Prevalence and Genotypic Analysis and Antibiotic Resistance of *Salmonella* **Species Isolated from Imported and Freshly Slaughtered Chicken**

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Corresponding Author: Mahmoud E. Ezzat Department of Bacteriology, Immunology and Mycology, Faculty of Veterinary Medicine, Suez Canal University, Ismailia, Egypt Email: ezzat.scu@gmail.com Abstract: Salmonellosis is a bacterial disease caused by different types of Salmonella species and associated with clinical disease in both human, birds and livestock species. Salmonellosis associated with severe economic impacts either from losses in poultry farm due to pathological picture losses, cost of medication, condemnation in slaughterhouse or zoonotic important. This study focused on the prevalence of Salmonellae in imported frozen and freshly slaughtered chicken in Dakahlia Governorate, Egypt. A total of 273 samples were collected from 66 examined birds (24 from Balady chickens "Native Bred", 23 from Broiler chicken and 19 from frozen chicken). Nineteen out of 66 chickens (6.95%) Salmonella species were isolated, representing: 10 from Balady (native bred) chicken (8.3%). eight from broiler chicken (7%) and one from imported frozen chicken (2.61%). The serotyping of the Salmonellae isolated from chickens as follow (5) (26.3%) S. typhimurium, (5) (26.3%) S. kentucky, (3) (15.8%) S. infantis (2) (10.5%) S. enteritidis, (2) (10.5%) S. tamale, (2) (10.5%) S. newport. PCR assay was carried out on nine serovars (2) S. enteritidis, (2) S. typhimurium, (2) S. kentucky, (1) S. infantis, (1) S. tamale, (1) S. newport detect the presence of InvA (invasion protein InvA), bla_{TEM} (beta-lactamase TEM). aacC (Aminoglycoside N (3)-acetyltransferase IV), gepA (Quinolone efflux pump), tetA(A) (Tetracycline resistance protein, class D) and sull(Dihydropteroate synthase type-1) genes, the percentage respectively was 100%, 100%, 33.3, 33.3%, 88.89%, 77.78% in examined Salmonella strains. The susceptibility of Salmonella serovars to chemotherapeutic agents showed high sensitivity to levofloxacin and gentamycin, but the highest resistance was to sulfamethoxazole. The increased prevalence rate of Salmonella species isolation from chicken and chicken meat carrying Multidrug Resistance (MDR) genes for commonly used antimicrobial agents, consider a critical threat for public health issues. Further studies are required to understand Salmonella spp epidemiology and to limit the spread of multidrug-resistant types.

Keywords: Salmonella, Fresh Chicken, Frozen Meat, Food Poisoning

Introduction

Salmonellosis is a significant global public health issue and causes high morbidity rate and has a significant economic impact. Foodborne diseases persist in many countries and consider the most important public health problem. There is global improvement in food processing practices, hygiene measures, education of food handlers on the potential risks and information to consumers for proper utilization, but still so far to



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eliminate the foodborne pathogen problem (Ruban et al., 2010). Infection with Salmonellae is still the first reported cause of bacterial foodborne illness all over the world and associated with gastroenteritis disorders (Huong et al., 2006; WHO, 2004). Chicken meat Contaminated by Salmonellae is the most significant potential hazard of human gastroenteritis worldwide; as several reports correlate the human salmonellosis cases to the consumption of contaminated chicken meat and byproducts with different salmonella species (Beli et al., 2001). The majority of human salmonellosis infections are derived from the consumption of contaminated chicken meat, although there are several other transmission routes of infection (Hassanein et al., 2011). Contaminated frozen chicken products and byproducts have been considered potential source for salmonellosis; where the presence of Salmonella species in frozen chicken products maybe associated with potential infection risk to human if the product is improperly cooked (Dominguez and Schaffner, 2009). Chicken stored at variable temperatures and chickens brought from wet markets has higher levels of salmonella than those brought from other hygienic retail store; also, chickens produced by integrated companies have lower levels of salmonella than non-integrated companies due to higher biosecurity measures (Donado-Godoy et al., 2014). The risk of salmonellosis from chilled chickens higher than frozen chicken carcasses, which may associate to the preparation practices and hygienic measures (Donado-Godoy et al., 2012). Antibiotics administration is an important tool in the reduction the incidence and mortalities associated with avian salmonellosis (Duarte et al., 2009). The recent reports highlighted the increased rate of MDR salmonella species which may correlate to the massive and improper usage of antimicrobial agents in human and veterinary medicine and it represent a global public health problem (Cruchaga et al., 2001; Mosalem, 2016) (Eid et al., 2016; Elfeil et al., 2016; Enany et al., 2018). The problem of MDR salmonella spread all over the world with higher prevalence rate in in the developing nations with no clear prevalence mapping to such genes (Eid et al., 2019). The studying of drug resistance and MDR patterns between the different Salmonella strains gives not only important evidence to the clinician for the best therapeutic regime and suitable choices to control the reported cases but also an important tool in devising and scheduling a proper chemoprophylactic and chemotherapeutic drug regimens within a geographical area and highlighted the need for developing suitable antimicrobial agents against certain pathogen (Abouelmaatti et al., 2013; Elfeil et al., 2012; Murugkar et al., 2005). In Egypt, Salmonella reported the highest rate of foodborne infection via chicken's meat (WHO, 2004; 2007). The problem in Egypt is old

where: a prevalence survey has been conducted on year 1979 at upper Egypt region (Assiut, New-Valley and Suhag Governorates) and examined bacteriologically dead chickens and ducks as well as 2325 fecal swabs from living chickens and ducks suffered from paratyphoid infections; they isolated 17 Salmonella serotypes, where the most common types were S. hessarek, S. orian and S. miami (Bayoumi et al., 1979). Moreover, In Belbais El-Sharkia Governorate, another prevalence report applied on 1989 and has been recorded salmonella isolation from 10.7% of chickens and ducks in contact with human diarrheic cases (Taha, 1989) that similar to other prevalence report in east Egypt regions applied on 2016 and recorded *salmonellae* isolation from 10.5% and 14.3% of chicken and pigeon in contact with human diarrheic cases (El-Demerdash et al., 2016). In Dakahlia Governorate, salmonella species were isolated from 13.3% of chickens of different ages from farms in Bilkas and Gamasa (Moawad, 2009). While in Alexandria governorate, salmonella species were isolated from layer flocks in poultry farms with a percentage of 11.4% (Draz et al., 1997). Lower percentages than the previously mentioned were reported by Sadoma, (1997) who isolated Salmonellae from six out of 300 (2%) cloacal swabs collected from 30 chicken farms at different localities in Gharbia (Sadoma, 1997). Nabil and Yonis (1999) isolated S. typhimurium, S. anatum and S. pullorum from 2.5% of chickens and 4% of ducks samples from fattening and laying farms in Kafr El-Shaikh Governorate and Ahmed et al. (2009); isolated salmonella from chickens reared in a rural village in El-Sharkia province with a percentage of 1.7%; the serotyping study of the isolated strains showed that they were grouped into two different serogroups, S. enteritidis and S. typhimurium constituting 80% and 20%, respectively. A study was performed in Qalyubia governorate, Egypt and highlighted the prevalence of S. kentucky, S. infantis, S. enteritidis, S. typhimurium, S. chiredzi and S. tsevie in commercial broiler chicken flocks (El-Ghany et al., 2012). This study focused on examining random samples from different stores and slaughter houses and examine the prevalence of different salmonella species in frozen and freshly slaughtered chicken's mean and check the presence of antimicrobial resistant, MDR and virulence genes in collected isolates, which can seriously affect the human health and the medication regime and can return again to poultry farms through works in the poultry industry.

Materials and Methods

Sample Collection

A total of 273 samples (235) from freshly slaughtered and (38) from imported frozen chickens were aseptically collected from birds' markets in Dakahlia Governorate, Egypt. The samples were (liver, cecum, lung, breast muscle and thigh muscle). All samples were collected in sterile plastic bags and transferred immediately in icebox to Reference Laboratory for Veterinary Quality Control on Poultry Production for bacteriological examination.

Isolation of Salmonella

The collected samples weighted then inoculated in buffered peptone water (1:10 dilution) then incubated at 37±1°C for 16-20 h According to ISO 6579 (2002). The pre-enrichment broth was mixed after incubation, then sub culture in two different broths: where 0.1 mL of the broth was transferred into a tube containing 10 mL of Rappaport-Vassiliadis medium with soya (RVS broth) and incubated at 41.5°C for 24 h; another 0.1 mL of the pre-enrichment broth was transferred into a tube has 10 mL of Muller-Kauffmann Tetrathionate-Novobiocin, broth (MKTTn broth) and incubated at 37°C for 24 h. After incubation, a loop-full of the RVS and MKTTn broth were streaked separately onto the surface of Xvlose Lysine Deoxycholate Agar (XLD agar) and Hektoen Enteric (HE agar). The plates were incubated in an inverted position at 37°C for 24 h then checked for growth of typical Salmonella colonies. Typical colonies of Salmonella grow on XLD agar with a black center and a lightly transparent zone of reddish color due to the color change of the indicator. Lactose-positive Salmonella is yellow with or without blackening on XLD agar. On HE colonies appeared as deep blue.

Biochemical Identification

Purified isolates were examined by different biochemical reactions According to ISO 6579 (2002) either by oxidase, urea hydrolysis, H_2S production on TSI, lysine decarboxylation, citrate utilization and indole as shown in Table 1.

Serogrouping of Salmonella

The isolates that were identified biochemically as *Salmonella* were subjected to serological identification according to Kauffman-White Scheme (Kauffmann, 1972) for determination of somatic (O) and flagellar (H) antigens (Cruickshank, 1975).

Antimicrobial Susceptibility Testing

The pure identified *Salmonella* strains were tested for antimicrobial susceptibility; where the test was done by the agar disc diffusion method as previously described (Finegold and Martin, 1982).

Detection of Virulence Genes and Antibiotics Resistance Genes

The purified *salmonella* isolates subjected to DNA Extraction using QIAamp DNA mini kit (Qiagen, Germany) according to manufacturer's instructions. the extracted DNA subjected to PCR screening for the presence of selected virulence and antimicrobial resistant genes using specific Oligonucleotide primers sets listed in Table 2.

Table 1: Biochemical identification of suspected Salmonella species

Biochemical test	Reaction	Result
Oxidase reaction	Negative	Colorless
Urease hydrolysis test	Negative	Yellow (Medium remain original yellow)
H2S production in Triple sugar iron agar (TSI)	Positive	Alkaline (red) slant, acid (yellow) butt with or without - blackening due to H2S production. (k/A/G/H ₂ S)
Lysine decarboxylation test	Positive	Purple color with H2S at the middle of the tube
Citrate utilization	Positive	Development of deep blue color within 24-48 h
Indole reaction	Negative	Yellow ring.

Table 2: Oligonucleotide	primers sec	quences Source
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Primer	Sequence	Amplified product	Reference
tetA(A)	GGTTCACTCGAACGACGTCA	576 bp	Randall et al. (2004)
	CTGTCCGACAAGTTGCATGA	-	
blatem	ATCAGCAATAAACCAGC	516 bp	Colom <i>et al.</i> (2003)
	CCCCGAAGAACGTTTTC	_	
aacC	GGCGCGATCAACGAATTTATCCGA	448 bp	Lynne et al. (2008)
	CCATTCGATGCCGAAGGAAACGAT		
sul1	CGGCGTGGGCTACCTGAACG	433 bp	Ibekwe <i>et al.</i> (2011)
	GCCGATCGCGTGAAGTTCCG		
qepA	CGTGTTGCTGGAGTTCTTC	403 bp	Cattoir <i>et al.</i> (2008)
	CTGCAGGTACTGCGTCATG	-	
invA	GTGAAATTATCGCCACGTTCGGGCAA	284 bp	de Oliveira et al. (2006)
	TCATCGCACCGTCAAAGGAACC	_	

Results and Discussion

Poultry industry suffer from several pathogen threats in Egypt either bacterial like Salmonella, Pasteurella, Clostridium or viral types like Avian influenza virus (AIV), Newcastle Disease Virus (NDV), Infectious Bronchitis Virus (IBV), Infectious Bursal Disease Virus (IBDV) or parasitic as coccidia and it associated with severe economic on the industry and human public heath impact (Algammal and Elfeil, 2015; Ayoub et al., 2019; Diab et al., 2019; Eid et al., 2016; 2019; Elhady et al., 2018; Sedeik et al., 2018; 2019; Sultan et al., 2019a; 2019b). Salmonella consider one of the major bacterial threats affect poultry industry and associated with severe economic losses. In this study, 66 birds, (24) from Balady chickens, (23) Broiler chickens and (9) Frozen chickens were examined for the presence of Salmonella; the number and percentage of positive samples from Balady chickens" Native breed" (10; 8.3%), Broiler chickens (8; 7%) and Frozen chickens (1; 2.61%); where the highest percentage of Salmonella isolation was from Balady chickens, while the lowest percentage was from frozen chickens (Table 3). The obtained results matched with other reports describing the prevalence of salmonella in chicken and chicken products in other Egyptian governorates, where Ezzat et al. (2014); who found that Salmonella species isolates, representing: 7.5% from apparently healthy chicken in east Egypt regions and Ibrahim et al. (2013); who highlighted the Salmonella species occurrence as 9-10% in examined breeders flocks; Rabie et al. (2012); who reported the prevalence of Salmonella was 14% and 4% in chickens and raw chickens meat respectively in Toukh, Egypt and Hassanein et al. (2011); reported 52% of frozen chicken fillet and 36% of frozen chicken leg samples in Assiut, Egypt and the identified dominant serovar was S. enteritidis. The obtained data agreed with other reports outside Egypt, as in brazil Medeiros et al. (2011), isolated Salmonella in 0.0-8.9% from the collected chicken carcass parts and Le Bouquin et al. (2010); who carried out a nation-wide survey in France on the period from October 2005 to September 2006 and examined 370 randomly selected French commercial broiler chicken flocks to determine the prevalence of different Salmonella species and the results showed prevalence of different serotypes in 8.6% of the examined flocks. In Croatia, a study highlighted isolation of Salmonella from 66 samples of fresh, retail-cut chicken meat and frozen ground chicken meat with 10.6% incidence among the examined samples (Kozačinski et al., 2006). In Pakistan, Soomro et al. (2010); recorded Salmonella species in Hyderabad, Pakistan of 38% from poultry meat retail markets. Regarding post slaughter carriage rates the obtained

results matched with previous reports in different countries, where; Chenu et al., (2011); reported that the post slaughter carriage rates of Salmonella on broiler carcasses present with variable rates according to the country and continents with may correlated to the hygienic and processing practice; where the prevalence rate was 36.7% in Australia; 21% in the USA and 15.7% in Europe.; White et al. (2001); who recorded that 3.0% of the retail examined meat samples (turkey, chicken, beef and pork) were positive for different Salmonella strains in the Greater Washington region. USA; Kaushik et al. (2014); who reported that the incidence of Salmonella in chicken meat samples was 23.7% in India, Donado-Godoy et al. (2012); who recorded 27% of the carcasses sampled and Chilled chickens were positive for Salmonella in Colombia; The European Food Safety Authority; carried out a European Union-wide baseline prevalence survey on different strains of Salmonella on broiler carcasses, around 10132-broiler production batches were collected and examined from around 561 slaughterhouses in 26 different European Union (EU) Member States and from two countries not belonging to the EU; the survey analysis showed a salmonella Community prevalence of 15.61% with wide margin of variation in between the different EU states (0.0% to 26.6%) and in one state the prevalence rate reach 85.61% in the examined samples and the most predominant serotypes was S. infantis (Authority, 2010).

Salmonella strains were serotyped using poly and monovalent "O" and "H" antisera and the results revealed that 19 strains were serotyped as: S. typhimurium (5; 26.3%), S. kentucky (5; 26.3%), S. infantis (3; 15.8%), S. enteritidis (2; 10.5%), S. tamale and (2; 10.5%) and S. newport (2; 10.5%) as shown in Table 4. These results vary from country to another, where Medeiros et al. (2011) reported different percentage in brazil where S. enteritidis (48.82%), S. infantis (7.6%) and S. typhimurium (7.2%) from chicken, Ezzat et al. (2014); who reported that the serotyping of the Salmonellae from chickens were (8; 21.6%) S. enteritidis, (5; 13.5%) S. kentucky, (2; 5.4%) S. newport, were isolated from broilers. Where in EU; baseline Community prevalence survey of Salmonella showed the most predominate serotypes in broiler carcass was Salmonella infantis and Salmonella enteritidis and accounted as 1/3 and 1/6 of the Salmonella isolates, respectively (Authority, 2010); in France, Le Bouquin et al. (2010); reported that the most prevailing serovar was S. hadar followed by S. mbandaka and S. anatum ; while in Pakistan, Soomro et al. (2010); reported the most prevailing serogroups among chickens were S. typhi, S. enteritidis, S. typhimurium and S. pullorum. Mahmoud E. Ezzat et al. / American Journal of Animal and Veterinary Sciences 2020, 15 (2): 134.144 DOI: 10.3844/ajavsp.2020.134.144

Types of chicken meat	Types of samples	No. of examined samples	No. of positive samples	Proportion
Balady chicken	Liver	24	2	8.30
	Cecum	24	2	8.30
	Lung	24	2	8.30
	Breast	24	2	8.30
	Thigh	24	2	8.30
	Total	120	10	8.30
Broiler chicken	Liver	23	2	8.70
	Cecum	23	2	8.70
	Lung	23	0	0.00
	Breast	23	2	8.70
	Thigh	23	2	8.70
	Total	115	8	7.00
Frozen chicken	Breast	19	1	5.26
	Thigh	19	0	0.00
	Total	38	1	2.60
Total		273	19	6.95%

 Table 4: Numbers and percentage of different serotypes of Salmonellae isolated from chickens

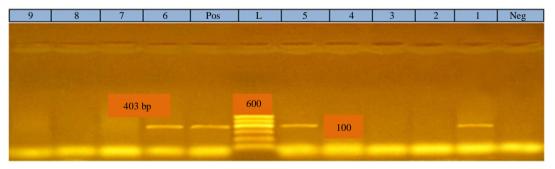
Isolated serotypes			
Serotype	No. of serotypes	%	
S. Typhimurium	5	26.3	
S. Enteritidis	2	10.5	
S. Kentucky	5	26.3	
S. Infantis	3	15.8	
S. Tamale	2	10.5	
S. Newport	2	10.5	
Total	19	100.0	

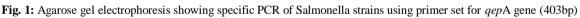
Regarding antimicrobial resistant among the examined 19 different salmonella isolates; the highest resistant to sulfamethoxazole (19/19; 100%), then resistance to tetracycline and amoxicillin (15/19; 78.9%), resistance to neomycin (12/19; 63.2%), resistance to chloramphenicol (8/19; 42.1%), resistance to doxycycline and colistin (5/19; 26.3%), resistance to ciprofloxacin and gentamycin (4/19; 21.1%) and the lowest resistant was to levofloxacin (3/19; 15.8%) as shown in Table 5. The data obtained from this study nearly in coordinated with Ahmed et al. (2014); who conducted a survey on salmonella MDR genes in Egypt and concluded that 47/69 Salmonella isolates (68.1%) showed MDR phenotypes to at least three different antimicrobial classes, Yang et al. (2002); who tested 22 S. typhimurium and 14 S. enteritidis isolates from animals and birds at Korea during the period from 1994 to 2001 for their antibiotic patterns and results showed high resistance to sulfisoxazole and tetracycline as 95% and 86%, respectively, Hui et al. (2015) who reported Salmonella strains resistance to doxycycline (28.21%) in central China and Sodagari et al. (2015); who observed in Iran, high antimicrobial resistance to tetracycline was 81% in Iran. However, the obtained results differ from de Oliveira et al. (2006); who tested The antimicrobial resistance of 79 S. enteritidis isolates from foods including beef meat and poultry meat individually and resistance observed was to gentamycin (12.71%) and neomycin (17.7%), Van et al. (2007); who isolated Salmonella from raw food samples including chicken and tested for their susceptibility to antibiotics in Vietnam and recorded that out of 91 serotypes, 40.7%, 22.1% and 16.5% were resistant to tetracycline, ampicillin and sulfafurazole respectively, Foley and Lynne (2008); who reported that multidrug resistance for ampicillin, chloramphenicol, sulfonamides, tetracycline and streptomycin was detected in 9.3% of Salmonella strains tested, Hyeon et al. (2011); who estimated the antibiotic resistance of Salmonellae isolated from chicken meat and the resistance observed to tetracycline and chloramphenicol (16.7%) in Seoul, South Korea in 2009, Capuano et al. (2013); who reported that none of the isolates were resistant to ciprofloxacin and enrofloxacin, while low resistance to gentamicin, ceftazidime (1.7% each), cephalothin and Colistin sulfate (2.6% each) and most resistance was to ampicillin (45.6%), tetracycline (48.21%), streptomycin (52.59%) and sulfonamides (53.5%) in Italy.

PCR is a perfect tool for accurate detection of Salmonella resistant genes revealed that genotypic analysis of Salmonella strains *qepA* (a resistant gene for quinolones) was reported with a percentage of (3 out of 9 isolates) 33.3% as in Fig. 1. Our result differs from Silva-Sanchez et al. (2011); who reported that no Salmonella isolates showed the presence of *qepA* during his study and Lunn et al. (2010); who found that one isolate (2.4%) showed the presence of qepA. The bla_{TEM} gene, a resistant gene encoded for B- lactamases was reported in this study with a percentage of 100% in all Salmonella serotypes as in Fig. 2. This result agreed but with higher percentage with previous report as in Canada. the percentage of bla_{TEM} gene in Salmonella isolates was 17% (Aslam et al., 2012), in Korea, 90.5% penicillin-resistant S. Enteritidis carried the *bla*_{TEM} gene (Hur *et al.*, 2011) and in china, *bla*_{TEM} gene detected in 51.6% of 62 ceftriaxone resistant Salmonella isolates (Yang et al., 2010). The tetA-gene, a gene encoded for tetracycline resistance was detected in this study with a percentage of 88.89% (8 out of 9 isolates) as in Fig. 3. This result nearly in coordinated with Shahada et al. (2006); who reported that in Japan oxytetracycline-resistant S. 89% of Infantis from poultry carried the tet (A) gene, while this result differs from a study performed by Zhang et al. (2011); who recorded the presence of tetA gene in Salmonella isolates from poultry with a percentage of 73.1% and Ahmed and Shimamoto (2012); who identified tetracycline resistance gene tetA in 14 out of 21 (66.7%) Salmonella isolates. The sull gene, a gene encoded for sulfonamide resistance was reported in this study with a

percentage of 77.78% (7 out of 9 isolates) as in Fig. 4. This result nearly similar with those of Zou et al. (2009); who detected sul1 in 11 of 16 isolates with a percentage of 68.7%. The aacC gene (a resistance gene for Aminoglycosides) was reported in this study with a percentage of 33.3% (3 out of 9 isolates) as in Fig. 5. This result differs from Lynne et al. (2008); who reported that aacC gene in 5 from 7 isolates with the percentage of 71.4%. The high rate of antimicrobial genes detection in salmonella isolated from human food (chicken meat) consider potential risk for the human health and narrow the available medication choices to human cases in addition to increase the risk of retransmission salmonella species from human to poultry farms through the infected or carrier people engaged with the poultry industry.

Antimicrobial agent	Number of resistant strains (NO)	Percentage of resistance strains	Number of sensitive strains (NO)	Percentage of sensitive strains (%)
Levofloxacin (Lev)	3	15.8	15	78.9
Sulfamethazine (Smz)	19	100.0	0	0.0
Tetracycline(T)	15	78.9	4	21.1
Amoxicillin (AX)	15	78.9	3	15.8
Neomycin (N)	12	63.2	5	26.3
Doxycycline (Do)	5	26.3	9	47.4
Ciprofloxacin (Cip)	4	21.1	11	57.9
Chloramphenicol (C)	8	42.1	9	47.4
Colistin (CL)	5	26.3	9	47.4
Gentamycin (Cn)	4	21.1	15	78.9





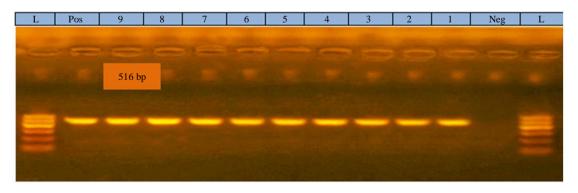


Fig. 2: Agarose gel electrophoresis showing specific PCR of Salmonella strains using primer set for blaTEM gene (516 bp)

Mahmoud E. Ezzat et al. / American Journal of Animal and Veterinary Sciences 2020, 15 (2): 134.144 DOI: 10.3844/ajavsp.2020.134.144

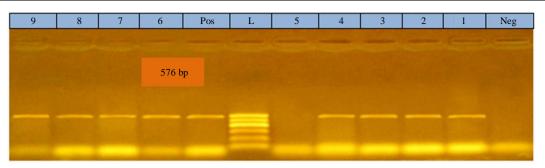


Fig. 3: Agarose gel electrophoresis showing specific PCR of Salmonella strains using primer set for tetA (A) gene (576 bp)

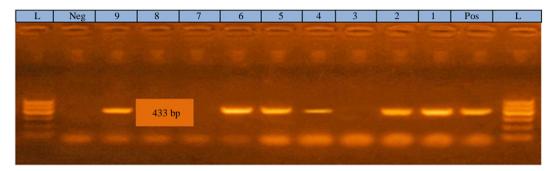


Fig. 4: Agarose gel electrophoresis showing specific PCR of Salmonella strains using primer set for sul1 gene (433 bp)

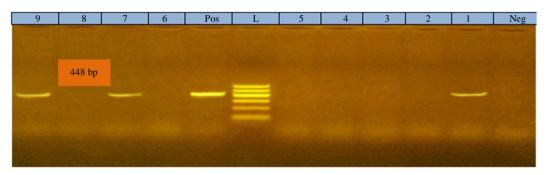


Fig. 5: Agarose gel electrophoresis showing specific PCR of Salmonella strains using primer set for gene aacC (448 bp)

Conclusion

Salmonellosis is an important global public health problem that has negative effects in poultry industry all over the world. As a result of extensive and in proper usage of antimicrobial agents in human and veterinary medicine specially as growth promotors in poultry feeds, there are dramatically increase in the MDR *Salmonella*; so that this study was focused on detection of some resistance genes to different antimicrobial agents that used in poultry farms such as *qepA* for quinolones, *sul1* for sulfonamide, *bla_{TEM}* for β - lactams, *aacC* for aminoglycosides and *tetA* (*A*) for tetracycline. Because of much higher resistance of *Salmonella* to different antimicrobial agents that reported in this study using sensitivity tests and PCR it's recommended to use antimicrobial agents in proper dose for recommended

time after applying sensitivity tests on the isolated *Salmonellae* to avoid these resistance that have adverse effect on poultry industry.

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Author's Contributions

All authors equally contributed in this work.

Ethics

The experiment design approved by the Suez canal university ethical committee.

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