Bacteriological and Molecular Characterization of *Salmonella* Species Isolated from Humans and Chickens in Sharkia Governorate, Egypt

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**Abstract**

This study was carried out to investigate the occurrence of *Salmonella* species in 690 different samples collected from Sharkia Governorate, Egypt. The samples comprised of chicken meat, organs, eggs, cloacal swabs and wooden surface swabs from pluck shop outlets and hand swabs from workers. In addition, stool swabs from patients with gastroenteritis attending the Outpatient Clinics at Zagazig and Abo-Hammad cities. *Salmonella* spp. were detected in 5.9%, 2.6%, 4.2%, 10.4%, 10% and 10% of chicken meat, organs, egg shell, cloacal swabs, surface swabs and hand swabs, respectively. The isolation rate from stool swabs was 0.8%. Biochemical Identification revealed that *Salmonella* spp. were identified in 129 out of 690 examined samples (18.7%). Meanwhile, molecular identification using *invA* gene revealed that only 29 *Salmonella* isolates were detected (4.2%). *S. Typhimurium* and *S. Enteritidis* were identified from different samples with the percentages of 1.2 and 1.01, respectively. The other identified *Salmonella* serovars were *S. Newport* (0.9%), *S. Kentucky* (0.7%), and *S. Infantis* (0.4%). Virulence associated genes including *avr A*, *hilA* and *pef* were identified in 100, 91.3 and 10.3% of the examined isolates. In conclusion, a proportion of chicken carcasses and giblets sold in Sharkia Governorate, Egypt was contaminated with *Salmonella* spp. including potentially virulent *S. Typhimurium* and *S. Enteritidis*, posing risk for human consumers.

**Keywords:** *Salmonella*, Serotyping, PCR, *avr A*, *hilA*, *pef*

**Introduction**

*Salmonella* is an important foodborne pathogen with an estimated 1.3 billion incidences of nontyphoidal salmonellosis worldwide annually [1]. More than 2500 *Salmonella* serotypes are recognized, of which, *S. Enteritidis*, *S. Typhimurium* and *S. Kentucky* are identified as the most frequently causative agents causing disease burden on consumers [2]. Poultry is incriminated in many outbreaks of human salmonellosis worldwide, which in turn causes concern to public health [3]. Traditional identification methods including phenotyping and serotyping are time consuming and labor intensive. For these reasons, the use of PCR for identification of *Salmonella* serovars is an attractive alternative to the most traditional techniques. *Salmonella*-specific PCR with primers for the invasion (*invA*) gene is a rapid, sensitive, and specific tool for monitoring *Salmonellae* at the genus level in a variety of clinical samples [4]. The *invA* gene encodes a protein in the inner membrane of bacteria, which is necessary for invasion of epithelial cells of the host [5]. Several reports had also confirmed the successful detection of 100% of *Salmonella* isolates from poultry using specific primers for the *invA* gene with no false positives or negative [6-8]. *Salmonella* spp. had some genes responsible for its virulence, for instance, the *avrA*; is an effector protein of the type three secretion system (TTSS) complex that contributes to the virulence of *Salmonella* spp. by limiting the host’s inflammatory response through the inducement of cell apoptosis, especially macrophages [9, 10]. Also, the hyper invasive locus A (*hilA*) gene is one of the important virulence determinants which is necessary for the expression of the TTSS components required to invade epithelial cells [11, 12]. The plasmid encoded fimbriae (*pefA*) gene plays an important role in the pathogenicity of *Salmonella* spp. because the fimbriae promote their attachment to epithelial cells [13]. This study was planned to estimate the occurrence of *Salmonella* serovars in chickens and humans inhabiting the same areas using bacteriological and molecular methods. *Salmonella* isolates were further characterized by virulence gene profiling, focusing on three virulence determinants associated with SPIs and plasmids that have been shown to be relevant.

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for the success of *Salmonella* as an intracellular pathogen.

**Material and Methods**

**Sampling**

A total of 690 different samples from Abo-Hammad and Zagazig cities, Sharkia Governorate were collected for the isolation of *Salmonella* species. The samples comprised of 152 chicken meat samples (breast and thigh, 76, each), 152 chicken organs (liver and heart, 76, each), 140 egg samples (egg shell and contents, 70, each), 76 cloacal swabs and 20 surface swabs from wooden cutting boards at poultry pluck shop outlets. In addition, 20 hand swabs from poultry workers at the outlets and 130 human stool samples from Outpatient Clinic at Hospitals in Abo-Hammad and Zagazig, Sharkia Governorate, Egypt were collected. The study was conducted during the period from November 2016 to April 2017. The collected samples were immediately transported to the laboratory for bacteriological analysis.

**Isolation and bacteriological identification**

Twenty-five grams from raw chicken meat (breast and thigh) and organs (liver and heart) samples were homogenized in 225 mL of buffered peptone water (BPW, Oxoid) and incubated at 37°C for 24 h for pre-enrichment. Sterile swabs were used to sample the egg shells and then, they were incubated in BPW tubes. After sterilization of the egg shells, it was cracked and the contents were mixed with BPW. All other swab samples (cloacal, hand, stool and surface swabs) were collected in BPW tubes and pre-enriched as previously described.

Isolation of *Salmonella* spp. was carried out according to ISO-6579: 2002 standard [14, 15]. Following pre-enrichment in BPW, a portion (0.1 mL) of the pre-enriched broth was transferred to 10 mL Rappaport-Vassiliadis Soy Peptone (RVS, Oxoid) broth followed by incubation at 41.5°C overnight (18-24 h). Another portion (one mL) from pre-enriched broth was added to 10 mL of Muller-Kauffmann Tetrathionate/Novobiocin broth (Oxoid), and then incubated at 37°C for 24 h. From each enrichment broth, a loopful was streaked onto the surface of Xylose Lysine Desoxycholate (XLD, Oxoid) agar and Hekton Enteric agar (Oxoid) and the plates were incubated at 37°C for 24 h. The suspected colonies were purified and then identified morphologically and biochemically using Gram staining and biochemical screening tests including oxidase test reactions on triple sugar iron agar, indole production, carbon utilization in Simmon’s citrate agar and urea hydrolysis [16].

**Molecular identification of Salmonella spp.**

Identification of genus *Salmonella* was performed using the invasion gene (*invA*) specific primer pairs F-139 5'-GTG AAA TTA TCG CCA CGT TCG GGC AA-3' and R-141 5'-TCA TCG CAC CGT CAA AGG AAC C-3’ which produce 284 bp amplicon size [8]. The reaction was performed in a volume of 25 µL containing 12.5 µL of readymade power Emerald Amp GT PCR Mastermix (Takara), 20 pmol of each primer and 6 µL of the purified DNA. A negative control (no added DNA) and a positive control (*S. Enteritidis*) was also run. The positive control was kindly provided by the Reference Laboratory for Veterinary Quality Control on Poultry Production, Animal Health Research Institutes, Giza.

The reaction conditions consisted of a primary denaturation at 94°C for 5 min, followed by 35 cycles of denaturation at 94°C for 30 sec, annealing at 55°C for 30 sec and extension at 72°C for 30 sec, followed by a final extension at 72°C for 7 min. The PCR products (10 µL) were mixed with loading buffer (3 µL) and loaded in 1.5% agarose gel beside 5 µL of 100 bp DNA ladder (Qiagen, USA). The gels were then run in 1x TBE and 5 uM ethidium bromide (Sigma) for at least 45 min at 100 volts and then visualized using ultraviolet transilluminator (Gel Documentation System, Consort-Belgium).

**Serotyping**

Molecularly confirmed *Salmonella* isolates were subjected to serotyping following the Kauffmann-White Scheme with commercial antisera (Difco Laboratories Detroit) for cell wall (O) and flagellar (H) antigen identification [Kauffmann and Das-Kauffmann, 2001]). Serotyping was carried out at the Bacteriology Department, Faculty of Veterinary Medicine, Benha University.
**Molecular identification of S. Enteritidis and S. Typhimurium**

Molecular confirmation of S. Enteritidis and S. Typhimurium, was further carried out using primer sets specific for S. Typhimurium with the sequences F: 5’-GGT GGC AAG GGA ATG AA-3’ and R: 5’-CGC AGC GTA AAG CAA CT-3’ [17] and S. Enteritidis with the sequences F: 5’-GCA GCG GTT ACT ATT GCA GC-3’ and R: 5’-TGT GAC AGG GAC ATT TAG CG-3’ [18] producing amplicons with molecular weight of 915 bp and 310 bp, respectively. The amplification mixture composition and visualization of the products were as previously described for invA PCR. The reaction conditions for the three genes consisted of a primary denaturation at 94°C for 5 min, followed by 35 cycles of denaturation at 94°C for 30 sec, annealing at 50°C for 1 min and extension at 72°C for 10 min. While, the reaction conditions for S. Enteritidis sefA PCR consisted of a primary denaturation at 94°C for 5 min, followed by 35 cycles of denaturation at 94°C for 30 sec, annealing at 52°C for 30 sec, extension at 72°C for 30 sec and then a final extension at 72°C for 7 min.

**Molecular identification of virulence associated genes**

Molecular detection of three virulence associated genes: hila F: 5’-CAT GGC TGG TCA GTT GGA G-3’ and R: 5’-CGT AAT TCA TCG CCT AAA CG-3’ [19], pefa F: 5’-TGT TTC CGG GCT TGT GCT-3’ and R: 5’-CAG GGC ATT TGC TTA TTC C-3’ [20], and avrA F: 5’-CCT GTA TTG TTG AGC GTC TGG-3’ and R: 5’-AGA AGA AGA GCT TCG TTG AAT GTC C-3’ [21] producing amplicons with molecular sizes of 150 bp, 700 bp and 422 bp, respectively. The reaction mixture composition and visualization were as previously described for invA PCR. The reaction conditions for the three genes consisted of a primary denaturation at 94°C for 5 min, followed by 35 cycles of denaturation at 94°C for 30 sec, annealing at 60°C for 30 sec (hila), 58°C for 45 sec (avra), 55°C for 45 sec (pefa), extension at 72°C for 45 sec and a final extension at 72°C for 10 min.

**Results and Discussion**

**Identification of Salmonella spp.**

Bacteriological examination revealed that 18.7% of the samples under investigation were suspected to be contaminated with Salmonella species (Table 1). However, molecular amplification of the invA gene revealed that only 4.2% were confirmed. In accordance, de Freitas et al. [22] have reported that increased uncertainty in morphological and biochemical testing is attributed to variations among Salmonella strains. The rapidity and higher accuracy of PCR compared to bacteriological typing methods is promising in diagnosis of infections [17, 23].

The invA gene is used as a marker for the molecular identification of the genus [17, 23] and was widely used for the detection of Salmonella spp. in different samples. Consistent with our findings, invA gene was amplified in 100% of Salmonella serovars isolated from chicken and humans in Egypt [6, 24-26].

**Table 1:** Proportion of salmonella isolates identified by bacteriological and molecular testing in different samples

<table>
<thead>
<tr>
<th>Samples</th>
<th>No. of bacteriologically suspected isolates (%)</th>
<th>No. of molecularly confirmed Salmonellae*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chicken Meat (n=152)</td>
<td>30 (19.7%)</td>
<td>9 (5.9%)</td>
</tr>
<tr>
<td>Chicken Organs (n=152)</td>
<td>26 (17.1%)</td>
<td>4 (2.6%)</td>
</tr>
<tr>
<td>Chicken Egg (n=140)</td>
<td>17 (12.1%)</td>
<td>3 (2.1%)</td>
</tr>
<tr>
<td>Cloacal swabs (n=76)</td>
<td>23 (30.3%)</td>
<td>8 (10.5%)</td>
</tr>
<tr>
<td>Surface swabs (n=20)</td>
<td>6 (30%)</td>
<td>2 (10%)</td>
</tr>
<tr>
<td>Hand swabs(n=20)</td>
<td>3 (15%)</td>
<td>2 (10%)</td>
</tr>
<tr>
<td>Stool swabs(n=130)</td>
<td>24 (18.5%)</td>
<td>1 (0.8%)</td>
</tr>
<tr>
<td>Total (n=690)</td>
<td>129 (18.7%)</td>
<td>29 (4.2%)</td>
</tr>
</tbody>
</table>

*Molecular identification was based on amplification of invA gene.*
The most prevalent zoonotic *Salmonella* serovars in the current study (Table 2) upon serotyping were *S. Typhimurium* (1.2%) and *S. Enteritidis* (1.01%), followed by *S. Newport* (0.9%), *S. Kentucky* (0.7%) and *S. Infantis* (0.4%).

Human non-typhoidal salmonellosis is mainly caused by *S. Typhimurium* and *S. Enteritidis* [17, 27]. Different primers specific for molecular identification of *S. Typhimurium* and *S. Enteritidis* were developed to target O-antigens, H1 and H2 antigens [28-30]. At least five primer sets should be used to amplify these antigens, rendering these primers in appropriate for routine diagnosis [17]. Primers targeting *stm*-4495 gene of *S. Typhimurium* are more sensitive and specific for the identification of *S. Typhimurium* [17]. A sequence in *S. Enteritidis* SEF14 fimbral antigen gene (*sefA*) were used for developing primer sets specific for the diagnosis of the serovar [31]. In the current study, *stm*-4495 and *sefA* genes were respectively detected in all *S. Typhimurium* (n=8) and all *S. Enteritidis* (n=7) isolates using corresponding specific primers that produced amplicons with molecular weight of 915 bp and 310 bp, respectively (Figures 1,2). The *sefA* gene was reported in 100% of *S. Enteritidis* isolates from human gastroenteritis cases [31], meat products in Egypt [32] and poultry [33].

![Sample of gel picture showing amplification of 284 bp of invA gene for the molecular identification of *Salmonella* species from different sources. L: 100 bp ladder, lanes 1-8: examined samples, 9: Positive control of *Salmonella* DNA obtained from the Biotechnology Unit, Reference laboratory for Veterinary Quality Control on Poultry Production, Animal Health Research Institute, Dokki, Giza, Egypt, 10: Negative control.](image)

**Occurrence of Salmonella spp. in different samples**

The overall isolation rate of *Salmonella* spp. from chicken meat samples in the current study (Table 2) was 5.9%. Such percentage was nearly similar to 6, 9.8 and 5% previously reported in India [34], China [35] and Egypt [26], respectively. Slightly higher percentages of 10.6 and 11.8 were reported in Croatia [36] and India [37], respectively. However, higher isolation rates of 14% [38] and 44% [39] from chicken meat were reported in Egypt. The difference in prevalence rates between different studies could be attributed to diversity in sampling methods, season and isolation techniques [40].

The results in Table (2) showed the isolation of *Salmonella* spp. from breast (6.6%) and thigh muscles (5.2%). Higher percentages of *Salmonella* spp. isolation from breast meat; 15.4% in Croatia [36], 26% in Egypt [41] and 20% in Egypt [42] were previously reported. While, higher prevalence rates of isolation from thigh muscles; 33.3% [42] and 33% [43] were also documented.
Contamination of poultry meat is usually originated from microorganisms present in the intestinal tract, on skin and feather during feather plucking, evisceration, washing and storage [36]. Although the low contamination rate of chicken meat in the study area, hygienic sanitation methods during poultry rearing, transportation, slaughtering, evisceration, in addition to, cross contamination from cutting boards, knives, equipment and hands of workers, should be applied to minimize public health risk [44].

Salmonella spp. were isolated from 2.6% of liver and heart samples, each (Table 2). Nearly similar percentage of 5.6% for Salmonella spp. in liver samples were previously recorded in Egypt [45]. The percentage of Salmonella spp. isolated from heart samples during the current study was nearly similar to 6% in Iran [46]. In contrary, higher percentages of 48% and 14.1% were reported in Egypt [39] and Iran [40], respectively. The previously reported high isolation of Salmonellae from liver and heart samples could be attributed to the contamination of these organs from the crop and the intestinal contents during evisceration [47].
Table 2: Occurrence of *Salmonella* species in the examined samples

<table>
<thead>
<tr>
<th>Samples</th>
<th>Number examined</th>
<th>S. Typhimurium</th>
<th>S. Enteritidis</th>
<th>S. Kentucky</th>
<th>S. Newport</th>
<th>S. Infantis</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Meat</td>
<td>152</td>
<td>1 (0.7%)</td>
<td>2 (1.3%)</td>
<td>2 (1.3%)</td>
<td>2 (1.3%)</td>
<td>2 (1.3%)</td>
<td>9</td>
</tr>
<tr>
<td>Breast</td>
<td>76</td>
<td>0</td>
<td>2 (2.6%)</td>
<td>1</td>
<td>1 (1.3%)</td>
<td>1</td>
<td>5</td>
</tr>
<tr>
<td>Thigh</td>
<td>76</td>
<td>1</td>
<td>0 (1.3%)</td>
<td>1</td>
<td>1 (1.3%)</td>
<td>1</td>
<td>4</td>
</tr>
<tr>
<td>Organs</td>
<td>152</td>
<td>0</td>
<td>3 (2%)</td>
<td>0</td>
<td>1 (1.3%)</td>
<td>0</td>
<td>4</td>
</tr>
<tr>
<td>Liver</td>
<td>76</td>
<td>0</td>
<td>2 (2.6%)</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>Heart</td>
<td>76</td>
<td>0</td>
<td>1 (1.3%)</td>
<td>0</td>
<td>1 (1.3%)</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>Egg shell</td>
<td>70</td>
<td>1</td>
<td>0 (1.4%)</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td>3</td>
</tr>
<tr>
<td>Egg content</td>
<td>70</td>
<td>0</td>
<td>0 (1.4%)</td>
<td>0</td>
<td>0 (1.4%)</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Cloacal swabs</td>
<td>76</td>
<td>2</td>
<td>1 (2.6%)</td>
<td>2</td>
<td>2 (2.6%)</td>
<td>1</td>
<td>8</td>
</tr>
<tr>
<td>Surface swabs</td>
<td>20</td>
<td>1</td>
<td>1 (5%)</td>
<td>0</td>
<td>0 (5%)</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>Hand swabs</td>
<td>20</td>
<td>2</td>
<td>0 (10%)</td>
<td>0</td>
<td>0 (10%)</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>Stool swabs</td>
<td>130</td>
<td>1</td>
<td>0 (0.8%)</td>
<td>0</td>
<td>0 (0.8%)</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>Total</td>
<td>690</td>
<td>8</td>
<td>7 (1.2%)</td>
<td>5 (1.01%)</td>
<td>6 (0.7%)</td>
<td>3 (0.9%)</td>
<td>29</td>
</tr>
</tbody>
</table>

*Salmonella enterica* could be transmitted to humans following consumption of contaminated eggs produced by infected laying hens. There was an association between the contamination of eggs and egg products with *Salmonella* and human infection [48]. The results shown in Table (2) verified that 4.2% of egg shell samples were contaminated with *Salmonella* species. Comparable isolation rates from egg shell samples were previously recorded in Egypt [49, 50]. However, higher isolation rates of 20% in India [34] and 34% in Spain [51] were documented.

Absence of *Salmonellae* in the contents of eggs (Table 2), was consistent to other findings [51]. In contrary, in India, Nagappa *et al.* [52], and different studies in Egypt [49, 50, 53] detected *Salmonella* spp. in egg content samples. The low and sporadic egg content contamination could be attributed to the protective effect of complex system membrane barriers and the antibacterial effect of egg albumin [51].

The results in Table (2) showed that 10.4% of the examined cloacal swabs were positive for *Salmonella* species. Similarly, in Egypt, Abd El-Ghany *et al.* [45] isolated *Salmonella* spp. from 6.3% of cloacal swabs. Moreover, higher isolation rates of 64.5% in Bangladesh [54] and 92% in India [55] were documented. Lower rates of 2.6% in Northern Thailand [56] and 4% in Spain [51] were reported.

Meat contact surfaces are considered sources of contamination of meat served for human consumption [57]. In the present study, 10% of the surface swabs from wooden cutting boards were contaminated with *Salmonella* spp. (Table 2). A study in Nigeria reported the isolation of *Salmonella* spp. in meat contact surfaces [57]. Contamination of the cutting boards could be originated from water used in washing. Another study in Nigeria revealed that no *Salmonella* spp. were isolated from the wooden surface swabs in poultry outlets [58]. The variation in the isolation rates from the wooden contact surfaces indicates variation in hygienic practices applied during processing in the pluck shop outlets. Other studies reported the identification of *Salmonella* spp. in chopping boards at retail chicken outlets with the percentage of 18.8 in India [59] and 100% in Malaysia [60].

*Salmonella* infection in humans frequently occurs as a result of cross contamination from
equipment, utensils and workers’ hands due to subsequent handling of raw carcasses and products. In addition, consumption of undercooked poultry meat is another important route of transmission [47]. Table (2) shows that the overall prevalence of *Salmonella* spp. in 130 examined stool samples of human participants residing the same localities from which chicken samples were obtained was 0.8%. Nearly similar results of 1.3 and 2% were reported in Japan [61] and Egypt [50], respectively. In Egypt, 10% [62], 6% [41] and 4% [38] prevalence rates of *Salmonellae* in stool samples were reported.

The frequency of *Salmonella* spp. in hand swabs from workers at pluck shop outlets was 10% (Table 2). Nearly similar results of 14.3% in India [59] and 8.9% in Egypt [63] were reported. However, Ahmed et al. [41] documented that 4% of hand swabs collected from poultry pluck shop workers in Dakahlia, Egypt, were contaminated with *Salmonellae*. Therefore, hand cleaning and disinfection before and after contact with eggs and chicken carcasses are essential to minimize the risk of cross contamination [64].

**Serotypes identified in the examined samples**

*S.* Typhimurium and *S.* Enteritidis are the most predominant isolated organisms in most cases associated with the consumption of contaminated poultry and their products [65]. The predominance of *S.* Typhimurium (1.2%) followed by *S.* Enteritidis (1.01%) and *S.* Newport (0.9%) in the current study was in concordance with the Centers for Disease Control and Prevention (CDC) [66]. In addition, the predominant serovars present in Egyptian poultry farms are *S.* Typhimurium and *S.* Enteritidis[25, 45]. In contrary, a study in Egypt, reported that *S.* Enteritidis predominated and was followed by *S.* Typhimurium in samples of chicken origin [47].

The identification of *S.* Typhimurium in different samples during the current study highlights the importance of such serovar to pose a potential risk to poultry consumers and pluck shop workers. *S.* Enteritidis has shown an increase in frequency in different parts of the world in the last two decades [67]. It is also the most predominant serovar associated with chicken egg production [61].

*S.* Kentucky currently ranks among top ten serovars causing gastroenteritis in humans [68]. The poultry source of this serovar was previously reported [69]. Isolation rates of 41%, 20%, 14.3% and 10.8% for *S.* Kentucky isolated from chicken samples were reported in Ireland [70], USA [71] and Egypt [43, 47], respectively.

The isolation of *S.* Newport from chickens was previously reported in Patna [37], Northeastern Thailand [13] and Iran [40] with the percentages of 2.6%, 1.3% and 8%, respectively. The CDC annual summary identified *S.* Enteritidis, *S.* Typhimurium and *S.* Kentucky as the most frequent causative agents causing disease burden on consumers in the United States [2].

In the present study, *S.* Infantis was only identified in chicken meat samples and cloacal swabs (1.3%, each). Likewise, Kaushik et al. [37] isolated *S.* Infantis from poultry meat in India with an isolation rate of 0.4%. Also, isolation of this serovar from broiler flocks suggested that chicken meat is one of the sources for human infection [61, 72, 73]. *S.* Infantis has been reported previously in Japan from asymptomatic cases and food handlers [61].
Molecular identification of virulence associated genes

Various virulence determinants in *Salmonella* spp. are associated with chromosomal and plasmid factors [8]. All 29 identified *Salmonella* strains were subjected to PCR genotyping for detection of some virulence determinants. The results revealed the detection of *avr*A, *hil*A and *pef* genes in 100, 91.3 and 10.3% of the examined isolates (Figure 3). The detection of *avr*A gene in 100% of *Salmonella* isolates was also reported in other studies [12, 24]. However, lower frequencies of 80% [74] and 50% [75] were reported in *Salmonella* enterica isolates. This variation could be attributed to recombination which frequently occurs in the location of this gene [76]. Consistent with the current results of *hil*A gene, it was previously identified in 100 and 88.2% of *Salmonellae* isolated from chicken samples in Brazil [12] and Egypt [6], respectively. While the gene was identified in 8.3% [38] and 8.6% [18] of *Salmonella* isolates. Regarding to results of *pef* gene, the obtained low frequency of *pef*A gene was comparable with other findings [24, 77]. However different studies in Egypt reported the detection of the gene with higher frequencies ranging from 100% [38] to 41.2% [6]. The considerable differences in virulence determinants of *Salmonella* serovars are attributed to the variation in sample sources, types of serovars and presence or absence of plasmids carrying virulence associated genes [78].

**Conclusion**

It could be concluded that a proportion of chicken carcasses and giblets sold in Sharkia, Egypt is contaminated with *Salmonella* spp., predominantly *S. Typhimurium* and *S. Enteritidis*. the majority of the isolates harbored virulence associated genes, hence, chicken meat and their products constitute a significant problem for public health. Thus,
this calls for better measures to control cross contamination of poultry meat during slaughter and handling with pathogenic bacteria must be taken.

**Conflict of interest**

The authors declare no conflict of interest.

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primary production stage. https://www.iso.org/standard/42109.html


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الملخص العربي

التوصف البكتريولوجي والجزيئي لأنواع السلمونيلا المستفردة من الأدميين والذبج بمحافظة الشرقية - مصر

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تمت الدراسة الحالية لتحديد نوعان السالمونيلا في عد 89 عينة فردية بمحافظة الشرقية - مصر. وشملت العينات على لوح النجاح، حوار النجاح، بيض، عينات قنها المجمعة، مسحة أسطح محاذا وحوار النجاح، وضع في محلات عيد وتحضير النجاح ومسحات آيدي عمال تلك المحال. وكذلك تم أخذ عينات راز من الإنتاج المعدي المترشدين على العيان الخارجية مستشفى الأنهار. وقد تم عزل أنواع السالمونيلا بنسبة 90.6%، 8.0%، 4.0%، 0.4%، 0.0%، 10% من عينات لمحوم النجاح، والحوار وقشر البيض ومسحات قنها المجمعة ومسحات أسطح المعاشر ومسحات آيدي عمل تلك المحال على التوالي. ووجد أن نسبة العزل من مسحات النجاح 90.6%، وبالمفصلي الكيميائي الحيوي تم التعرف على أنواع السالمونيلا في 49 من 124 عينة وحوزة موجبة للسالمونيلا 

تم تحديد عدد 54 عينة باستخدام جين invA عنبر (8.7%). بينما بالتصنيف الجزيئي باستخدام جين

وعلل أن نسبة العزل من مسحات النجاح 0.4%، ويتصل السالمونيلا تيفيموريوم والسالمونيلا انتريتيدين من عينات مختلفة بنسبة 0.2%، 1.0% على التوالي. ونسبة 91.5% و10.3% في نسبة 100% من السالمونيلا بالبيومورت (0.9%) والسالمونيلا كتاكامي (0.7%) والسالمونيلا انتريتيدين (0.4%). كما تم تحديد جينات الضراوة المتطرفةBI40083665 (4.4%) من المستندات المختبرة. ونستنتج من الدراسة الحالية أن يوجد نسبة ثابتة بالسالمونيلا في محاولات التجارب المدواخل بمحافظة الشرقية بمصر وتحت ظروف البحث الحالي. ونسود السالمونيلا تيفيموريوم والإنتريتيدين الضارة أنواع السالمونيلا الأخرى مما يشكل خطراً محتملاً على المستهلك.