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# Prevalence of Antibiotic Resistance and Biofilm Producing *Salmonella spp*. in Some Egyptian Foods.

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**Abstract:** A total of 400 food samples including 200 meat (luncheon, frozen lean beef, raw lean beef and frozen chicken meat) and 200 dairy products (white cheese, kareesh cheese, turkey cheese and pasteurized milk) were examined for *Salmonella* prevalence. *Salmonella* could be detected in 14 (28%), 4 (8%), 11 (22%) and 7 (14%) of luncheon, frozen lean beef, raw lean beef and frozen chicken meat samples, respectively. On the other hand, the percentage of positive samples in the case of dairy samples represented 6 (12%), 19 (38%) and 13 (26%) of white cheese, kareesh cheese and turkey cheese respectively, while it could not be detected in any of pasteurized milk samples. Serotyping of the isolated *Salmonella* (71 isolates) from meat and dairy samples using slide agglutination test revealed that, the highest predominant serovar was 35 *S.* Typhimurium (49.3%), followed by 18 S. Newport (25.4%), 16 *S.* Enteritidis (22.5%) and two *S.* Kentucky (2.8%; 2 from meat samples only). The results of antibiotics susceptibility profiles highlighted the presence of multi-drug resistance by several serovars of *Salmonella*, especially *S.* Typhimurium. Regarding the formation of the biofilm, the obtained results showed that the serovars were differed in their ability to form it where *S.* Typhimurium was the strongest producing serovars with 10 isolates (10/35; 28.57%), followed by *S.* Enteritidis were 3 isolates (3/16; 18.75%). The presence of *Salmonella* in Egyptian foods it raises the alarm that control over food quality and safety must be tightened.

**keywords**: *Salmonella*; Meat; Dairy products; Multidrug-resistant; Biofilm; Egypt**.**

## 1.Introduction

Foodborne infections are estimated to cause 600 million cases and 420,000 deaths per year around the world. Unsafe foods constitute a threat to human health and the economies of countries, affecting primarily populations at risk of exclusion, migrants, and people living in conflict zones (**1**). Microbial contamination of food can occur at any point along the process, including harvesting, slaughtering, processing, and distribution ("farm to fork"), and can be caused by pollution of the environment, such as water, soil, or air (**1**). Foodborne bacteria cause a wide range of diseases, all of which have substantial economic and health implications (**2**). The most common symptoms of foodborne infections are gastrointestinal, such as diarrhea, but kidney and liver failure, brain and neurological abnormalities, reactive arthritis, and other complications can also occur. Children, pregnant women, the elderly, and individuals with a compromised immune system are particularly vulnerable to these infections (**3**).

Food safety is a major public health concern around the world. Unsafe food has the potential to spread a variety of foodborne illnesses and epidemics. According to a recent WHO research, an estimated 600 million individuals

(about one out of every ten people) become unwell after eating contaminated food each year, with 420,000 deaths, resulting in the loss of 33 million disability-adjusted life years (**4**).

*Salmonella* is a major cause of foodborne illnesses that have resulted in increased morbidity and mortality around the world (**5**). Non-typhoidal *Salmonella* is a leading cause of foodborne gastroenteritis worldwide, with a significant illness burden and economic losses (**6**). Additionally, it was responsible for 59,000 deaths worldwide, out of a total of 2,30,000 deaths caused by foodborne diarrheal disease agents (**7**). So, it can be said that Salmonellosis is the third most common foodborne cause of death worldwide, after norovirus (120 million cases) and Campylobacter spp. (96 million cases), as well as the most common foodborne illness (7.6 million cases) (**7**). Moreover, salmonellosis has lately been observed at an unusually high rate (**8**). Meat products (chicken, beef, sausage etc..) has also been found as a source of non-typhoidal *Salmonella* resistant to clinically relevant antibiotics, with a higher prevalence in middle-income nations (**9**). In addition to chicken meat may be a common source of microbial foodborne pathogens such as *Salmonella* spp. and E. coli (**10**). Recently, cheese has become the main component of patient meals, as it is an energy-dense, nutritional, and concentrated form of milk with a long shelf life (**11**). Regardless, dairy products may include harmful bacteria such as *E. coli* and *Salmonella* if they were inadequately heat-treated and pasteurized, rendering them inappropriate or potentially dangerous causes of foodborne illness (**12**). *E.coli* and *Salmonella* pathogenic strains were regularly found as the most common foodborne pathogens connected to consumption of raw or insufficiently heat-treated dairy products offered in rural regions and local grocery stores in Mansoura city (**13**,**14,15**). Antimicrobial-resistant *Salmonella* infections, particularly multidrug-resistant (MDR) bacteria, are on the rise and spreading, posing a huge public health threat (**9**). Multidrug resistance (MDR) in *Salmonella* strains is known to be of zoonotic origin, meaning it can be passed from animals to people via contaminated products (**16,17**). As a result, determining antibiotic resistance profiles in field isolates is critical for amending applicable legislation and implementing more effective national antibiotic resistance prevention programs. Bacterial biofilm means attachment, aggregation of bacterial cells and then engraved in a matrix of extracellular polymeric substance. This biofilm consists of water, proteins, lipids, enzymes, polysaccharides and DNA, which protects against environmental challenges and increase resistance to many antimicrobials (**18**). Therefore, the aim of this study to reveal the presence of antibiotics resistant *Salmonella* in more prevalent types of Egyptian foods, to shed light on the increased the quality and safety of these foods.

## 2. Materials and methods

### 2.1. Tested food samples.

During December 2019 and March 2020, a total of 400 meat and dairy food samples (100 g each) were collected from different retail supermarkets and specialty food shops from rural areas and small cities in Kafr El-Sheikh Governorate, Egypt. Meat products, were luncheon (50), frozen lean beef (50), raw lean beef (50) and frozen chicken meat (50) samples. Dairy products collected were white cheese (50), kareesh cheese (50), turkey cheese (50) and pasteurized milk (50) samples. Samples were collected in clean sterile plastic bags (disposable; Alexandria, Egypt), placed in an ice-boxes and transferred to the microbiology lab (plant viruses and bacteriophage Lab of Botany and Micro. Dept., Fac. of Sci., Al-Azhar Univ. Cairo, Egypt) for examination without delay or stored at a freezing temperature of $-20^{o}$C until analysis.

### 2.2. Samples preparation

Twenty-five (25g) of each hard sample was mixed and homogenized in sterile mixer with 225 ml sterile buffered peptone water (BPW), (Oxoid), while 25 ml of each milk sample was added directly to 225 ml of BPW. Ten-fold dilutions of homogenates samples were prepared and ready for microbiological testing (**19**).

### 2.3. Isolation of *Salmonella*

Each prepared food sample (25g sample/225 ml BPW) was overnight incubated at 35°C, followed by inoculation (1:10) into Rappaport-Vassiliadis broth (Oxoid) and overnight

incubated again at 40 °C. A loopfuls (100 µl) from each enriched culture were separately streaked onto each of *Salmonella* and *Shigella* (SS) agar (Oxoid; CM0099) and xylose lysinedeoxycholate agar (XLD), (Oxoid; CM0469). The plates were incubated at 37°C for 24 h, after incubation, the expected colonies were creamy with or without black centers on SS agar and red in color with or without black centers on (XLD) agar. From each selective medium, 2-3 colonies were selected and streaked onto Tryptone Soya Agar (Oxoid, Hampshire, UK) slope which overnight incubated at 37 °C, for further identification (ISO 6579, 2002).

### 2.4. Identification of the *Salmonella* isolates

All *Salmonella* isolates firstly have been identified morphologically (**20**), biochemically (Indole, Triple sugar iron (TSI), Oxidase, Catalase, Voges –Proskauer, Methyl red, Citrate utilization (**21**) and Urease test (**22** ), and serologically (slide agglutination test) (**23,24**) in the second step of identification, the Biomerieux VITEK 2 system (**25,26**) was used automatically to confirm the *Salmonella* isolates.

### 2.5.AntibioticResistanceProfilesof*Salmonella* Isolates

The antibiotic-resistant profiles of all Salmonella isolates were determined using the disc diffusion technique (**27**). The following medications were used in this test: streptomycin (10 g), kanamycin (30 mcg), flucloxacillin (5 mcg), tetracycline (30 mcg), and levofloxacin (30 mcg) (5 mcg). Rifamycin (30 mcg), Erythromycin (15 mcg), Amoxicillin/clavulanic acid AMC (20 g), Clindamycin (2 mcg), Gentamicin (10 mcg), Cephradine (30 mcg), Ciprofloxacin (5 mcg), and Ampicillin (10 mcg). The antibiotic resistance or sensitivity of the studied Salmonella was determined here by evaluating the growth inhibitory zone around each antibiotic disc after 24 h of incubation at 37 °C. Based on the defined protocols of the National Committee for Clinical Laboratory Standards (NCCLS) in 2007 (**28**), the obtained results were labelled as S (sensitive), I (intermediate sensitive), and R (resistant).

### 2.6. Biofilm Forming Activity of *Salmonella* Isolates

The qualitative evaluation of biofilm formation activity of all antibiotic resistant Salmonella isolates was performed using the tissue culture plate method (**29**). The production of biofilms was assessed in 96-well tissue culture polystyrene plates with a flat bottom and cover in this test (Sigma-Aldrich, Costar, USA). In each well, 200 l of TSB media enriched with 0.25 percent glucose was added in each well plus 20 µl of $10^5$CFU/ml of a bacterial suspension. The plates were aspirated and rinsed with phosphate-buffered saline (PBS) after an overnight incubation at 37 °C . After removing the ethanol by washing, the adsorbed bacteria were fixed to the polystyrene wells with 95 percent ethanol and then stained with crystal violet (0.1 percent). At the Botany and Micro. Dept., Fac. of Sci., Al-Azhar Univ. Cairo, Egypt, the dye was solubilized in 1% w/v SDS and the optical densities were measured photometrically at O.D.570 nm using an ELISA reader (Sunrise™-TECAN, Switzerland). The experiment was carried out in triplicate. Depending on Stepanovi's *et al*., (2007) interpretation, the developed biofilm was described as low, moderate, or strong(**29**).

## 3. Results

### 3.1. Identification of isolated *Salmonella* spp.

Typical colonies for *Salmonella* on XLD (pink colonies with or without black centers), as well as on SSA (smooth, round, black or colorless colonies) were taken for biochemical tests. All Gram's stain negative, urease negative, oxidase negative isolates with production of black colonies on TSI agar were considered as biochemically confirmed *Salmonella* isolates Table (1). In the affirmative identification by Biomerieux VITEK 2 system, the results of the isolates were all *Salmonella* spp.

### 3.2. Prevalence of *Salmonella* spp. in meat samples and Serotyping

Out of 200 examined meat sample (50 each for luncheon, frozen beef, raw beef, and frozen chicken meat) the prevalence of *Salmonella* spp. was summarized in Table (2). The highest level of contaminated samples was found in luncheon, where 14 (28%) samples showed a positive result for the presence of *Salmonella*,

followed by the raw beef 11 (22%), frozen chicken meat 7 (14%) and 8% of frozen beef (4 out of 50 samples).

**Table.1**: Morphological and Biochemical characterization of various isolates suspected to *Salmonella* spp.

| Test | Isolate result |
|---|---|
| Gram reaction | -ve |
| Shape of colonies | circular |
| Color of colonies | Greyish white |
| Spore | -ve |
| Indole | -ve |
| Triple sugar iron (TSI) | +ve |
| Oxidase | -ve |
| Catalase | +ve |
| Voges –Proskauer (VP) | -ve |
| Methyl red (MR) | +ve |
| Citrate utilization | -ve |
| Urease test | -ve |

**Table.2:** Prevalence of *Salmonella* spp. in meat samples

| Sample type | Number of samples | Positive samples | % samples |
|---|---|---|---|
| Luncheon | 50 | 14 | 28 |
| Frozen beef | 50 | 4 | 8 |
| Raw beef | 50 | 11 | 22 |
| Frozen chicken meat | 50 | 7 | 14 |
| **Total** | 200 | 36 | 18 |

The results of serotypes in the current study showed that, *Salmonella* isolates from meat samples were 16 *S.* Typhimurium (48.5%; 6 from luncheon, 3 from frozen beef, 4 from raw beef and 3 from frozen chicken meat ), 5 *S.* Enteritidis (15%; 3 from luncheon and 2 from raw beef), 10 *S.* Newport (30.3%; 4 from luncheon, 1 from frozen beef, 3 from raw beef and 2 from frozen chicken meat) and two *S.* Kentucky (6%; 1 from luncheon and 1 from frozen chicken meat) As shown in Table (3) and Figure (1).

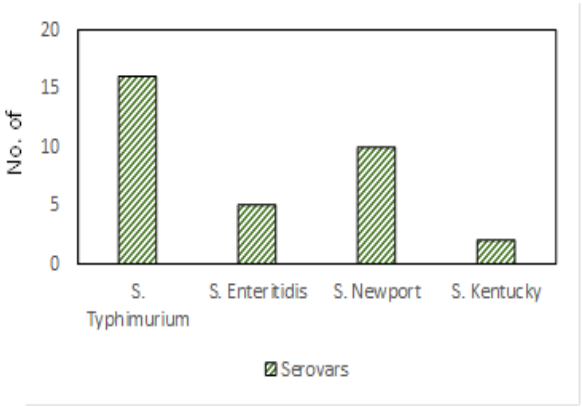


**Figure 1:** Serovars of *Salmonella* isolates from meat samples

**Table 3.** Serotyping of isolated *Salmonella* from meat samples

| Sample type | Serovars / (No.) | | | | No. of isolates |
|---|---|---|---|---|---|
| | *S.*Typhimurium | *S.*Enteritidis | *S.* Newport | *S.* Kentucky | |
| Luncheon | + (6) | + (3) | + (4) | + (1) | 14 |
| Frozen beef | + (3) | - | + (1) | - | 4 |
| Raw beef | + (4) | + (2) | + (3) | - | 9 |
| Frozen chicken meat | + (3) | | + (2) | + (1) | 6 |
| **Total** | 16 | 5 | 10 | 2 | 33 |

### 3.3. Prevalence of *Salmonella* spp. in Dairy samples and Serotyping

In dairy samples (200), the highest contamination percentage (38%) was found in Kareesh cheese (19 out of 50 samples). The turkey cheese samples showed 13 (26%) positive for *Salmonella*, while 6 (12%) samples of white cheese were positive. On the other hand, pasteurized milk samples did not show any positive results for the presence of *Salmonella* spp. all data were outlined in Table (4).

**Table.4:** Prevalence of *Salmonella* spp. in dairy samples

| Sample type | Number of samples | Positive samples | % samples |
|---|---|---|---|
| White cheese | 50 | 6 | 12 |
| Kareesh cheese | 50 | 19 | 38 |
| Turkey cheese | 50 | 13 | 26 |
| Pasteurized milk | 50 | ND | - |
| **Total** | 200 | 38 | 19 |

ND= not detected

Thirty-eight isolates of *Salmonella* were serotyped as 19 *S.* Typhimurium (50%; 3 from white cheese, 9 from kareesh cheese and 7 from turkey cheese), 11 *S.* Enteritidis (28.9%; 1 from white cheese, 6 from kareesh cheese and 4 from turkey cheese) and 8 *S.* Newport (21%; 2 from white cheese, 4 from kareesh cheese and 2 from turkey cheese), Table (5) and Figure (2).

**Table.5:** Serotyping of isolated *Salmonella* from dairy samples

| Food sample | Serovars / (No.) | | | No.of isolates |
|---|---|---|---|---|
| | *S*.Typhimurium | *S*.Enteritidis | *S*.Newport | |
| White cheese | + (3) | + (1) | + (2) | 6 |
| Kareesh cheese | + (9) | + (6) | + (4) | 19 |
| Turkey cheese | + (7) | + (4) | + (2) | 13 |
| Pasteurized milk | - | - | - | - |
| **Total** | 19 | 11 | 8 | 38 |

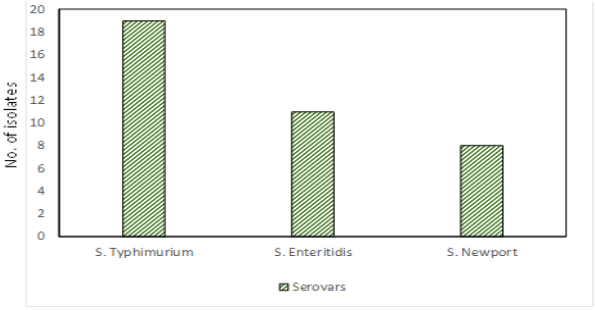


**Figure 2:** Serovars of *Salmonella* isolates from meat samples

Concerning the distribution of *Salmonella* serotypes in examined meat and dairy samples, the obtained results were illustrated in Figure (3). Seventy-one isolates of *Salmonella* were typed as 35 *S*. Typhimurium (49.3%; 16 from meat and 19 from dairy samples), 16 *S*. Enteritidis (22.5%; 5 from meat and 11 from dairy samples), 18 *S*. Newport (25.4%; 10 from meat and 8 from dairy samples) and 2 *S*. Kentucky (2.8%; 2 from meat samples). The most prevalent serovar detected in this study was *S*. Typhimurium.

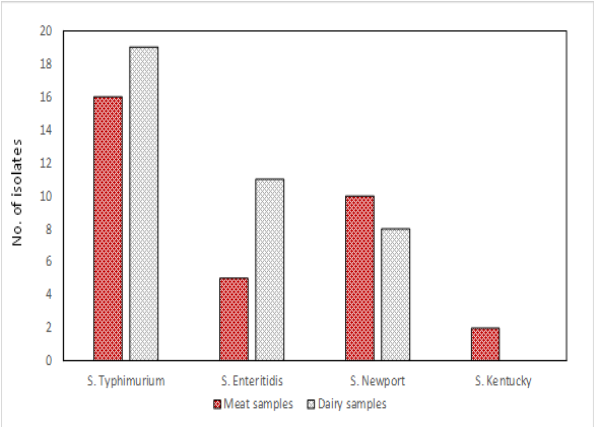


**Figure 3.** Distribution of *Salmonella* serovars in examined meat and dairy samples

### 3.4. Antibiotics susceptibility profiles

The results of determination of antibiotics resistance of different *Salmonella* serovars (*S*. Typhimurium, *S*. Enteritidis, *S*. Newport and *S*. Kentucky) to sixteen antibiotics are presented in Tables (6). At least 23 isolates of *S*. Typhimurium exhibited resistance to all tested antibiotics, so this serovar is considered the most resistant strain. Four isolates of *S*. Enteritidis were resistant to streptomycin, Rifamycin and Erythromycin, seven to Kanamycin, six to oxacillin and gentamicin, while two to three isolates were resistant to flucloxacillin, AMC, ciprofloxacin, cephradine and ampicillin. *S*. Newport showed more resistance to oxacillin (10 out of 18 isolates), while its resistance to other antibiotics was low. Two *S*. Kentucky isolates showed resistance to flucloxacillin, levofloxacin, oxacillin and ampicillin, while only one was resistant to streptomycin, tetracycline, tobramycin and ciprofloxacin.

**Table.6**: Antibiotic sensitivity profiles of isolated *Salmonella* spp.

| Antibiotics | *S*. Typhimurium (n=35) | | | *S*.Enteritidis(n=16) | | | *S*. Newport(n=18) | | | *S*. Kentucky(n=2) | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | R (No.) | I (No.) | S (No.) | R (No.) | I (No.) | S (No.) | R (No.) | I (No.) | S (No.) | R (No.) | I (No.) | S (No.) |
| Streptomycin | 23 | 9 | 3 | 4 | 7 | 5 | 3 | 7 | 8 | 1 | 0 | 1 |
| Kanamycin | 23 | 7 | 6 | 7 | 0 | 9 | 0 | 3 | 15 | 0 | 0 | 2 |
| Flucloxacillin | 23 | 11 | 1 | 3 | 12 | 1 | 0 | 9 | 9 | 2 | 0 | 0 |
| Tetracycline | 31 | 2 | 2 | 0 | 2 | 14 | 0 | 10 | 8 | 1 | 1 | 0 |
| Levofloxacin | 28 | 4 | 3 | 0 | 6 | 10 | 4 | 6 | 8 | 2 | 0 | 0 |
| Tobramycin | 23 | 3 | 9 | 0 | 11 | 5 | 2 | 9 | 7 | 1 | 1 | 0 |
| Aztreonam | 24 | 2 | 7 | 0 | 9 | 7 | 2 | 0 | 16 | 0 | 0 | 2 |
| Oxacillin | 23 | 10 | 2 | 6 | 0 | 10 | 10 | 0 | 8 | 2 | 0 | 0 |
| Rifamycin | 23 | 9 | 3 | 4 | 0 | 12 | 0 | 14 | 14 | 0 | 1 | 1 |
| Erythromycin | 34 | 1 | 0 | 4 | 0 | 12 | 0 | 4 | 14 | 0 | 1 | 1 |
| AMC | 23 | 3 | 9 | 3 | 5 | 8 | 0 | 3 | 15 | 0 | 1 | 1 |
| Clindamycin | 23 | 12 | 0 | 0 | 8 | 8 | 5 | 6 | 7 | 0 | 0 | 2 |
| Gentamicin | 29 | 0 | 6 | 6 | 8 | 2 | 2 | 0 | 16 | 0 | 1 | 1 |
| Cephradine | 23 | 0 | 12 | 2 | 0 | 14 | 4 | 0 | 14 | 0 | 1 | 1 |
| Ciprofloxacin | 23 | 12 | 0 | 3 | 0 | 13 | 0 | 8 | 10 | 1 | 0 | 1 |
| Ampicillin | 23 | 11 | 1 | 2 | 0 | 14 | 6 | 0 | 12 | 2 | 0 | 0 |

R = indicates the isolate is resistant to antibiotics, I = intermediate resistance, S= the isolate is sensitive; AMC= Amoxicillin/clavulanic acid; (No.) = indicates the number of isolates.

### 3.5. Biofilm forming capacity of isolated *Salmonella* spp.

Biofilm formation of isolated *Salmonella* was investigated using tissue culture plate method and the obtained results were outlined in Table (7) and illustrated in Figure (4). The serovars exhibited different abilities in biofilm formation, where *S.* Typhimurium was the strongest producing serovars with 10 isolates (10/35; 28.57%), followed by S. Enteritidis were 3 isolates (3/16; 18.75%). The biofilm produced by S. Newport and S. Kentucky was not strong.

**Table.7:** Biofilm forming capacity of isolated *Salmonella* spp.

| **Biofilm activity** | *S.* Typhimurium (n=35) | | *S.* Enteritidis (n=16) | | *S.* Newport (n=18) | | *S.* Kentucky (n=2) | |
|---|---|---|---|---|---|---|---|---|
| | No. | % | No. | % | No. | % | No. | % |
| Strong | 10 | 28.57 | 3 | 18.75 | 0 | 0 | 0 | 0 |
| Moderate | 17 | 48.57 | 9 | 56.25 | 6 | 33.33 | 0 | 0 |
| Weak | 8 | 22.85 | 4 | 28 | 12 | 55.55 | 2 | 100 |

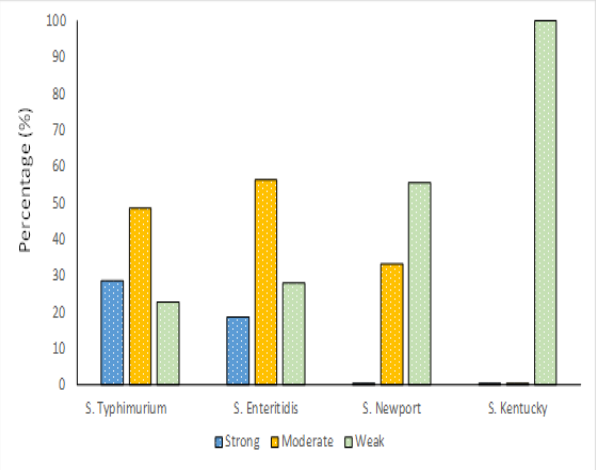


**Figure 4.** Biofilm formation activity by different *Salmonella* serovars

## 4. Discussion

*Salmonella* is a common foodborne bacterium that causes gastroenteritis, intestinal diseases, foodborne outbreaks, and mortality all over the world (**30**, **31**, **32**). *Salmonella* is responsible for roughly 13 billion gastroenteritis cases worldwide each year, with clinical manifestations ranging from simple intestinal infections to systemic illnesses (**33**). In general, humans become infected by eating tainted animal-derived foods such as eggs, milk, poultry, pig, beef, and other meats (**31, 33**). As a result, it is critical to implement a rapid, easy, and ideally visible detection technology to detect the presence of *Salmonella* in various food substrates in order to manage *Salmonella* in foods of animal origin (**31**). In the present study, a total of 400 food samples including 200 meat (luncheon, frozen lean beef, raw lean beef, and frozen chicken meat) and 200 dairy products (white cheese, kareesh cheese, turkey cheese, and pasteurized milk) were examined for *Salmonella* prevalence. *Salmonella* could be detected in 14 (28%), 4 (8%), 11 (22%), and 7 (14%) of luncheon, frozen lean beef, raw lean beef, and frozen chicken meat samples, respectively. On the other hand, the percentage of positive samples in the case of dairy samples represented 6 (12%), 19 (38%), and 13 (26%) of white cheese, kareesh cheese, and turkey cheese respectively, while it could not be detected in any of pasteurized milk samples. In consistence with our results, *Salmonella* was found in 19 percent of packed broiler meat from retail grocery stores in research done in Pennsylvania (**33**). As well as the results of the present study were in agreement with many previous studies in Egypt, where who detected *Salmonella* spp. in chicken meat with a prevalence of 14% (**34**) and beef (**35**). Additionally, the current findings were also in agreement with many reports from other countries, such as 14.5% from Nepal (**36**), 14% from Canada (**37**), 19.2% from South Africa (**38**), and 12% from Turkey (**39**). The presence of *Salmonella* in meat products may be attributed to the fact that this product is made from raw meat (contamination during slaughtering, scalding, de-feathering, evisceration, carcass cutting, and handling). In addition, the natural casing is frequently utilized in the manufacturing process, which could be an important source of *Salmonella* especially if proper hygienic measures are neglected (**40**). Pathogenic bacteria may contaminate carcasses after

slaughter and then spread through cut meat or meat materials destined for further processing into meat products (**41**). In disagreement with our results in many previous studies in Egypt by **(42)**, (**43)** and **(44)** failed to isolate *Salmonella* from processed meat samples. On the other hand, our findings indicate the presence of *Salmonella* spp. in dairy products and this is in accordance with **(45)** who reported the presence of *Salmonella* in cheese samples. *Salmonella* failed to be recovered from cheese samples in many earlier studies (**46, 47, 48)**. The presence of some pathogenic microorganisms especially *Salmonella* in dairy products may be due to using raw milk in the production accompanied by improper sanitary practices during manufacturing, handling, and selling. The isolation of this pathogen hence these foods could be of high risk in transmitting enteric pathogens. These results are supported by the findings by (**49**) they found that *Salmonella* spp., were the reason for all 29 outbreaks.

Serotyping of the isolated *Salmonella* (71 isolates) from meat and dairy samples using slide agglutination test revealed that the highest predominant serovar was 35 S. Typhimurium (49.3%), followed by 18 S. Newport (25.4%), 16 S. Enteritidis (22.5%) and two S. Kentucky (2.8%; 2 from meat samples only). In consistence with our findings, many studies showed that the most commonly isolated serotype from different organs was S. Enteritidis and S, Typhimurium, the same results were detected in Egypt by (**50**; **51** and **52**) who confirmed the prevalence of S. Enteritidis and S. Typhimurium by (58.33% and 41.66%), respectively from chickens. In addition, S. Enteritidis and S. Typhimurium were predominant in Saudi Arabia, by (55.56% and 22.22%, respectively) among the detected *Salmonella* serovars from chickens (**53**). In previous investigations, (**34**) and **(54)** found that S. Typhimurium was the most common serotype of *Salmonella* isolates from chicken meat in Egypt and India, respectively.

Antibiotic susceptibility profiles revealed the occurrence of multi-drug resistance in various *Salmonella* serovars, particularly S. Typhimurium. Our findings are in accordance with another previous research by **(55)** who detected 100% resistance of *Salmonella* to some tested antibiotics. Additionally**, (56)** was reported that *Salmonella* isolates from chicken were highly resistant to tetracycline, sulphamethoxazole, trimethoprim, and lower resistance to ciprofloxacin and cefotaxime, **(56)**. The resistance in *Salmonella* may be attributed to the presence of the blaTEM gene. This interpretation was agreed with **(57)** who showed blaTEM-1 and blaTEM-104 from gram-negative bacteria isolated from farms in Egypt. Moreover, **(58)** analyzed the mechanisms of multidrug- resistance in 21 isolates of S. Enterica serovar Enteritidis and four isolates of S. Enterica serovar Typhimurium also, identified bla cmy-2 in isolates of S. Enterica serovar Enteritidis. As well as **(59)** identified the blaTEM-1 in S. Enterica serovars in the United States and China.

Regarding the formation of the biofilm, the obtained results showed that the serovars have differed in their ability to form it where S. Typhimurium was the strongest producing serovars with 10 isolates (10/35; 28.57%), followed by S. Enteritidis were 3 isolates (3/16; 18.75%). Many previous studies indicated that *Salmonella* spp. has demonstrated the capacity to form biofilms on several surfaces (**60**, **61**,**62** ,**63)** which suggest that biofilm formation capacity may be an important factor for the persistence of *Salmonella* on food and other contact surfaces (**64**). Inconsistent with our results other studies decided similar results, The S, Typhimurium serotype has been characterized as the most powerful biofilm producer (**65, 66**). Several stud¬ies describe research on the resistance of *Salmonella* bio¬film to antimicrobials, such as ampicillin, ciprofloxacin, gentamicin, tetracycline, or third-generation cephalo¬sporins such as ceftriaxone and cefotaxime (**67, 68**). Additionally, it was reported that S. Typhimurium biofilms performed on polystyrene micro¬ plates also exhibited up to 200-fold greater resistance to ciprofloxacin compared to planktonic cells (**69**).

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