Prevalence of *Salmonella enteritidis* in aquatic bird's eggs (*Anas platyrhncha*) from farmer's houses in Basrah marshes, Iraq.

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#### Summary

The present study was performed to detect the incidence of *Salmonella enteritidis* contamination of water bird's eggs from farmer's houses in Basrah city marshes. 90 samples were collected from eggs. Bacteriological examination and antimicrobial susceptibility test was done and the results showed that the rate of contamination of eggs shells and contents was (4.4%,2.2%) respectively.

## Introduction

Salmonella consider a major cause of food borne human disease in most parts of world (Soultose et al., 2003) and transmitted to humans through raw poultry and products (Kimura et al., 2004). Salmonella are fermentative facultatively anaerobic, oxidasenegative, gram-negative rods. They are generally motile, aergenic, nonlactose fermenting, urease negative, citrate utilizing and acetyl methyl carbinol negative. They are simple to cultivate and submitted themselves readily to biochemical testing (Barrow and Feltham, 1992). The genus salmonella consists of two species, Salmonella enterica and Salmonella bongori, within the species S. enterica there are six subspecies differentiated by biochemical variations, namely entrica, Salamae, arizonae, diarizonae, houtenae and indica (Old and Threlfall, 1998). Serovars within the subspecies other than subspecies, entrica are designated by their antigenic formula, for example, S. enteric. Arizonae 61:K:1,5,7. Salmonella cause localized infection of the gastrointestinal tract but can also multiply in the reticuloendothelial system resulting in systemic infection and death. Eggs are familiar, nutritious, versatile,

economical, quick and easy to prepare food, also they provide a unique well balanced source of nutrients for all ages.

Moreover, their high quality, law caloric value and case of digestibility make eggs valuble in many therapeutic diets for adults (Buley and Vadehra. 1989; Bufano, 2000). *Salmonella* is one of the primary causes of human foods poisoning

throughout the world (Fantasia and Filetici, 1994). World wide, salmonellosis is a serious medical and veterinary problem and raises great concern in food industry. Poultry is the most potential source of *Salmonella* food presenting in man. Moreover, contamination of the poultry meat with *Salmonella* in the poultry slaughterhouses is very important (Ashton, 1990). The dominant type of *Salmonella* food poisoning was *Salmonella typhimurium* but since 1982, *S. entritidis* Curried out by chickens and poultry products is the major source of human intestinal infections (Fantasia and Filetici, 1994).

Salmonella contamination in and /or on eggs has been shown to make a significant contribution to human diseases in several counties (Lake *et al.* 2004). However, contamination of the food after illness could not be discounted (ESR, 2006). Eggs and eggs products are nutritious foods and they form an important part of the human diet eggs and eggs products that are improperly handled can be a source of foodborne diseases , such as Salmonellosis.

Salmonellosis is a leading food borne disease worldwide. A wide range of foods has been implicated in such as disease. However, foods of animal origin, especially poultry and poultry products, including eggs, have been consistently implicated in sporadic cases and outbreakes of human Salmonellosis (FAO/WHO). Salmonella reside in the intestinal tract and are shed in the faeces of infected animals, and humans as well. Many foods, particularly those of animal origin and those subject to sewage

pollution, have been identified as vehicles for transmitting those pathogens (ICMSF, 1996).

Eggs contents may be contaminated with *Salmonella* by 2 routes, transovorion or horizontal transmission (FAO/WHO). The nutrients that make eggs a high- quality food for humans are also a good growth medium for bacteria, any foods particularly protein –rich

animal foods such as eggs can carry microorganisms that cause disease or spoil the food (Frazier and Westhoff, 1986).

Material and Methods

Collection of samples:

A total of ninety aquatic bird eggs were collected from farmer's houses in Garmmat Ali City. Each ten eggs constituted a group, each 10 eggs (one group) were placed in a sterile plastic bag and transferred to the laboratory with out delay to prepared and examined for salmonella species.

# Preparation of samples:

-Egg shells: Egg shells were tested by a surface rinse method as described by Moats (1979).

-Egg contents: Eggs were prepared for evacuation of their contents according to Speak(1976).

# Microbiological analyses:

Replicate portions of ten fold dilutions of samples in sterile peptone water were made for all samples collected, the preparations were hemogenized and 0.1 ml each of appropriate dilutions was plated using the pour plate method (Harrigan and MeCance, 1976). enumeration of the total viable counts was conical out using plate count Agar.

Salmonella- Shigella (SS) agar was used for the isolation and enumeration of Salmonella and Shigella after enrichment of samples at tetrathionate broth. Plates were incubated at 37C° for 48 h. Surface swabs of shells of the eggs were taken and cultured on the same media.

All colonies from incubated plates were purified by repented subculturing before being examined microscopically for gram reaction (Claus, 1992). Cell morphology (using 24h old cultures ), motility, all isolation were purification and identification according to gram stain, culture characteristics and biochemical reactions (Harrigan and MaCance, 1976).

Biochemical analysis included catalase and oxiodase reactions ; sugar fermentation, urea hydrolysis (Christensen, 1946; (Harrigan and MeCance, 1976).

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Antibiotic susceptibility test ; Determination of the susceptibility of isolated strains to antibiotic discs (oxoid) was adapted using the disc diffusion method according to Finegold and Martin (1982).

#### Results

# Table 1: Incidence of *Salomenlla enteritidis* from the examined Egg shells and contents.

samples Total sam		nples	Salmoi		monela	ela enteritidis	
				No		%	
Egg shells 90				4		4.4	
Egg contents 90		ł		2		2.2	
Table 2: Antimicrobial sensitivity test in the examined samples.							
Antibiotics		Conc.	μg	Percentage of effect on isolated S.			
			enteritidis strain		s strair	ns (n=6 isolates)	
				S	R		
Tetracycline		30		2(33)	4(66	6.4)	
Neomycin		30		2(33)	4(6	6.4)	
Ampicillin		20		1(16.4)	5(8	5(83)	
Gentamicin		10		3(50)	3(5	3(50)	
Chloramphenicol		30		4(66.4)	2(3	3)	
Enterofluxacin		5		4(66.4)	2(3	2(33)	
Ciprofloxacin		5		5(83)	1(1	6.4)	
Trimethoprim-		25		2(33)	4(6	6.4	
Sulfomethoxazol				. ,	•		
S- sonsitivo P- resistanco							

S= sensitive R= resistance

#### Table 3: biochemical tests for diagnosis of Salmonella enteritidis

The test	Result
Motility	+
Gram's stain	-
Cellular morphology	Rods
Catalase activity	+
Oxidase activity	-
Urea hydrolysis	-
Citrate utilization	+
Acetyl methyl carbinol	-
Sugar fermentation	
Glucose	+
Lactose	-
Maltose	-
Xylose	-

# Discussion

The obtained results in table(1) showed that the *S. enteritidis* was isolated from eggs of water birds with an incidence of (4.4%, 2.2%) this results was approach with that obtained by Molback and Neimam, (2002) who recorded results (3.4%) from raw eggs These results can be explained as collected eggs had transovarin transmission from carrier birds which had no signs of a disease but shedding of *S. entenitidis* in eggs been occurred.

It has been documented that storage of eggs at the cell outlets, depending storage conditions, particularly the temperature and duration, may effect the microbial load of both egg shells and contents but not the prevelance of bacteria (Jones etal, 2004).

Freshly laid eggs are generally semi- sterile, however they may constitute, if contaminated, a public health hazard, and cause economic losses through their spoilage (Perales and Audicana, 1989, ACMSF, 1993).

This variation in recovery rate may be attributed to differences in environmental temperature, variation of bird husbandry practices or even variation in methods of isolation (WHO, 1988,ICMSF,1996)

Regarding the antimicrobial sensitivity test table(2) using .disc diffusion method, to the isolated *S. enteritidis*. strains from the previous source, showed that Ciprofloxacin was the drug of choice. Followed by enterofloxacin, gentamicin, neomycin, tetracyclin, chloramphenicol. trimthprim-Sulfamethaxazol and the lowest sensitivity was to ampicillin.

The results assured by Duarte *et al*, (2009), Murugkgr *et al*, (2005), Yagoub *et al*, (2006), Smith *et al*, (2009) who found that all isolated *S. entenitidis* was highly sensitive to ciprofloxacin followed by enterofloxacin then chloramphenicol and low sensitive to gentamicin, trimthprim-Sulfamethaxazol but highly resistant to ampicillin.

However, the nutrients that make eggs a high- quality food for human are also a good growth medium for bacteria (Frazier and Westhoff, 1986). Eggs were considered a vehicle for transmission of certain pathogens to man if such eggs are consumed raw or semi- raw. Bacteria on egg shell have been emplicated as source of. Bacterial contamination of broken out eggs (Solowey *et al*, ,1986; Kraft *et al*, 1967). Motile bacteria on shells may easily penetrate the shells to interior (Board, 1968). The rate of penetration is influenced by humidity and storage temperature. At which the eggs were produced and stored (Beard and Fuller, 1994; Cox *et al*, 2000).

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## مجلة جامعة ذي قار للبحوث الزراعية ، المجلد ١ ، العدد (٢) ٢٠١٢

تواجد جرثومة Salmonella enteritidis في بيض الطيور المائية (الخضيري) من بيوت الفلاحين في أهوار البصرة غازي مالح جابر \*، عيسى طه محمد، امجد نعمة الابريسم، مجيد شناوه \* قسم الاحياء البحرية، مركز علوم البحار، جامعة البصرة.

الخلاصة

أنجزت هذه الدراسة لتحديد تواجد التلوث بجرثومة السالمونيلا في بيض الطيور المائية المنتج من بيوت الفلاحين في أهوار مدينة البصرة أذ تم جمع ٩٠ عينة من البيض وأجري عليها الفحص الجرثومي وكذلك أجريت اختبارات الحساسية الدوائية على العزلات الناتجة وقد أشارت النتائج بأن معدل التلوث كان بنسبة ٤.٤% في قشرة البيض و٢.٢% في محتويات البيض.