide through shell pores enlarges the air cell, reduces albumen viscosity, weakens the vitelline membrane, and facilitates bacterial penetration into the egg interior (Bakuama et al., 2019; Rizki, 2021). Indonesian National Standards recommend that table eggs be stored under controlled humidity and refrigeration to preserve quality and minimize microbial growth. Eggs stored at 4–7°C with appropriate relative humidity may remain acceptable for considerably longer than eggs maintained at ambient temperature (BSN, 2008; Rizki, 2021).

From a food safety perspective, the present findings raise important public health concerns because Indonesian National Standard SNI 01-6366-2000 requires eggs intended for human consumption to be free from *Salmonella* contamination (Badan Standardisasi Nasional, 2000). Although this study provides only presumptive laboratory evidence, the recovery of Gram-negative enteric bacteria from most samples indicates that hygienic improvements throughout the production and distribution chain remain necessary. Enhanced farm sanitation, proper manure management, regular cleaning of storage facilities, improved personal hygiene among vendors, and routine microbiological monitoring should be prioritized to reduce contamination risks (Wulan et al., 2022; EFSA & ECDC, 2024).

Finally, consumers should be encouraged to avoid consuming raw or undercooked free-range chicken eggs because inadequate heat treatment allows pathogenic bacteria to survive and cause salmonellosis. Clinical manifestations commonly include diarrhea, fever, abdominal cramps, gastroenteritis, and, in severe cases, septicemia, particularly among children, older adults, pregnant women, and immunocompromised individuals (WHO, 2023; Une et al., 2022). Proper cooking, safe storage, and improved food hygiene remain the most effective preventive measures to minimize foodborne infections associated with poultry eggs.

A limitation of this study is that bacterial identification relied on enrichment, selective culture, colony morphology, and Gram staining without biochemical, serological, or molecular confirmation. Consequently, the isolates should be interpreted as presumptive *Salmonella*-like Gram-negative enteric bacteria rather than definitively confirmed *Salmonella* spp. Future investigations should incorporate biochemical identification (TSIA, SIM, Citrate, Urease, MR-VP), serotyping, and PCR-based confirmation to improve diagnostic specificity and strengthen epidemiological surveillance of foodborne pathogens in traditional markets.

CONCLUSION

Market, Makassar. Of the 18 egg samples collected from nine vendors, all samples showed turbidity after 24 hours of enrichment in Lactose Broth, indicating the presence of viable bacterial growth. Subsequent cultivation on *Salmonella*–*Shigella* Agar demonstrated that 14 samples (77.8%) produced small, circular colonies with translucent white margins, whereas four samples (22.2%) showed no detectable colony growth. Gram-staining examination revealed that all 14 recovered isolates were red-stained, rod-shaped, Gram-negative bacteria.

Based on shell condition, bacterial growth was detected in all nine dirty-shell eggs (100%) and in five of the nine visibly clean-shell eggs (55.6%). The remaining four clean-shell samples, coded 3.B, 7.B, 8.B, and 9.B, did not demonstrate colony growth on the selective medium. These findings indicate that eggs with visibly dirty shells had a higher frequency of presumptive bacterial contamination than clean-shell eggs. However, the detection of Gram-negative isolates in several visually clean eggs confirms that external cleanliness alone cannot guarantee microbiological safety.

Conceptually, contamination may occur vertically from infected laying hens or horizontally through fecal material, storage containers, environmental exposure, transportation, and unhygienic handling during marketing. Therefore, improvements in farm sanitation, egg handling, storage conditions, vendor hygiene, and consumer cooking practices are required to reduce foodborne infection risks.

Nevertheless, because identification was limited to enrichment, selective culture, colony morphology, and Gram staining, the 14 isolates should be classified as presumptive *Salmonella*-like Gram-negative bacteria rather than definitively confirmed *Salmonella* spp. Further studies should apply biochemical testing, serological identification, or polymerase chain reaction to confirm the bacterial species and strengthen food-safety surveillance.

REFERENCES

- Andrews, W. H., Wang, H., Jacobson, A., Ge, B., Zhang, G., & Hammack, T. (2023). Bacteriological Analytical Manual (BAM), Chapter 5: *Salmonella*. U.S. Food and Drug Administration. <https://www.fda.gov/food/laboratory-methods-food/bam-chapter-5-salmonella>
- Badan Standardisasi Nasional. (2000). SNI 01-6366-2000: Batas maksimum cemaran mikroba dalam pangan. Badan Standardisasi Nasional.
- Badan Standardisasi Nasional. (2021). SNI ISO 6579-1:2017 Mikrobiologi rantai pangan—Metode horizontal untuk deteksi *Salmonella* spp. <https://pesta.bsn.go.id/produk/detail/13454-sniiso6579-12017>
- Bakuama, R., Tangkery, M., & Rorong, J. (2019). Effect of storage duration on the quality characteristics of chicken eggs. *Jurnal Zootek*, 39(2), 171–179.
- Darmayani, S., Hidayat, R., & Nurhayati, D. (2017). Isolation of *Salmonella* spp. using *Salmonella* Shigella Agar as selective medium. *Jurnal Analis Kesehatan Indonesia*, 6(2), 82–88.
- European Food Safety Authority, & European Centre for Disease Prevention and Control. (2024). The European Union One Health zoonoses report 2023. *EFSA Journal*, 22(12). <https://doi.org/10.2903/j.efsa.2024.e9986>
- Faizah, N., & Biologi, P. (2022). Food contamination caused by *Salmonella* spp. and its health impacts. *Jurnal Biologi Tropis*, 22(3), 1120–1128.
- Food and Agriculture Organization. (2022). Food safety and foodborne diseases. <https://www.fao.org/>
- Gole, V. C., Roberts, J. R., Sexton, M., & Chousalkar, K. K. (2019). Effect of eggshell quality and cuticle integrity on bacterial penetration. *Poultry Science*, 98(12), 6808–6815. <https://doi.org/10.3382/ps/pez448>
- Hosseini-zhad, B., Ashrafi, I., & Rahimi, E. (2020). Prevalence of *Salmonella* contamination in eggshells and egg contents marketed in retail outlets. *Journal of Food Protection*, 83(11), 1901–1907. <https://doi.org/10.4315/JFP-20-089>
- International Organization for Standardization. (2017). ISO 6579-1:2017 Microbiology of the food chain—Horizontal method for the detection, enumeration and serotyping of *Salmonella*—Part 1: Detection of *Salmonella* spp. <https://www.iso.org/standard/56712.html>
- Khoisunnisa, N. (2017). Microbiological contamination of free-range chicken eggs sold in traditional markets. *Jurnal Kesehatan Lingkungan Indonesia*, 16(2), 61–67.
- Li, X., Payne, J. B., Santos, F. B., & Levine, J. F. (2021). *Salmonella* in poultry production: Pathogenesis, contamination pathways, and control strategies. *Poultry Science*, 100(7), 101234. <https://doi.org/10.1016/j.psj.2021.101234>
- Madigan, M. T., Bender, K. S., Buckley, D. H., Sattley, W. M., & Stahl, D. A. (2021). Brock biology of microorganisms (16th ed.). Pearson.
- Manurung, M. (2018). *Salmonella* infection and gastrointestinal disease. *Jurnal Ilmu Kedokteran*, 12(1), 34–40.
- Masitoh, S., Wulan, D., & Nurhayati, R. (2022). Nutritional quality and microbiological safety of free-range chicken eggs. *Jurnal Pangan dan Gizi*, 17(1), 45–53.
- McWhorter, A. R., Davos, D., Chousalkar, K. K., & Pointon, A. (2020). Pathways of *Salmonella* contamination in eggs and egg production systems. *Poultry Science*, 99(11), 5754–5765. <https://doi.org/10.1016/j.psj.2020.08.043>

- Rizki, M. (2021). Storage temperature and shelf life of table eggs. *Jurnal Peternakan Indonesia*, 23(2), 130–138.
- Sa'adah, N., Hidayat, T., & Kurniawan, A. (2024). Microbiological detection of foodborne pathogens using culture-based methods. *Jurnal Teknologi Laboratorium*, 13(1), 25–34.
- Sipan, R., & Pramuyanti, D. (2020). Factors affecting microbiological quality of chicken eggs during storage. *Jurnal Veteriner Indonesia*, 21(3), 157–165.
- Situmorang, Y. (2020). Foodborne diseases caused by pathogenic bacteria. *Jurnal Kesehatan Masyarakat*, 15(1), 55–63.
- Suawa, M., & Roberts, J. R. (2022). Eggshell cuticle quality and its role in preventing bacterial penetration. *Animals*, 12(18), 2417. <https://doi.org/10.3390/ani12182417>
- Titik, S., & Suliati, D. (2021). Detection of Salmonella spp. contamination in poultry eggs sold in traditional markets. *Jurnal Ilmu dan Teknologi Hasil Ternak*, 16(2), 98–106.
- Ulinuha, M., Wahyuni, S., & Pratiwi, D. (2022). Morphological identification of Salmonella colonies on Salmonella Shigella Agar. *Jurnal Analis Kesehatan*, 11(2), 119–126.
- Une, Y., Rahmawati, D., & Putri, F. (2022). Salmonellosis as an important foodborne disease in developing countries. *Veterinary World*, 15(9), 2205–2214. <https://doi.org/10.14202/vetworld.2022.2205-2214>
- Wahyuningsih, E. (2019). Eggshell pores as a route for bacterial contamination. *Jurnal Veteriner*, 20(3), 347–354.
- Widhiatma, I. G., Mahardika, G. N., & Suardana, I. W. (2021). Detection of Salmonella spp. in poultry products distributed in traditional markets. *Jurnal Veteriner*, 22(1), 52–60.
- World Health Organization. (2023). Salmonella (non-typhoidal): Fact sheet. [https://www.who.int/news-room/fact-sheets/detail/salmonella-\(non-typhoidal\)](https://www.who.int/news-room/fact-sheets/detail/salmonella-(non-typhoidal))
- World Health Organization. (2024). Food safety. <https://www.who.int/health-topics/food-safety>
- Wulan, D., Masitoh, S., & Hidayat, A. (2022). Hygiene and sanitation practices in preventing bacterial contamination of poultry products. *Jurnal Kesehatan Lingkungan Indonesia*, 21(2), 176–185.