

Bacteriological Studies on Pathogens in Egyptian Pigeons.

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The prevalence rate of bacterial isolates of public health importance in pigeons was (28.16%). The incidence of bacterial pathogens differed according to health status of examined pigeons and ages either squabs or adults, as it gave the higher incidence in freshly dead squabs (33.33%) and in adults (28.57%) followed by diseased squabs (31.03%) and adults (26.67%) then finally slaughtered pigeons (25.56%). There was a wide range of bacterial pathogens isolated from nasal and cloacal swabs of diseased pigeons including *C. jejuni*, *Citrobacter freundii*, *D. pneumoniae*, *E. coli*, *K. oxytoca*, *K. pneumoniae*, *Mannheimia haemolytica*, *P. aeruginosa*, *Salmonella spp.*, *S. aureus* and *Y. enterocolitica*. There were variations between the incidence and the species of pathogens isolated from cloacal and nasal swabs either in squabs or in adults *K. oxytoca*, *Mannheimia haemolytica* and *Y. enterocolitica* never isolated from adult. It was appeared that the deaths usually occurred due to combination of more than one bacterium. On the examination of internal organs slaughtered pigeons, there were differences in the incidences of bacterial isolation from different organs. Serological identification of most prevalent isolates revealed 5 *Salmonella* serovars including, 3 *P. aeruginosa* serogroups and 6 *E. coli* serogroups. All examined pathogens were sensitive to enrofloxacin followed by gentamicin then ciprofloxacin. In contrast, streptomycin then erythromycin and colistin sulphate showed the lowest effect. Among the isolates tested, *P. aeruginosa* was resistant to the most used antibiotics..Most isolated strains of *E. coli*, *P. aeruginosa*, *Salmonella spp.* and *Y. enterocolitica* from pigeons were elaborating enterotoxin. *S. paratyphi A* and *S. typhimurium* var. copenhagen were 100% enterotoxigenic followed by *S. typhimurium*(83.33%) , *E. coli* O₈ and *Ps. aeruginosa* I (75%) in each. On other hand, lower enterotoxin production was observed in *Ps. aeruginosa* A (46.15%) and *E. coli* O₁₁₁ (44.44%).

Pigeons are widely distributed in Egypt and considered as an important bird for many people especially in fest, for hunting and racing, many farmers reared them in their houses and considered an essential food for rich people. Pigeons are potential reservoirs for several pathogenic microorganisms, their faeces and other body fluids may harbour several pathogenic bacteria, which contributed to contaminate ponds, lacks, parks, garden and soils. Tanaka *et al.*, (2005) considered pigeons faeces as a source of several zoonotic agents for birds, animals and humans especially *Salmonella*, *E. coli* O157 as well as *Mycobacterium spp.* also pigeons considered as a possible reservoir of Shiga toxin producing *E. coli* pathogenic to human (Sonntag *et al.*, 2005). *Campylobacter jejuni* was the most frequently isolated microorganism, found in pigeons (Casanovas *et al.*, 1995). In human, these bacteria can cause gastroenteritis, respiratory symptoms, septicemia, and even mortality, also, they cause low production and mortalities in infected birds, as *S. Typhimurium* var. Copenhagen mostly causes intestinal problems,

diarrhoea and can also penetrate the intestinal tract and cause internal and neurotic problems.

Uncontrolled use of antibiotics in medicine and animal husbandry over the course of decades has fostered the selection of resistant bacteria (Tomasz, 1994). The rise in multi-drug resistant pathogenic bacteria is of global concern, because it can lead to increase human and domestic animals health care costs and increased morbidity and mortality (Williams and Heymann, 1998). Zoonotic bacteria especially salmonellae were shown to survive and multiply in the dropping for up to one month after their deposition by pigeons and the dropping distributed over a large environmental conditions. Hence, the goal of this study was conducted to the following items.

Bacteriological examination of samples collected from Egyptian pigeons and complete identification of the pathogenic isolates of public health importance, serological identification of some pathogenic isolates and determination of antimicrobial susceptibility also insurance of the virulence of the most public health important pathogenic isolates by inoculation in

experimentally laboratory animals (mice and chicks), finally detection of enterotoxigenic strains of some pathogenic isolates.

Materials and methods

Samples. A total of 778 samples were collected from 206 pigeons in different farmer houses, Giza zoo and poultry shops located throughout Egypt during the period from November, 2006 up to April, 2007. These samples were classified into groups as follows:

Diseased pigeons. The samples were collected from 58 squabs and 30 adult pigeons which showed signs of illness as nasal discharges, diarrhea (as green and watery droppings), dull and ruffled feathers. Samples included nasal and cloacal swabs from each bird.

Slaughtered and freshly dead pigeons. The samples were collected from 90 slaughtered and (21 squabs and 7 adults) of freshly dead pigeons from either poultry shops or farmer houses. Samples included crops, pharyngeal swabs, lungs, livers, intestines and oviducts. Each sample was collected in sterile polyethylene plastic bag, while samples for Campylobacter examination were collected in sterile screw capped bottles containing transport broth with supplement. All samples were transferred in an icebox to the laboratory without delay.

Bacteriological examination.

Isolation and identification of Campylobacter.

All samples in transport broth Campylobacter medium were cultured directly onto modified Campylobacter charcoal differential agar medium (C.C. D.A.) with supplement in duplicate. The inoculated plates were incubated at 37°C and 42°C for 72 hours in 10% CO₂ tension in anaerobic jar using gas generating kit. Motility test was done by using hanging drop technique to see the corkscrew like motion specific for Campylobacter and Gram's stain was also done to detect the characteristic comma shape of Campylobacter for all suspected colonies. The isolates were identified according to Koneman *et al.*, (1997).

Isolation and identification of other bacteria.

Each sample was cultured in peptone broth and incubated at 37°C for 24 hours then a loopfull was plated onto following solid media according to its specificity: Nutrient agar, 5% sheep blood agar, MacConkey agar, DAS media, mannitol salt agar, S.S agar, medium contain cetrimid for *P. aeruginosa* and Yersinia selective agar with selective supplement. On the other hand, samples used for salmonellae examination were firstly incubated in Rappabort Vassiliadis at 42°C for

24 hours then plated onto S.S. agar. All inoculated plates were incubated aerobically at 37°C for 24- 48 hours. The colonies were identified and confirmed biochemically according to Koneman *et al.*, (1997); Boone and Castenholz (2001); Quinn *et al.*, (2002).

Serological identification of the most isolates.

E. coli and *P. aeruginosa* isolates were serogrouped according to the procedure outline by Edwards and Ewing (1972); Homma (1982) respectively. Meanwhile, Salmonella isolates were serologically identified according to Kauffmann - White scheme as described by Kauffmann (1973). The typing antisera were obtained from Denka Seiken Co. Ltd, Tokyo, Japan using agglutination technique.

Antimicrobial susceptibility.

This was done using disc diffusion standard technique according to Finegold and Martin (1982); Quinn *et al.*, (2002) with Mueller Hinton medium, in Campylobacter investigation the medium was supplemented with 5% sheep blood using the following (Oxoid) discs, cefadroxil (30µg), chloramphenicol (30µg), ciprofloxacin (5µg), colistin sulphate (50µg), doxycycline (30µg), enrofloxacin (5µg), erythromycin (15µg), flumequine (30µg), gentamicin (10µg), nitrofurantion (300µg), nalidixic acid (30µg) and streptomycin (10µg). the results were interpreted according to the manual supplied by Oxoid Company.

Pathogenicity test.

Mice (mouse lethality test). Two hundred & sixty four (264) albino white mice with average weight of about 18- 20 g and aged 28-30 days old were used to investigate the pathogenicity of various strains of 22 pathogens including *C. jejuni*, *Citrobacter freundii*, *D. pneumoniae*, *E. coli* serogroups (O8, O78, O86, O111, O157 and O166) *K. oxytoca*, *K. pneumoniae*, *Mannheimia haemolytica*, *P. aeruginosa* A, *P. aeruginosa* G, *P. aeruginosa* I, *S. Kentucky*, *S. Paratyphi A*, *S. Typhimurium*, *S. Typhimurium* var. *Copenhagen*, *S. Virginia*, *S. aureus* and *Y. enterocolitica*. All mice were examined bacteriologically to ensure their freedom from pathogens. The mice were divided into groups each of 3 mice and inoculated I.P. with 0.1 ml of 5 X 10⁸ C. F. U/ mouse of the tested strain and kept separately, last group was kept as control and injected only with saline. Mice were kept under observation for 7-10 days, the numbers of dead mice were recorded and re-isolation of inoculated strains was done.

Chick. Chicks of 3 days old obtained from El-

Table (1): Prevalence rate of bacterial isolates of public health importance in pigeons.

Health status of examined pigeons	Age	No. of examined pigeons	Bacteriologically Positive	
			No.	%*
Freshly dead	Squabs	21	7	33.33
	Adults	7	2	28.57
Diseased	Squabs	58	18	31.03
	Adults	30	8	26.67
Slaughtered	Adults	90	23	25.56
Total		206	58	**28.16

*The percentage was calculated according to the number of each type of examined pigeons.

**The percentage was calculated according to the total number of examined pigeons.

Wadi Company. Ten chicks were randomly taken and subjected to clinical, postmortem and bacteriological examination, which proved to be apparently healthy. All chicks were examined bacteriologically to exclude any bacterial pathogens. Four hundred & forty four (440) chicks were divided into equal groups containing 5 chicks in each. Chicks of each group were individually infected per os with 0.5 ml containing (5×10^8 C.F.U.) of the tested strain, last group was kept as control and injected only with saline. Chicks of all groups were kept under observation for 12 days, clinical signs and mortalities were recorded and postmortem and bacteriological examination were done to dead bird. The study was terminated at 12 days post infection and re-isolation of infected strain was done.

Infant mouse assay. Detection of heat stable enterotoxin by Robins-Brown *et al.*, (1993). Preparation of culture filtrate via inoculation of each isolate in tryptone soya broth and incubated over night at 37°C. Then 10 ml of culture was placed in 200 ml of medium containing 2% casamino acid, 1% yeast extract and 0.4% glucose pH (8.5) in 250 ml flask. The inoculated flasks were incubated on a rotatory shaker 200 rpm at 37°C for 18 hours then centrifuged at 12000 x g for 10 minutes. The supernatant was filtrated through Millipore membrane filter pore 0.45µm and stored at -20°C until used. A part of sterile medium was used as control. Infant mouse assay 0.1 ml of each filtrate was injected through the abdominal wall into milk filled stomach of each 3 mice 2-4 days old for each examined strain and 3 infant mice were injected by 0.1 ml of sterile medium and were used as negative control. After 4 hours, the mice were killed and the entire intestine was removed. The intestine and remaining body were weight to calculate the ratio of intestine weight/remaining

body weight. Ratio greater than (0.083) was recorded as positive test for enterotoxin.

Results and Discussion

Pigeons are free-living birds so spreads of infectious agents occur through faecal contamination of drinking water sources, pastures and agricultural crops and may also come into close contact with domestic birds enabling direct transfer of infectious agents to take place, especially when they are kept out of doors. In general the prevalence rate of bacterial isolates of public health importance in pigeons was (28.16%), this incidence may be indicated that pigeons mainly affected by viral and parasitic causes other than bacterial one. The incidence of bacterial pathogens differed according to health status of examined pigeons and age either squabs or adults, as it gave the higher incidence in freshly dead squabs (33.33%) and in adults (28.57%) followed by diseased squabs (31.03%) and adults (26.67%) then finally slaughtered pigeons (25.56%). This may be attributed to the location of samples collection, husbandry, farmer management and kind of feed and finally to grassing area, as represented in table (1). It was clear that incidence of bacterial pathogens in squabs was higher than adults which may be due to lower immunity in young.

The data achieved in table (2) showed that there was a wide range of bacterial pathogens isolated from nasal and cloacal swabs of diseased pigeons including *C. jejuni*, *Citrobacter freundii*, *D. pneumoniae*, *E. coli*, *K. oxytoca*, *K. pneumoniae*, *Mannheimia haemolytica*, *P. aeruginosa*, *Salmonella* spp, *S. aureus* and *Y. enterocolitica*. There were variations between the incidence and the species of pathogens isolated from cloacal and nasal swabs either in squabs or in adults. *K. oxytoca*, *Mannheimia haemolytica* and *Y. enterocolitica* never isolated from adults.

Table (2): Prevalence rate of bacterial pathogens of public health importance isolated from diseased pigeons.

Isolated Pathogens	Nasal		Cloacal					
	Squabs (58)		Adults (30)		Squabs (58)		Adults (30)	
	No.	%*	No.	%**	No.	%*	No.	%**
<i>C. jejuni</i>	0	0	0	0	4	6.70	2	6.66
<i>Citrobacter freundii</i>	0	0	0	0	2	3.45	1	3.33
<i>D. pneumoniae</i>	3	5.17	1	3.33	0	0	0	0
<i>E. coli</i>	3	5.17	1	3.33	5	8.62	3	10.00
<i>K. oxytoca</i>	0	0	0	0	1	1.72	0	0
<i>K. pneumoniae</i>	3	5.17	1	3.33	0	0	0	0
<i>Mannheimia haemolytica</i>	1	1.72	0	0	0	0	0	0
<i>P. aeruginosa</i>	2	3.45	1	3.33	2	3.45	1	3.33
<i>Salmonella spp</i>	0	0	0	0	3	5.17	1	3.33
<i>S. aureus</i>	3	5.17	2	6.66	0	0	0	0
<i>Y. enterocolitica</i>	0	0	0	0	1	1.72	0	0

*The percentage was calculated according to the total number of examined diseased squabs (58).

**The percentage was calculated according to the total number of examined diseased adult pigeons (30).

In this concern, Casanovas *et al.*, (1995) recorded that *C. jejuni* was the most isolates found in 105 pigeons (26.2%) then *Salmonella spp.* were isolated from six specimens (1.5%) and *Yersinia spp.* was isolated from only one pigeon in the city of Barcelona. In addition, Adesiyun *et al.*, (1998) mentioned that (5%) of racing pigeons that originated from 8 fanciers in Trinidad yielded *Salmonella spp.* all of which were *S. Typhimurium* while only (1%) was positive for *Campylobacter spp.* Otherwise, Methner and Lauterbach (2003) isolated *Salmonella* in an incidence of (7.04%) from faecal samples in purebred pigeons. While, Dove *et al.*, (2004) recorded that *Salmonella spp.* were isolated from (5.7%) of the cloacal swabs of free-living pigeons in the city of Ljubljana, Slovenia. Otherwise, Pasmans *et al.*, (2004) isolated (22.8%) of *S. Typhimurium var. Copenhagen* from pooled faecal samples from pigeon lofts from the city of Ghent (Belgium). Furthermore, Sonntag *et al.*, (2005) considered the pigeons as a possible reservoir of Shiga toxin 2f-producing *E. coli* associated with human disease. Meanwhile, Lillehaug *et al.*, (2005) detected *Campylobacter jejuni* in fresh faecal samples of six out of 200 feral pigeons while all samples were negative for salmonellae. In addition, Tanaka *et al.*, (2005) isolated salmonellae from (3.9%) of faecal samples from feral pigeons in Japan. McCrea *et al.*, (2006) isolated *Campylobacter* and *Salmonella spp.* from live bird to prepackaged carcass for 3

flocks of squab. Pedersen *et al.*, (2006) detected *S. enterica* in an incidence of (3.2%) from pigeons captured in Fort Collins, Colorado.

Concerning the results in tables (3-4) *C. jejuni*, *Citrobacter freundii*, *D. pneumoniae*, *E. coli*, *K. oxytoca*, *K. pneumoniae*, *Mannheimia haemolytica*, *P. aeruginosa*, *Salmonella spp.*, *S. aureus* and *Y. enterocolitica* were isolated from freshly dead squabs. While in 2 freshly dead adult pigeons *C. jejuni*, *E. coli*, *Salmonella spp.*, and *S. aureus* were isolated. From this data, it was appeared that the deaths usually occurred due to combination of more than one bacterium. The prevalence rate of *E. coli* was very high, this may be attributed to the fact that *E. coli* overgrows the normal intestinal flora and could produce toxins that could reach the blood and caused intoxication, so young pigeons become sick and died. In addition, *S. Typhimurium* could penetrate the intestinal tract and caused internal and neurotic problems in addition to deaths. The result proved that these pathogens not only constitute public hazard for human beings but also cause deaths of birds. It is very important to get rid of dead pigeons in good hygienic measures to prevent spread of pathogenic organisms to human and other livestock and avoid contamination of environment. This data agree with those reported by Pennycott *et al.*, (2005) who recovered *S. Typhimurium* from 8 carcasses and *E. coli* O86 from 3 carcasses found in dead birds in south-west Scotland.

Table (3): Prevalence rate of bacterial pathogens of public health importance isolated from freshly dead squabs.

Isolated pathogens	Crops		Pharyngeal swabs		Lungs		Livers		Intestines	
	No.	%	No.	%	No.	%	No.	%	No.	%
<i>C. jejuni</i>	1	4.76	0	0	0	0	2	9.52	3	14.29
<i>Citrobacter freundii</i>	1	4.76	0	0	0	0	1	4.76	1	4.76
<i>D. pneumoniae</i>	0	0	1	4.76	2	9.52	1	4.76	0	0
<i>E. coli</i>	2	9.52	2	9.52	3	14.29	4	19.05	5	23.81
<i>K. oxytoca</i>	0	0	0	0	0	0	1	4.76	1	4.76
<i>K. pneumoniae</i>	0	0	1	4.76	1	4.76	1	4.76	0	0
<i>Mannheimia haemolytica</i>	0	0	0	0	2	9.52	1	4.76	0	0
<i>P. aeruginosa</i>	1	4.76	1	4.76	1	4.76	2	9.52	2	9.52
<i>Salmonella spp</i>	1	4.76	0	0	0	0	2	9.52	4	19.05
<i>S. aureus</i>	0	0	1	4.76	3	14.29	1	4.76	0	0
<i>Y. enterocolitica</i>	0	0	0	0	0	0	1	4.76	1	4.76

The percentage was calculated according to the total number of examined freshly dead squabs (21).

Table (4): Prevalence rate of bacterial pathogens of public health importance isolated from freshly dead adult pigeons.

Isolated pathogens	Crops		Pharyngeal swabs		Lungs		Livers		Intestines	
	No.	%	No.	%	No.	%	No.	%	No.	%
<i>C. jejuni</i>	1	14.29	0	0	0	0	1	14.29	1	14.29
<i>E. coli</i>	1	14.29	1	14.29	1	14.29	2	28.57	2	28.57
<i>Salmonella spp</i>	0	0	0	0	0	0	2	28.57	2	28.57
<i>S. aureus</i>	0	0	1	14.29	1	14.29	1	4.76	0	0

The percentage was calculated according to the total number of examined freshly dead adult pigeons (7).

On the examination of internal organs including crops, pharyngeal swabs, livers, intestines and lungs of slaughtered pigeons, it was clear that there were differences in the incidences and organs from which the following pathogens *C. jejuni*, *D. pneumoniae*, *E. coli*, *K. oxytoca*, *K. pneumoniae*, *Mannheimia haemolytica*, *P. aeruginosa*, *Salmonella spp*, *S. aureus* and *Y. enterocolitica* were isolated as achieved in table (5). Many investigators concerned with bacteria isolated from slaughtered pigeons as Jeffrey *et al.*, (2001) on microbiological testing for *Campylobacter* and *Salmonella* in squabs processing plant in three separate trials, found that the overall prevalence of positive samples in trial (1) was (1.4%) for *Salmonella spp.* and (11.1%) for *C. jejuni* in trial (2) (4.3 and 0%) for *Salmonella spp.* and *C. jejuni* and in trial (3) (4.1 and 4.8%) for *Salmonella spp.* and *C. jejuni* respectively. Otherwise, Pasmans *et al.*, (2004) isolated (3.3%) *S. Typhimurium* var. Copenhagen from pooled samples of livers, intestines and spleens of feral pigeons from the city of Ghent

(Belgium). Furthermore, Losito *et al.*, (2005) isolated *Staphylococcal* strains from a pigeon slaughterhouse in central Italy.

Otherwise, Soncini *et al.*, (2006) isolated *Campylobacter* from (15.8%) of pigeon s neck skin, (12.5%) of pigeon s meat samples. The isolation of these pathogens from slaughtered pigeons may be attributed to their illness before slaughtering or contamination during slaughtering due to bad hygienic measures, using of contaminated utensils or surface contamination during evisceration of the birds from rupture of intestinal tract or occur via infected human during handling of raw pigeons.

The presences of the pathogens in oviduct were very important as it may disseminate these pathogens to eggs causing economic losses and causing zoonotic diseases to consumers. *E. coli*, *P. aeruginosa* and *Salmonella spp.* were isolated from oviducts of examined slaughtered pigeons. Meanwhile only *E. coli* and *Salmonella spp.* were isolated from oviducts of examined dead pigeons, as demonstrated in table(6).

Table (5): Prevalence rate of bacterial pathogens of public health importance isolated from slaughtered pigeons.

Isolated pathogens	Crops		Pharyngeal swabs		Lungs		Livers		Intestines	
	No.	%	No.	%	No.	%	No.	%	No.	%
<i>C. jejuni</i>	2	2.22	0	0	0	0	5	5.56	8	8.89
<i>D. pneumoniae</i>	0	0	2	2.22	7	7.78	4	4.44	0	0
<i>E. coli</i>	4	4.44	6	6.66	5	5.56	6	6.67	8	8.89
<i>K. oxytoca</i>	2	2.22	2	2.22	0	0	2	2.22	5	5.56
<i>K. pneumoniae</i>	0	0	2	2.22	4	4.44	3	3.33	0	0
<i>Mannheimia haemolytica</i>	0	0	1	1.11	3	3.33	2	2.22	0	0
<i>P. aeruginosa</i>	1	1.11	2	2.22	3	3.33	2	2.22	3	3.33
<i>Salmonella spp</i>	2	2.22	0	0	0	0	3	3.33	4	4.44
<i>S. aureus</i>	0	0	5	5.56	8	8.89	6	6.67	0	0
<i>Y. enterocolitica</i>	1	1.11	0	0	0	0	2	2.22	3	3.33

The percentage was calculated according to the total number of examined slaughtered pigeons. (90).

Table (6): Prevalence rate of bacterial pathogens of public health importance isolated from oviducts of examined pigeons.

Isolated Pathogens	Type	Slaughtered (n = 10)		Dead (n = 2)	
		No.	%	No.	%
<i>E. coli</i>		5	50	2	100
<i>P. aeruginosa</i>		1	10	0	0
<i>Salmonella spp.</i>		2	20	1	50

The percentage was calculated according to the number of each type of tested pigeons.

Table (7) Serological identification of most prevalent isolates.

Serovars	Salmonella serovars		<i>P. aeruginosa</i>			<i>E. coli</i>		
	NO.	%	Serogroups	NO.	%	Serogroups	NO.	%
<i>S. kentucky</i>	3	11.11	A	11	44	O8	12	16.90
<i>S. paratyphi A</i>	3	11.11	G	7	28	O78	15	21.13
<i>S. typhimurium</i>	12	44.44	I	4	16	O86	13	18.31
<i>S. typhimurium</i> var. Copenhagen	7	25.93	Untyped	3	12	O111	9	12.68
<i>S. virgina</i>	2	7.41				O157	7	9.86
						O166	13	18.31
						Untyped	2	2.81
Total	27	100		25	100		71	100

N.B. Number of isolate was from all examined samples even in the same bird and the percentage was calculated according to the total number of each isolate.

The results presented in table (7) revealed 5 Salmonella serovars including *S. kentucky*, *S. paratyphi A*, *S. typhimurium*, *S. typhimurium* var Copenhagen and *S. virgina*. On identification of *P. aeruginosa* isolates serogroups A, G and I were identified. On the other hands, *E. coli* isolates were serogrouped as O8, O78, O86, O111, O157 and O166. These results were partially agree with that reported by Methner and Lauterbach (2003) who found that all

Salmonella isolated from faecal samples in purebred pigeons belonged to the serovar Typhimurium var. Copenhagen. In addition, Tanaka *et al.*, (2005) isolated *S. typhimurium* and *S. cerro* from feral pigeons in Japan. Meanwhile, similar results were reported by Pasmans *et al.*, (2004) who isolated (22.8%) of *S. typhimurium* var. Copenhagen from pooled faecal samples from pigeon lofts from the city of Ghent (Belgium).

The antimicrobial susceptibility of many strains of 22 pathogens isolated from pigeons revealed variable results in susceptibilities and zones of inhibition to different chemotherapeutic agents and antibiotics, all examined pathogens were sensitive to enrofloxacin, followed by gentamicin then ciprofloxacin. In contrast to streptomycin then erythromycin and colistin sulphate showed the lowest effect. Among the isolates tested, *P. aeruginosa* was resistant to the most used antibiotics, as illustrated in table (8). In general, there were good susceptibilities to many examined agents these might due to fact that pigeon not take antibiotic continuously as other animals. In this aspect Kimpe *et al.*, (2002) on examination of antimicrobial susceptibility of 60 *E. coli* and 18 *S. Typhimurium* var. Copenhagen strains isolated from homing pigeons, found that *E. coli* strains was resistant to all antibiotics tested. Over one-half of them were resistant to tetracycline, penicillins, and ampicillin, however, none showed extended spectrum beta-lactamase activity, implying that the cephalosporins remained active. Resistance to trimethoprim, aminoglycosides and fluoroquinolone ranked next. In contrast to the *E. coli* strains, the *S. enterica* strains were susceptible to all the antimicrobials tested. Also, Seepersadsingh and Adesiyun (2003) recorded that the highest prevalence of resistance was (83.3%) to streptomycin among salmonella isolates from birds. Meanwhile, Losito *et al.*, (2005) mentioned that *S. aureus* isolated from pigeon slaughterhouses in central Italy were sensitive to amoxicillin/clavulanic acid, cephalothin, gentamicin, kanamycin, oxacillin, rifampicin, tobramycin, trimethoprim-sulfamethoxazole, vancomycin. Some (15.2%) of the strains were resistant to ampicillin and to penicillin G; (6.8%) were resistant to chloramphenicol, (20.3%) to enrofloxacin, (16.9%) to erythromycin and to ciprofloxacin, (8.5%) to clindamycin, and (11.9%) to lincomycin. The highest percentages of strains were resistant to tetracycline (37.3 %). Only one strain had a multiple antibiotic resistance. The results achieved in tables (9-10) showed the virulence of different pathogens as demonstrated by the sequence of mortality of mice injected with test pathogens separately I/P. It was clear that *D. pneumoniae*, *E. coli* O157, *Mannheimia haemolytica*, *P. aeruginosa* I, *S. paratyphi* A and *S. typhimurium* var. Copenhagen were highly virulent strains for mice followed by *E. coli* O78 and *P. aeruginosa* G. On the other hand, *K. oxytoca* and *Y. enterocolitica* then *C. jejuni* were

lower in their virulence. In addition, showed the pathogenicity of the same pathogens in chicks it was appeared that there were great variations in morbidity and mortality rates and case fatality rate with different examined isolates. *P. aeruginosa* I showed (100%) mortality and case fatality rate then (90%) of *S. typhimurium* var. Copenhagen. Meanwhile, *K. oxytoca* and *S. kentucky* gave (0%). These variations in the data of pathogenicity tests may be attributed to pathogenic nature of the examined strains or the presence of virulence associated plasmid or production of endo or exotoxins and finally type of used laboratory animals. Similar result was reported by Helm *et al.*, (2004) who found that *S. typhimurium* frequently associated with pigeon infections were still virulent in mice. Otherwise, Pasmans *et al.*, (2004) recorded that *S. typhimurium* var. Copenhagen strains isolated from feral pigeons were the high virulence for mice. The present investigation achieved in table (11) shows that the most isolated strains of *E. coli*, *P. aeruginosa*, *Salmonella* spp. and *Y. enterocolitica* from pigeons were elaborating enterotoxin. *S. paratyphi* A and *S. typhimurium* var. Copenhagen were (100%) enterotoxigenic followed by *S. typhimurium* (83.33%), *E. coli* O8 and *P. aeruginosa* I (75%) in each. On other hand, lower enterotoxin production was observed in *P. aeruginosa* A (46.15%) and *E. coli* O111 (44.44%). It could be concluded that there is a potentially high risk of human exposure to excreta of pigeons that may serve as reservoirs for many zoonotic pathogens, so that live pigeons must be subjected to periodical examination. In addition, the importance of cleaning and sanitation programs and personal hygiene in farmer houses and poultry shops to avoid contamination and to get ride of dead pigeons in correct hygienic measures to avoid contamination of environment and spread of diseases to human and other live stocks.

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Table (8) Antimicrobial susceptibility of the most common bacterial isolates.

Antibiotic and chemotherapeutic agents Pathogens	Cefadroxil (30)		Chloramphenicol (30)		Ciprofloxacin (5)		Colistin sulphate (50)		Doxycycline (30)		Enrofloxacin (5)		Erythromycin (15)		Flumequine (30µg)		Gentamicin (10)		nitrofurantion (300µg)		Nalidixic acid (30)		Streptomycin (10)	
	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%
	<i>C. jejuni</i> 10	0	0	6	60	0	0	0	0	10	100	10	100	1	10	0	0	8	80	4	40	2	20	0
<i>Citrobacter freundii</i> 3	2	66.7	2	66.7	1	33.3	1	33.3	3	100	3	100	0	0	2	66.7	1	33.3	2	66.7	2	66.7	1	33.3
<i>D. pneumoniae</i> 7	6	85.7	5	71.4	2	28.6	0	0	5	71.4	7	100	0	0	0	0	1	14.3	0	0	0	0	1	14.3
<i>E. coli</i> O8 4	3	75	2	50	3	75	2	50	3	75	4	100	1	25	3	75	4	100	2	50	2	50	1	25
<i>E. coli</i> O78 5	3	60	1	20	4	80	0	0	3	60	5	100	0	0	3	60	5	100	3	60	3	60	1	20
<i>E. coli</i> O86 4	2	50	3	75	4	100	2	50	2	50	4	100	2	50	2	50	2	50	2	50	2	50	2	50
<i>E. coli</i> O111 3	3	100	2	66.7	3	100	1	33.3	3	100	3	100	2	66.7	2	66.7	3	100	1	33.3	0	0	0	0
<i>E. coli</i> O157 3	2	66.7	1	33.3	3	100	1	33.3	1	33.3	3	100	0	0	2	66.7	3	100	2	66.7	1	33.3	0	0
<i>E. coli</i> O166 4	2	50	2	50	4	100	2	50	2	50	4	100	1	25	3	75	4	100	2	50	2	50	1	25
<i>K. oxytoca</i> 3	2	66.7	0	0	3	100	0	0	2	66.7	3	100	0	0	3	100	3	100	2	66.7	1	33.3	0	0
<i>K. pneumoniae</i> 5	4	80	0	0	4	80	0	0	3	60	5	100	0	0	3	60	5	100	4	80	2	40	0	0
<i>Mannheimia haemolytica</i> 3	0	0	0	0	1	33.3	0	0	3	100	2	66.7	0	0	0	0	2	66.7	0	0	0	0	0	0
<i>P. aeruginosa</i> A 4	0	0	0	0	0	0	0	0	0	0	4	100	0	0	0	0	4	100	0	0	0	0	2	50
<i>P. aeruginosa</i> G 3	0	0	0	0	0	0	0	0	0	0	3	100	0	0	0	0	3	100	0	0	0	0	1	33.3
<i>P. aeruginosa</i> I 2	0	0	0	0	0	0	0	0	0	0	2	100	0	0	0	0	2	100	0	0	0	0	1	50
<i>S. Kentucky</i> 2	0	0	1	50	2	100	2	100	0	0	2	100	0	0	0	0	1	50	0	0	0	0	0	0
<i>S. Paratyphi</i> A 2	2	100	2	100	2	100	1	50	2	100	2	100	0	0	2	100	1	50	0	0	0	0	0	0
<i>S. Typhimurium</i> 3	0	0	2	66.7	3	100	0	0	3	100	3	100	0	0	3	100	1	33.3	1	33.3	3	100	2	66.7
<i>S. Typhimurium</i> var. Copenhagen 2	1	50	1	50	2	100	0	0	1	50	2	100	0	0	2	100	1	50	0	0	1	50	2	100
<i>S. Virginia</i> 2	0	0	2	100	2	100	1	50	0	0	2	100	0	0	0	0	1	50	0	0	1	50	0	0
<i>S. aureus</i> 10	6	60	8	80	8	80	0	0	7	70	10	100	5	50	8	80	10	100	9	90	4	40	5	50
<i>Y. enterocolitica</i> 3	2	66.7	3	100	1	33.3	0	0	3	100	3	100	2	66.7	2	66.7	3	100	1	33.3	3	100	1	33.3

*Concentration of used discs in µg.

**No. and percentage of sensitive strains to examined chemotherapeutic & antibiotic disc

Table (9) Pathogenicity of the bacterial pathogens of public health importance isolated from pigeons in mice.

Pathogens	No. of examined strains*	No. of examined mice	No. of dead mice per day											Total dead mice	
			1	2	3	4	5	6	7	8	9	≥10	No.	%**	
<i>C. jejuni</i>	10	30	0	4	2	1	1	0	0	0	0	0	0	8	26.67
<i>Citrobacter freundii</i>	3	9	0	1	1	2	1	0	0	0	0	0	5	55.56	
<i>D. pneumoniae</i>	7	21	8	5	3	5	0	0	0	0	0	0	21	100	
<i>E. coli O8</i>	4	12	0	3	2	2	1	1	0	0	0	0	9	75	
<i>E. coli O78</i>	5	15	5	4	3	2	0	0	0	0	0	0	14	93.33	
<i>E. coli O86</i>	4	12	3	2	2	1	1	0	0	0	0	0	9	75	
<i>E. coli O111</i>	3	9	0	0	2	2	0	1	1	0	0	0	6	66.67	
<i>E. coli O157</i>	3	9	3	2	2	2	0	0	0	0	0	0	9	100	
<i>E. coli O166</i>	4	12	0	1	2	1	2	1	1	0	0	0	8	66.67	
<i>K. oxytoca</i>	3	9	1	0	1	1	0	0	0	0	0	0	3	33.33	
<i>K. pneumoniae</i>	5	15	4	3	2	1	0	0	0	0	0	0	10	66.67	
<i>Mannheimia haemolytica</i>	3	9	3	3	2	1	0	0	0	0	0	0	9	100	
<i>P. aeruginosa A</i>	4	12	3	2	1	1	0	0	0	0	0	0	7	58.33	
<i>P. aeruginosa G</i>	3	9	3	2	2	1	0	0	0	0	0	0	8	88.89	
<i>P. aeruginosa I</i>	2	6	0	3	2	1	0	0	0	0	0	0	6	100	
<i>S. Kentucky</i>	2	6	0	2	1	0	0	0	0	0	0	0	3	50	
<i>S. Paratyphi A</i>	2	6	2	3	1	0	0	0	0	0	0	0	6	100	
<i>S. Typhimurium</i>	3	9	3	2	1	1	0	0	0	0	0	0	7	77.78	
<i>S. Typhimurium var. Copenhagen</i>	2	6	2	2	1	1	0	0	0	0	0	0	6	100	
<i>S. Virginia</i>	2	6	2	1	1	0	0	0	0	0	0	0	4	66.67	
<i>S. aureus</i>	10	30	5	3	3	2	1	1	0	0	0	0	15	50	
<i>Y. enterocolitica</i>	3	9	0	1	1	0	1	0	0	0	0	0	3	33.33	

*each isolate was inoculated in 3 mice.

**The percentage was calculated according to the number of examined mice of each isolates.

Table (10) Pathogenicity of the bacterial pathogens of public health importance isolated from pigeons in chicks.

Pathogens & No. of examined strains	No. of examined chicks	No. of infected chicks	Morbidity rate	No. of died chicks	Mortality rate	Case fatality rate *
<i>C. jejuni 10</i>	50	6	12	2	4	33.3340
<i>Citrobacter freundii 3</i>	15	5	33.3	2	13.33	40
<i>D. pneumoniae 7</i>	35	35	100	11	31.43	31.42
<i>E. coli O8 4</i>	20	16	80	9	45	56.25
<i>E. coli O78 5</i>	25	25	100	19	76	76
<i>E. coli O86 4</i>	20	11	55	4	20	36.36
<i>E. coli O111 3</i>	15	7	46.67	3	20	42.86
<i>E. coli O157 3</i>	15	5	33.33	1	6.67	20
<i>E. coli O166 4</i>	20	13	65	6	30	46.15
<i>K. oxytoca 3</i>	15	5	33.33	0	0	0
<i>K. pneumoniae 5</i>	25	19	76	10	40	52.63
<i>Mannheimia haemolytica 3</i>	15	15	100	9	60	60
<i>P. aeruginosa A 4</i>	20	20	100	16	80	80
<i>P. aeruginosa G 3</i>	15	15	100	13	86.67	86.67
<i>P. aeruginosa I 2</i>	10	10	100	10	100	100
<i>S. Kentucky 2</i>	10	8	80	0	0	0
<i>S. Paratyphi A 2</i>	10	10	100	7	70	70
<i>S. Typhimurium 3</i>	15	15	33.33	10	66.67	66.67
<i>S. Typhimurium var. Copenhagen 2</i>	10	10	100	9	90	90
<i>S. Virginia 2</i>	10	10	100	2	20	20
<i>S. aureus 10</i>	50	9	18	2	4	22.22
<i>Y. enterocolitica 3</i>	15	5	33.33	1	6.67	20

*Case fatality rate = No. of died chicks / No. of infected chicks.

Table (11): Incidence of enterotoxigenic bacterial pathogens of public health importance isolated from pigeons.

Pathogens	<i>E. coli</i> O8	<i>E. coli</i> O78	<i>E. coli</i> O86	<i>E. coli</i> O111	<i>E. coli</i> O157	<i>E. coli</i> O166	<i>P. aeruginosa</i> A	<i>P. aeruginosa</i> G	<i>P. aeruginosa</i> I	<i>S. Kentucky</i>	<i>S. Paratyphi</i> A	<i>S. Typhimurium</i>	<i>S. Typhimurium</i> var. <i>Copenhagen</i>	<i>S. Virginia</i>	<i>Y. enterocolitica</i>
No. of examined strains	12	15	13	9	7	13	11	7	4	3	3	12	7	2	9
Enterotoxigenic Positive	No.	9	10	8	4	4	7	5	3	2	3	10	7	1	6
	%	75	66.67	61.54	44.44	57.14	63.64	71.43	75	66.67	100	83.33	100	50	66.67

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دراسات بكتريولوجية للميكروبات للمسببات المرضية في الحمام المصري

لقد وجد أن معدل إنتشار العترات البكتيرية في الحمام والتي تشكل أهمية للصحة العامة في الإنسان هي ٢٨,١٦% ولقد وجد أن معدل تواجد العترات البكتيرية في الحمام يختلف باختلاف حاله الصحية للحمام المفحوص كما يختلف باختلاف العمر وكانت أعلى نسبة (٣٣,٣٣%) وجدت في صغار الحمام النافق حديثاً و الحمام البالغ النافق (٢٨,٥٧%) و صغار الحمام المريض (٣١,٠٣%) و الحمام البالغ المريض (٢٦,٦٧%) وأخيراً الحمام المذبوح (٢٥,٥٦%). وهناك العديد من العترات البكتيرية تم عزلها من مسحات من الأنف والشرج من الحمام المريض شاملة العترات التالية: (*C. jejuni*, *Citrobacter freundii*, *D. pneumoniae*, *E. coli*, *K. oxytoca*, *K. pneumoniae*, *Mannheimia haemolytica*, *P. aeruginosa*, *Salmonella spp*, *S. aureus* and *Y. enterocolitica*). ولقد لوحظ إختلاف في نسب و أنواع الميكروبات المعزولة من الحمام البالغ و الحمام الصغير كما وجدت إختلافات للمسحات المختلفه حيث لم يتم عزل (*K. oxytoca*, *Mannheimia haemolytica* and *Y. enterocolitica*) من الحمام البالغ ومن الواضح أن النفوق نشأ عن عدوي بأكثر من نوع من البكتريا، وبفحص الأحشاء الداخليه للحمام المذبوح ووجد أن معدل تواجد العترات البكتيرية في الحمام يختلف باختلاف عينه ووجد بالتصنيف السيرولوجي للعترات المعزوله أنها تشمل ٥ أنواع من السالمونيليا، ٣ أنواع من السيدوموناص إيروجينوزا، ٦ أنواع من الإيشيريشيا كولي ووجد أن كل المعزولات لها حساسيه لمركبات الإنترولوكساسين ثم الجنتاميسين ثم السيبروفلوكساسين ووجد أن مركبات الإستربتوميسين، الإريثروميسين، سلفات الكولستين هي المركبات التي لها حساسيه ضعيفه، كما وجد أن السيدوموناص إيروجينوزا مقاومه لمعظم المضادات الحيويه، ومن جهة أخرى وجد أن الإيشيريشيا كولي، السيدوموناص إيروجينوزا، السالمونيليا، واليرسينيا إنتيروكوليتيكا والمعزولة من الحمام وجد لها تأثير سمي معوي بنسبه ١٠٠% من عترات (*S. paratyphi A* and *S typhimurium* var. *copenhagen*) و بنسبه ٨٣,٣٣% من عترة (*S. typhimurium*)، ٧٥% لكل من (*E.coli O8* and *Ps. aeruginosa*).

