

Synovitis associated with Salmonellosis infections of broiler chickens in Karbala Province

التهاب المفاصل المرتبط مع إصابات السالمونيلا لدرج اللحم في مدينة كربلاء

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Abstract

The study aimed in part to identify bacterium *Salmonella spp.* isolate of culturing and antimicrobial susceptibility test in diagnosis of this bacteria that causes arthritis isolated from chicken farm in Kerbala governorate, Iraq. A total of 40 joint swabs were collected from broiler suffering by arthritis isolated from chicken farm in Kerbala governorate, through a period of three months (from December 2014 to February 2015), in an attempt to detect the bacterium *Salmonella spp.* which play a role for Arthritis. The research was concluded that the isolated *salmonella spp.* belongs to the Enterobacteriaceae play important in the synovitis of broiler chicken, Ciprofloxacin and Chloramphenicol had the highest overall in all *salmonella spp.* vitro efficacy, but resistant 100% coverage of Amoxicillin against *salmonella spp.* that isolate and tested.

الخلاصة

هدفت الدراسة لتحديد عزلات بكتريا السالمونيلا عن طريق الزرع واختبارات الحساسية وتشخيص هذه البكتريا كالتهاب مفصلي. تم جمع ما مجموعه 40 مسحة مفصلية من فروج اللحم والذي يعاني من التهاب المفاصل وجمعت العينات من عدد من حقول الدواجن في محافظة كربلاء المقدسة- العراق، خلال فترة ثلاثة أشهر (من ديسمبر 2014 إلى فبراير 2015)، في محاولة للكشف عن بكتيريا السالمونيلا والتي تلعب دور مهم في التهاب المفاصل. استنتج البحث بأن أنواع السالمونيلا المعزولة والتي تنتمي إلى مجموعة المعويات لعبت دور مهم في التهاب مفاصل فروج اللحم، وأعطيت المضادات الحيوية Ciprofloxacin و Chloromphenicol أعلى نتيجة كفحص حساسية ضد جميع أنواع السالمونيلا المعزولة، ولكن لوحظ جميع أنواع عزلات السالمونيلا التي تم اختبارها لها مقاومة 100% للمضاد الحيوي Amoxicillin.

Introduction:

Salmonellosis was the most common infectious diseases in the world in both animals and humans (1), *Salmonella spp.* are among the leading causes of community acquired food borne bacterial gastroenteritis worldwide (2).

It is ubiquitous geographically and zoologically. Some serotypes are relatively host-specific (*S. dublin* - cattle; *S. typhisuis* - swine; *S. pullorum* - fowl) while others, notably *S. typhimurium*, *S. anatum*, and *S. newport*, affect a wide host range among which feral birds and rodents play important roles in interspecific dissemination of infection. Long periods of asymptomatic and convalescent shedding ensure widespread, unchecked distribution of the organisms.(3).

Salmonella enterica is a significant food-borne pathogen of humans transmitted via the consumption of meat, animal products, and food products (fruits and vegetables) contaminated with animal waste (4). Humans appear to be susceptible for all *Salmonella* serotype, the most important source for which are animals and their by-products (poultry and poultry products and eggs were a major source of *Salmonella* in humans). *Salmonella enteritidis* was especially adapted for egg transmission. Whether a person develops disease following ingestion of salmonellae from the environment depends upon the dose of organisms, the serotype of *Salmonella*, and the colonization resistance of the infected individual. *Salmonella typhimurium* is most common, usually producing gastroenteritis (5).

The aim from this study to diagnosis bacteria infected joints of broiler and cause synovitis ,lameness and recumbent of bird with sensitive test for antibiotics for this bacteria to decrease morbidity and mortality rate.

Materials and Methods:

The specimens was represented by ten farms in Kerbala city (Al-Hure\ Al-Kamalia aria) with morbidity rat arrived to 6% and mortality rat arrived to 5%, consisted of broiler chickens. At one of the series of broiler chickens, avian infectious synovitis was suspected. For the confirmation of diagnosis, this study had performed anatomical and bacteriological examinations. The anatomical examination was made in the farm and the laboratory exam was made in laboratory Department of Microbiology, Kerbala University- Veterinary college, with in the Clinical examination was made daily, after 20-23 days old, at this age were seen the first symptoms of disease as reddish enlargement of joint, lameness, un able to stand.

Samples Collection:

A total of 40 pus forming specimens were collected from broiler suffering from arthritis who were presented to Kerbala farms, 200 µl of pus specimen was placed in 5 ml of selenite broth, labeled and transported to the laboratory in portable container, then incubated for 18-24 hrs at 37 C°. This study was conducted during the period that extended from December 2014 to February 2015.

Isolation of *Salmonella spp.*:

A lapful of selenite broth culture was streaked on surface of S.S, XLD plates and then incubated at 37 C° for 24 hrs . The biochemical characters of non – lactose fermenting bacteria was determined by using TSI agar and urease test and other biochemical tests. Colonies that showed biochemical characteristics similar to that of *Salmonella spp.*(6).

Antimicrobials Susceptibility Testing:

The antimicrobial susceptibility testing for bacterium colonies was done by the agar discs diffusion method by using five antimicrobial disc like (Gentamycin, Amoxicillin, Chloromphenicol, Neomycin and Ciprofloxacin) as that described by (7).

Statistical analysis:

Chi-square recommended by (8) was used for statistical analysis to show if there is any significant differences between results

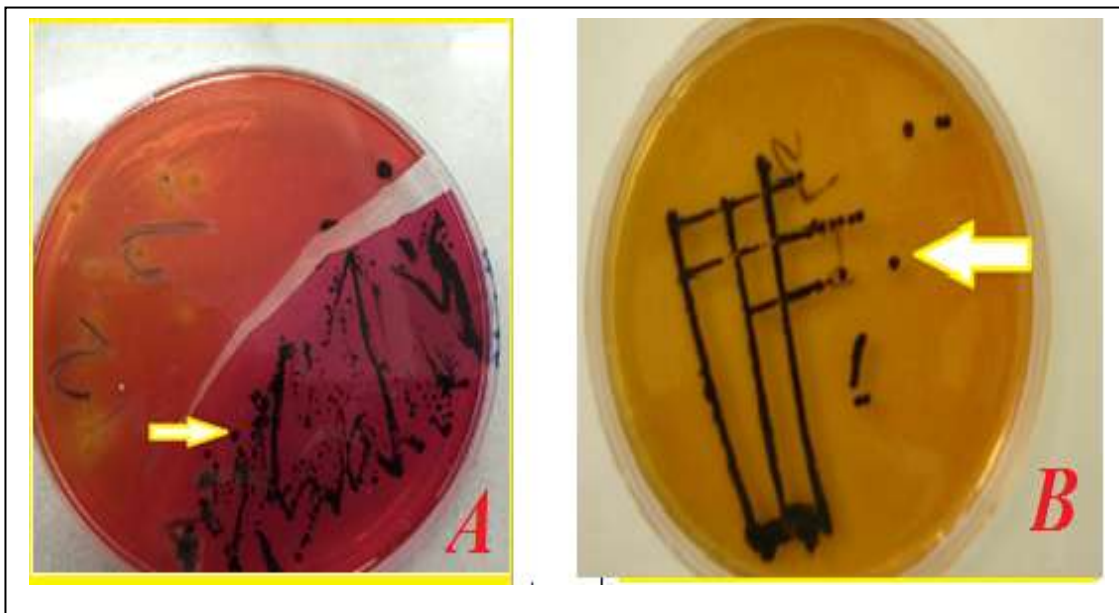
Results and Discussions:

Recent systematic studies of cause s of lameness in birds was reared in Iraq, therefore this study was aimed to detected some bacterial like *Salmonella spp.* which present in the ankle joint of the broiler chickens, in this study commercial broilers showed that long bone deformities were the main cause of lameness like arthritis and osteomyelitis (9)(figure 1) in the 21 days of age, the gross lesions was represented as pus forming, white to red crystals' areas of gaseous exudates around joint when it's opening by surgical seizer.



**(Figure 1) A: broiler, 22 days old. Note enlargement of ankle joint in the proximal end of femur
B: proximal femoral degenerative changes related to bacterial infection.**

The results obtained by culturing methods shown in table 1 indicated that out of 40 samples, 18 (45%) were positive for *Salmonella spp*, using the conventional culture methods of stool specimens on enrichment and selective media. According to the manufacture company (Difco) the colonies of *Salmonella spp*. on XLD agar were small, smooth, circular, convex and red in color with black center (Figure 2 A), while was pale in color with black center, circular, convex and smooth on S.S agar (Figure 2 B)



(Figure 2) A: the arrow showed colonies of *Salmonella spp*. On XLD agar as red colonies with black center, B: the arrow showed colonies of *Salmonella spp*. on S.S agar as pale colonies with black center.

The biochemical tests for all isolates showed positive results to H₂S production, glucose fermentation, citrate utilization, methyl red and motility tests while the results also confirm as negative reactions for indole production, urease, Voges-Proskauer,(10).

In naturally occurring enlargement of the proximal leg bones, birds may show signs of lameness or may simply be found dead. The onset of lameness may be associated with increased mortality rate in the flock due to weight loss it would be unlikely to obtain food or water and that lead to immune suppression (11).

By using the disc diffusion method, 18 isolates of *Salmonella spp.* were tested for their antimicrobial susceptibility toward 5 antibiotics, All tested isolates showed high susceptibility (100 %) toward ciprofloxacin and high resistance (100 %) against amoxicillin and half of isolates was sensitive to chloromphenicol. On the other hand, these isolates revealed varying percentages of susceptibility and resistance toward other antibiotics like gentamycin and neomycin.

Table (1): Antimicrobials susceptibility testing of *Salmonella spp.* toward certain antibiotics using discs diffusion method.

antibiotic	Sensitive		X ² value
	isolates	%	
Gentamicin	7	38	Cal.X ² =37.7 Tab. X ² =33.1 df= 4 Non-Significant (p>0.01)
Chloramphenicol	9	50	
Amoxicillin	0	100	
Ciprofloxacin	18	100	
Neomycin	11	61	

All bacterial isolates testing were found to be very effective against ciprofloxacin duo to The mode of action of all quinolones involves inhibition of bacterial DNA synthesis by blocking of the DNA gyrase but demonstrated some weakness against certain Gentamycin depending on protein inhibition drugs and some of strain resistant against it may be associate the resistance were under the control of transmissible plasmids(12).

On the other hand, Chloramphenicol was a potent inhibitor of protein synthesis in microorganisms, It blocks the attachment of amino acids to the nascent peptide chain on the 50S unit of ribosomes by interfering with the action of peptidyl transferase(13).

There are many different mechanisms by which *Salmonellosis* might exhibit resistance to Amoxicillin, the most important of them were Microorganisms produce enzymes that destroy the active drug by producing B-lactamase (14).

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