

Occurrence of Paratyphoid Infection Among Japanese Quails (*Coturnix coturnix japonica*) in Saudi Arabia

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Abstract. Integrated Quails Farm in the central region of Saudi Arabia with capacity of (50,000) birds a week was infected with Salmonella. Heavy mortality was up to 70% seen in Japanese quails, typical clinical signs of Salmonellosis were seen on the birds such as somnolence, weakness, nervous symptoms of torticollis, slight paralysis on some birds. Random samples collected from 14 houses including two breeding flocks and one rearing flock with different ages, were sent to the Poultry Disease Laboratory and subjected to bacteriological examination. Post-mortem examination showed characteristics lesions of necrotic foci in the liver, congestion of the lung, slight enlargement of spleen, acute enteritis. Consequently, salmonella was detected in 50% of the examined birds. Two distinct Salmonella Serogroups B (14 isolates) representing 63.64% and C1 (8 isolates) representing 36.36%, were isolated from Heart, Liver, Intestines, and Cloacal swab of the infected birds. Cox technique (rinse method) was carried out to isolate Salmonella from the processed quail carcasses of the same farm. Consequently, salmonella group B and C1 were isolated from the dressed quails on several occasions. Serotyping of the isolates revealed the following serotypes: *Salmonella*. Typhimurium, *Salmonella*. Virchow, *Salmonella*. Meleagridis, S. of group B 4, 12: I : - and S. of group C1 6,7: 1 : 1. Susceptibility test was carried out on the isolates against twelve antimicrobial drugs being used in the field of poultry.

Key Words: Paratyphoid infection ; Japanese quails; Saudi Arabia.

Introduction

Japanese quails (*Coturnix Coturnix japonica*) are members of the pheasant family (phasianidae). Japan is the native place of this species of quail which was domesticated as long ago as the 12th century, at the beginning of this century these birds have been bred in large numbers for dual-purpose of meat and eggs production. Traditionally, in the past quails production in the Kingdom of Saudi Arabia was mainly for sport falconry, but today quail meat and eggs have become highly required by the consumers due to their delicacy and low level of cholesterol. In addition, quail meat is tender and fortified with nutrients. Thus, the growing demand for meat and eggs of quails

has increased drastically. Nevertheless, many quail production farms were established around the country, some farms are fully integrated producing popular retailing packs contain 6 birds each, weighing approximately (600-800) grams. On the other hand, quail farms start gaining popularity in Saudi Arabia, because their, less feed requirements and space.

Salmonella are known to occur in poultry and poultry products and they are predominantly, such microorganism has been incriminated in food poisoning outbreaks around the world. Salmonella infection in quail is very important from the public health point of view as they can cause food poisoning in human beings. Reports of Salmonella in quails are scanty in literatures, although this particular infection has been documented on

Table 1. Source and serogroups of isolated Salmonella from Quail.

House No.	Age (day)	Isolated Group	Source of Isolates		
			Cloacal swab	Liver	Intestine
2	26	CI	+	+	+
3	40	B	+	-	-
		CI	-	+	+
4	12	B	+	+	-
		CI	-	-	+
5	5	B	-	+	-
		CI	+	-	-
7	33	B	+	-	+
8	9	CI	-	-	+
9	26	-ve	-	-	-
10	5	-ve	-	-	-
11	12	B	-	+	+
12	33	B	+	+	+
13	19	B	-	+	-
14†	DA**	-ve	-	-	-
15*	DA	-ve	-	-	-
16*	DA	B	+	-	-

-ve = negative *Breeders †Rearing **DA = Different ages

several occasions (Bryan 1980, Mohapatra 1992, Naveen and Arun 1992, and Sharma *et al.* 1995). To our knowledge this is the first report to be documented about Salmonella outbreak among quails in Saudi Arabia.

Materials and Methods

Samples collection and processing

Sick and apparently healthy quails, environmental samples (feces and Cloacal swab), and processed quail carcasses were collected from 14 houses including two breeding flocks and one rearing flock with different ages. The samples were submitted to the Poultry Disease Laboratory for bacteriological examination. At autopsy, pools from (Liver, Heart and Intestines) of 5 birds of

each house were examined individually.

The organs indicated above were chopped into small pieces with sterile scissors individually, then inoculated into selenite broth (SB) (Difco) in volumetric ratio 1: 10. The culture was incubated aerobically at 37°C for 18-24 hr. for Salmonella isolation, a loopful of the culture was streaked onto Brilliant Green Agar plates (BGA) (Difco), the plates were then incubated aerobically at 37°C for overnight. Suspected Salmonella colonies were transferred into Triple Sugar Iron Agar (TSIA), and lysine Iron Agar (LIA) (Difco), then tubes were incubated at 37°C for overnight.

Feces, Cloacal swabs

Approximately 25 gms of the fecal materials were added into 225 ml of (SB) in

Table 2. Number and percentage of isolated salmonella Serogroups.

Nature of Samples	Isolated Salmonella Serogroup			
	Group B		Group C1	
	No. Positive	% Positive	No. Positive	% Positive
Heart	1	4.54	0	0.00
Liver	5	22.73	2	9.09
Intestine	3	13.64	4	18.18
Cloacal Swab	5	22.73	2	9.09
Total	14	63.64	8	36.36

volumetric ratio 1: 10. A loopful of the culture was streaked onto (BGA), then the suspected salmonella colonies were transferred into (TSIA) and (LIA). Cloacal swabs collected from 5 birds of each house were placed in (SB) and incubated at 37°C for overnight. A loopful of the culture was streaked onto (BGA), and the suspected Salmonella colonies were transferred into (TSIA) and (TSIA) and (LIA).

Dressed quail carcasses

A modified method of Cox technique (rinse method) (Cox *et al.* 1983) was used to isolate Salmonella. Six quail carcasses (one retail pack weighing approximately 600-800 grams) were placed in a sterile heavy duty polyethylene bag, 100 ml of sterile distilled water was added then the bag was shaken vigorously for 1 minute. After shaking, the carcasses were drained into the bag for 15-30 seconds in a milk-dilution bottle. Ten milliliters of Selenite cystine broth (Difco) double strength (10X) was added to the rinse fluid and incubated at 37°C for 18-24 hr. Three loopfuls of the incubated culture were streaked onto (BGA) plates, and the plates were incubated aerobically at 37°C for 18-24 hr. Suspected salmonella colonies were transferred into (TSIA) and (LIA) for more identification.

Biochemical and Serological tests

Biochemical test was conducted according to the standard method of Ewing (1986). For the

cultures typical reaction of salmonella on TSIA and LIA.

Serological test was accomplished on pure culture of the isolates confirmed biochemically as Salmonella using slide agglutination technique, salmonella polyvalent "O" A-I and Vi and polyvalent "H" A-Z and specific somatic antisera (Difco, Detroit, Michigan, USA) were used. Salmonella cultures in pure form have been forwarded to LOHMANN TIERZUCHT GMBH Veterinary Laboratory, Cuxhaven, Germany, for further confirmation and serotypes identification.

Susceptibility test

Bauer *et al.* diffusion method (Bauer *et al.* 1980), was applied to examine the sensitivity of the salmonella isolates against twelve different antimicrobial drugs including. Ampicillin (AMP), Colistin (COL), Ciprofloxacin (CIP), Gentamycin (GEN), Kanamycin (KAN), Nalidixinsaur (NAL), Neomycin (NEO), Spectinomycin (SPE), Streptomycin (STR), Sulphamethoxazol+ Trimethoprim (SXT), Tetracycline (TET), Trimethoprim (TMP), where they predominantly used in the field of poultry.

Results

As shown in table (1), the source and the salmonella Serogroups which were isolated

Table 3. Salmonella serogroups isolated from processed quails.

Collection Number	Examined Number /	
	Bird	Isolated Salmonella Serogroups
1	12	Negative
2	36	B
		B
3	18	C1

Table 4. Representative Salmonella serovars isolated from quails.

Source	Serogroup	Serovar /	Seroformula
Processed quails	B	S. Typhimurium	1,4,12:i:1,2
Cloacal swab	B	S. Typhimurium	1,4,12:i:1,2
Cloacal swab	B	S. of group B	4,12:i:-
Cloacal swab	C1	S. Virchow	6,7:r:1,2
Intestines	C1	S. Virchow	6,7:r:1,2
Feces	C1	S. Meleagridis	3,10:eh:1w
Liver	C1	S. of group C1	6,7:-:-

from quail samples that were collected from 14 houses including two breeding flocks and one rearing flock with different ages. Salmonella Serogroups B or C1 were isolated from 10 houses (71.42%). Simultaneously, group B and C1 were isolated from houses 3,4, and 5. Salmonella group B was isolated from breeding flock of house No. 16, while the breeding flock of house No. 15 was negative for Salmonella. The isolates were recovered from Cloacal swab, liver, and intestines of the quails, and it could be isolated from one / and or two and three sources together.

Table (2) shows isolation percentages of group B and C1 of quail samples subjected to bacteriological examination. Serogroup B (5 isolates) (22.73%) were isolated from liver and Cloacal swab, while from intestines (3 isolates) (13.64%). On the other hand, Salmonella serogroup C1 was isolated twice from liver and Cloacal swab (9.09%), from intestines 4 times (18.18%). At the mean time, group B one isolate

(4.54%) was isolated from heart but not for C1.

As shown in table (3), Salmonella serogroups B and C1 were recovered from only two collections out of the three collections, which have been made for the processed quail carcasses during the outbreak.

Typing of the representative salmonella Serogroups revealed detection of S. Typhimurium from both Cloacal swab and processed quail carcasses of the same farm are shown in table (4). While, S. Virchow was isolated from Cloacal swab and intestines, S. Meleagridis was isolated from feces of the infected birds. S. of group B 4, 12: i: - monophasic was recovered from Cloacal swab and Non-motile S. of group C1 6,7: - - - isolated from liver of the birds.

Table (5) showing twelve different antimicrobial drugs commonly used in the therapeutic treatment of poultry. The results of the antibiotic susceptibility test of the salmonella isolates demonstrate that all tested

Table 5. Susceptibility of the isolated Salmonella from Quail against 12 antibiotics.

		Susceptibility of Salmonella to Antibiotics											
Source	Isolated Salmonella	AMP	COL	CIP	GEN	KAN	NAL	NEO	SPE	STR	SXT	TET	TMP
Processed quail	S. Typhimurium	S	R	R	R	S	R	S	S	S	S	S	S
Cloacal swab	S. Typhimurium	S	R	R	R	S	R	S	S	S	S	S	S
Cloacal swab	S. group B 4,12:1:-	S	R	R	R	S	R	S	S	S	S	S	S
Cloacal swab	S. Virchow	S	R	R	R	S	R	S	S	S	R	S	R
Intestines	S. Virchow	S	R	R	R	S	R	S	S	S	R	S	R
Feces	S. Meleagridis	R	R	R	R	S	R	S	S	S	R	S	R
Liver	S. group C1 6,7:-:-	S	R	R	R	S	R	S	S	S	R	S	R

(AMP) Ampicillin, (COL) Colistin, (CIP) Ciprofloxacin, (GEN) Gentamycin, (KAN) Kanamycin, (NAL) Nalidixinsure, (NEO) Neomycin, (SPE) Spectinomycin,

(STR) Streptomycin, (SXT) Sulphamethoxazol+Trimethoprim, (TET) Tetracycline, (TMP) Trimethoprim,

S = Sensitive

R = Resistant

strains were susceptible to five antibiotics namely: Kanamycin, Neomycin, Spectinomycin, Streptomycin, and Tetracycline. On the other hand, the isolates were completely resistant to four antibiotics namely: Colistin, Ciprofloxacin, Gentamycin, and Nalidixinsaurate. In addition, differences in susceptibility have been shown among salmonella isolates to three antibiotics namely: Ampicillin, Sulphamethoxazol + Trimethoprim, and Trimethoprim.

Discussion

Reports about Salmonellosis in quails seem to be scanty in literature. Recently, due to the intensive management and establishment of quail farms, reports of Salmonellosis in quail are coming regularly. Plenty of salmonella species have been isolated worldwide from quails infected with salmonella.

It is an acute septicaemic disease mostly encountered in young Chicks in first 1-3 weeks of age, the mortality sometimes may be as high as 100% (Mohapatra, 1992). Accordingly, the records of the infected farm in Saudi Arabia showed mortality up to 70% among the birds during the outbreak. Similarly, Janakirama Sarma *et al.* (1988) found the mortality among baby quails around 71% due to infection with *Salmonella Gallinarum* (Janakirama Sarma *et al.* 1988). Salmonella Serogroups B and C1 were isolated from the infected birds, serogroups B was more predominantly (63.64%) than serogroup C1 (36.36%). Isolation of group B from liver and Cloacal swab was equally (5 isolates) (22.73%), while (3 isolates) (13.64%) were from the intestines. On the other hand, group C1 was isolated (2 isolates) (9.09%) from each of liver and Cloacal swab, while (4 isolates) (18.18%) were from the intestines. Salmonella group B and C1 were isolated simultaneously from houses 3,4 and 5. Salmonella group B was isolated from the

breeding flock of house No. 16 but not from breeding flock of house No. 15. On the other hand, Salmonella serogroup B and C1 were isolated from processed quail carcasses of the same farm, and this phenomenon reflects the contamination which occurred during the evisceration process of the birds, at the mean time, give an idea about the sanitation status in the plant. Nevertheless, the hygienic farm management is a must. Seven salmonella isolates were chosen out of the recovered salmonella Serogroups to represent different sample sources.

The representative Salmonella serovars which have been recovered were as follows: *S. Typhimurium* was isolated from both Cloacal swab and processed quails of the same farm, and this is a conclusive evidence that the cross contamination is taking place in the processing plant, while the monophasic *S.* of group B 4, 12: i: - was isolated from Cloacal swab. On the other hand, *S. Virchow* was isolated from Cloacal swab and intestines, *S. Meleagridis* isolated from faces but not from Cloacal swab or intestines of the infected birds. In addition, a non-motile salmonella of group C1 6,7: - : - was isolated from liver of the infected birds. In contrast, this particular salmonella serovar had been isolated from fish-meal of the poultry farm in Saudi Arabia (Barbour *et al.* 1984). However, the antigenic formula of the isolated non-motile salmonella group C1 6,7: - : -, dose not conform to any known antigenic formula described by Kauffmann-White schema (Ewing 1986).

Recently, the resistant phenomenon among Salmonella bacteria is a worldwide increase, and this attributed to the extensive usage of the antibiotics in the field of the veterinary and human medicine practice. In our study the salmonella isolates were completely resistant to four antibiotics (33.3%) used in this study. Meanwhile, the isolates showed different

susceptibility against three antibiotics.

In conclusion, the author recommend that initiation hygiene management program and implementation of quality assurance system on the farms for production of safe food is a must. Moreover, prudence and restrictions in the use of antibiotics is mandatory to prevent the emergence of resistant salmonella strains. Also national antimicrobial susceptibility monitoring program should be implemented to provide data on trends of antimicrobial susceptibility in human and animals population in Saudi Arabia.

Acknowledgement

The author is indebted to Dr. M. Voss, from, LOHMANN TIERZUCHT GMBH Veterinary Laboratory. Cuxhaven, Germany, for assisting and typing the salmonella isolates. Our appreciation is extend to Mr. Jayaraman Moorthi the laboratory technician for helping in media preparation and samples analysis.

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إصابة طيور السمان اليابانية في المملكة العربية السعودية بمرض نظير التيفويد

حبيب مقبول النخلي

مختبر أمراض الدواجن - المركز الوطني لأبحاث الزراعة
ص.ب ١٧٢٨٥ ، الرياض ١١٤٨٤ ، المملكة العربية السعودية

ملخص البحث : مزرعة سمان متكاملة بالمنطقة الوسطى في المملكة العربية السعودية ، سعتها ٥٠٠٠٠ طائر في الأسبوع، أصيبت بالسالمونيلا. كانت نسبة النفوق ٧٠% بين طيور السمان حيث ظهرت على الطيور أعراض الإصابة بالسالمونيلا مثل الخمول، الضعف، أعراض عصبية، إصابة خفيفة بالشلل بين بعض الطيور.

جمعت عينات عشوائية من ١٤ حظيرة. بما فيها حضيرتين للامات وحظيرة تربية ذات أعمار مختلفة، أرسلت العينات لمختبر أمراض الدواجن للفحص البكتيريولوجي. الصفات التشريحية للطيور أظهرت وجود تتركزات على الكبد، احتقان الرئة، تضخم بسيط للطحال، التهاب حاد للأمعاء. أظهرت نتائج الفحص البكتيريولوجي تواجد جرثومة السالمونيلا في ٥٠% من الطيور المفحوصة. وقد تم عزل مجموعتين محددتين من السالمونيلا: مجموعة B (١٤ عزلة) (٦٤ و ٦٣%)، مجموعة C1 (٨ عزلات) (٣٦ و ٣٦%) وكانت العزلات من القلب، الكبد، الأمعاء والمسحات القطنية المأخوذة من فتحة المجمع للطيور.

وباختبار تحديد النوع المصلي لجراثيم السالمونيلا ظهرت الأنواع التالية:

S. Typhimurium, S. Virchow, S. Meleagridis, S. of group B 4, 12: 1:-, S. of group C1 6,7:-:-.

عند إجراء اختبار الحساسية للعزلات ضد ١٢ نوع من المضادات الحيوية والتي تستخدم في حقن الدواجن، خمسة أنواع منها فقط أظهرت فعاليتها وقد تضمنت الكاناميسين، نيومايسين، سبيكتينومييسين، ستريبتومايسين، والتيترا سيكلين.