

Detection and characterization *Salmonella enterica* from broiler chicken(Rose308) in Thi-Qar province and its relationship to broiler immunity and body weight

¹Noor Hassan Ghargan , ²Ali A. Abdulkareem , ³Zaman Kareem Hanan

^{1,2}Department of Animal Production, Faculty of Agriculture and Marshes, University of Thi-Qar, Iraq

³ College of Dentistry, University of Thi-Qar, Nassiryhia, Iraq

¹E-mail: lmbitpc@gmail.com

²E-mail: ali-ah@utq.edu.iq

³E-mail: zaman.k.bio@utq.edu.iq

Abstract

This study was conducted to isolate *Salmonella* bacteria from broiler (ROSS 308). A total of 184 stool samples were collected from chickens, showed symptoms of infection with salmonella bacteria from two different fields in Thi-Qar Governorate in Al-Islah district and Al-Fadeliyah district. Thirty isolates (16.2%) were diagnosed with Salmonella infection by biochemical and API 20 E tests. The pathological infection was confirmed in chickens by analyzing the level of antibodies in the blood of chickens, there was an increase in the level of immunoglobulin IgG and IgM, confirmed the existence of the disease and the stage of infection between a new infection and the acquisition of immunity, as a result of the previous injury and injuries close to recovery. The birds that suffer from a lack of weight differed in the stage of infection, this indicates that infection at any stage has a significant impact on the weights of birds infected with salmonella bacteria.

Keywords. *Salmonella enterica* , broiler chicken(Rose308) , broiler immunity

I. INTRODUCTION

The animal production sector is facing challenges all over the world, these challenges differ in different producing countries, whether developed or developing., so there must be effective solutions to these challenges, contribute to the preservation of animal production systems [1].

Livestock is an important aspect of the agricultural field, poultry is one of the most important economic sectors in economic development in many Arab countries. Poultry is considered part of the livestock, broilers come at the forefront of those poultry [2].

Broiler meat is one of the most important basic sources of human food, as one of the natural resources in most countries, it can be relied upon as an important food source [3]



Poultry, including broiler chicken, is susceptible to salmonella infection, which is a concern for researchers and public health because of the associated losses, where salmonella has been studied, as an economically important zoonotic pathogen by the World Health Organization [4].

Salmonella, which is a Gram-negative bacillus, it is a facultative anaerobe and belongs to the family Enterobacteriaceae, can cause gastrointestinal infections [5].

As a result of the importance of the aforementioned broiler, significantly affected by Salmonella bacteria due to the formation of biofilms, the study aimed to detection of *Salmonella enterica* from broiler chicken (Rose308) in Thi-Qar province and its relationship to broiler immunity and body weight.

II. MATERIALS AND METHODS

The research work period started from 28/11/2022 to 20/4/2023, the study period included two phases, the field work phase and the laboratory work phase, where 184 samples of broiler feces ROSS 308 were collected from two different fields from Al-Islah field (A) and Al-Fadhliyah field (B) in Thi-Qar governorate. Experiments were conducted on these models at Thi Qar University in the Microbiology Laboratory of the College of Dentistry.

Field work

Fecal samples were collected from broiler ROSS 308, it included 184 unsexed samples with two different ages from two different fields in Thi-Qar Governorate. The chicks were numbered and weighed at the age of 3 weeks, then weighed at the age of 5 weeks before the blood draw, were reared in the field on a litter of sawdust with a height of 5 cm, the field was heated with regularly distributed gas heaters to provide the ideal temperature, then, it decreased by three degrees every week, to stabilize at a rate of 25 °C, at the end of the fifth week, a mercury thermometer was placed to monitor the temperature at a height of 1m. The use of natural ventilation in the first weeks and other pullers system. Humidity was controlled by a humidifier in the range of 60-70. The lighting continues for 24 hours. The birds were fed ad libitum at both fields from 1 to 35 days of age on starter and growth diet, the amount of diet was calculated according to nutritional needs, the weight of feed consumed was calculated at the end of each week. The chicks were also vaccinated with the triple vaccine (Newcastle, Gumboro, and Coccidia), the birds were given vitamins and antibiotics for two days. The 3-week-old Newcastle Lasota vaccine was reintroduced by drinking water after thirsting for an hour, then the chicks were weighed using the electronic balance for each bird separately in the third week of life, then the weight at the end of the fifth week. Weight averages were taken to find out the differences in weights for each bird during the 35-day experiment.

Laboratory work

Bacteria isolation

After collecting 184 stool samples from broiler chickens, they were preserved in Buffer peptone water, 1 ml of medium was taken into tubes of Tetrathionate broth medium. Tubes were incubated at 37°C for 24 hours, after the bacteria were grown on the medium, they were cultured on solid selective media (XLD). The dishes were incubated



for 24 hours at a temperature of 37°C, then the colonies were purified from the medium and cultured on Nutrient agar, for biochemical and API 20 E testing (6).

Elisa test

The ELISA test was used to detect the level of IgG and Igm antibodies to salmonella in the blood serum of the birds used in the experiment [7], after drawing blood from the pterygoid vein of 52 broiler, who show signs of disease, blood was placed in sterile heparin-free test tubes at an angle, then the tubes are placed in the centrifuge for ten minutes at a rotation speed of (3000) rpm, the indirect method was used for testing according to OIE [8], using an ELISA test kit from Genrui Biotech Inc.

III. RESULTS AND DISCUSSION

After collecting 184 stool samples from broiler chickens from the chicken fields in Dhi Qar Governorate, with an average of 84 samples from the field (A). Fifty-one samples free of salmonellosis at the age of 21 days and the same age were obtained. 21 infected samples were obtained and 12 deaths were found, samples collected from the field (B) where 100 samples of chicken feces were collected, 46 samples free from infection at the age of 21 days and 35 days were obtained from the experiment, in addition to 31 infected samples at the two ages, with 23 deaths. The cause of death may be attributed to the lack of weight resulting from anorexia due to infection with bacteria, which was also accompanied by severe diarrhea, which leads to dehydration, the body's lack of resistance due to weak immunity, the inability of antibodies to overcome the antigen, as bacteria are able to overcome antibiotics, that can control the severity of the disease.

The results showed that the total number of infected samples from the two fields was 87 samples, showed symptoms of infection with salmonella outwardly. The pathological infection was confirmed in chickens by analyzing the level of antibodies in the blood of chickens, where there was an increase in the level of immunoglobulin IgG and IgM, which confirmed the presence of the disease and the stage of infection, as 23 samples obtained in field (A) produced antibodies to resist the bacteria, by the results of the analysis, the stage of infection was known, where 9 samples appeared indicating the presence of the new infection in the body of the bird, 8 samples had previously been infected with immunity inside the body, 6 samples are hit in the middle, in field (B), 31 samples were obtained that produced antibodies in the chicken body, the stage of infection also varies, as 12 samples were suffering from a new infection, 11 samples were exposed to infection and the occurrence of immunity in the body, 8 samples in which the infection is in the middle or close to recovery, as for the result of culturing questionable stool samples on selective media, salmonella colonies of 45 samples appeared pink, with a black center in the middle of XLD agar (Figure 1).





Figure 1. Salmonella colonies in pink with a black center.

The samples were subjected to biochemical tests and the result was negative for the oxidase test and positive for the catalase test. The results of the examination on the API 20 E strip was to obtain 30 samples (16.3%) belonging to *Salmonella* (Fig. 2).

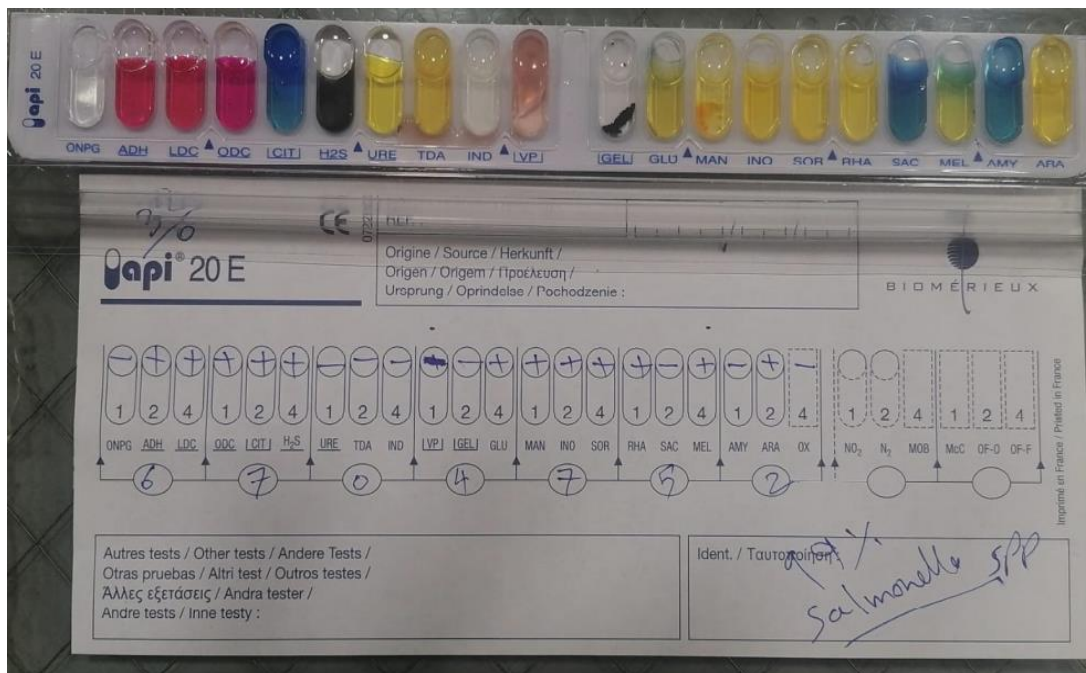


Figure 2. API 20 E strip test result.

After confirming the infection of broiler ROSS 308. Table (1) showed a relationship between weight loss at the age of 21, 1 day and 35 days due to infection with bacteria. The antibody analysis showed that infection with salmonella bacteria, whether the infection was old, that was, the bird acquired immunity, or the infection was new, the bird remains anorexic and suffers from weight loss.

Table 1. The relationship of low weights to the stage of infection.

Samples	21 days weight	35 days weight	Antibody resistance
S1	٨٠٠	١٦٠٠	infection in the middle
S2	٨٥٠	١٩٠٠	having an infection
S3	٨٦٠	١٦٠٠	having an infection
S4	٧٩٠	١٧٠٠	infection in the middle
S5	٨٠٠	١٦٠٠	injured
S6	٨٠٠	١٦٠٠	having an infection
S7	٧٥٠	١٦٠٠	injured
S8	٧٨٠	١٦٠٠	infection in the middle
S9	٨٥٠	١٦٠٠	infection in the middle
S10	٨٠٠	١٧٠٠	having an infection
S11	٨٥٠	١٩٠٠	injured
S12	٧٠٠	١٩٠٠	injured
S13	٧٥٠	١٩٠٠	injured
S14	٧٥٠	١٨٠٠	injured
S15	٧٥٠	١٨٠٠	having an infection
S16	٧٥٠	١٨٠٠	infection in the middle
S17	٨٠٠	١٨٠٠	injured
S18	٧٥٠	١٩٠٠	having an infection
S19	٨٠٠	١٨٠٠	having an infection
S20	٧٥٠	١٨٠٠	having an infection
S21	٧٥٠	١٨٠٠	having an infection
S22	٧٥٠	١٨٠٠	infection in the middle
S23	٧٥٠	١٨٠٠	having an infection
S24	٧٨٠	١٦٠٠	having an infection
S25	٧٥٠	١٨٠٠	having an infection
S26	٧٥٠	١٨٠٠	having an infection
S27	٧٥٠	١٨٠٠	infection in the middle
S28	٧٥٠	١٨٠٠	infection in the middle
S29	٧٥٠	١٨٠٠	infection in the middle
S30	٨٠٠	١٨٠٠	infection in the middle

IV. REFERENCES

1. Thbit I.A., Abdulkareemm A.A. and Salim. A.H. (2021). Effect of CAPN3 gene genotypes on productive traits and carcass traits of broiler. University of Thi-Qar, College of Agriculture and Marshlands, Department of Animal Production. 2708-9339 Volume 10, Issue 1 (2021) PP 13 -24.
2. Naji, S.A. (2017). Commercial production of poultry meat. p. 180, second edition.



3. Abdulkareem, A.A. (2020). The Genetic Variations in Mitochondrial D-loop Sequence for Local Ducks in Iraq. College of Agriculture and Marshes, University of Thi Qar, Iraq. Plant Archives Vol. 20, Supplement 1, 2020 pp. 277-281.
4. World Health Organization [WHO]. 2018. Salmonella (non-typhoidal) [Online]. Available: <http://www.who.int/mediacentre/factsheets/fs139/en/>
5. Fabrega, A. & Vila, J. (2013). Salmonella enterica serovar Typhimurium skills to succeed in the host: virulence and regulation. Clinical Microbiology Reviews, 26 (2), pp. 308-341.
6. Zaman K. Hanan Manal B Saleh, and., Ezat H. Mezal (2021). Molecular Detection of Quinolones Resistance Gens of Salmonella Typhi from Gallbladder of Patients Undergoing to Cholecystectomy in Thi-Qar province/Iraq. Annals of R.S.C.B., ISSN:1583-6258, Vol. 25, Issue 6.
7. Reen, D.J. (1994). Enzyme- linked immunosorbent assay (ELISA), In: Methods in molecular biology: basic protein and peptide protocol edited by walker J.M vol (32).
8. OIE (2008): Animal Health World Organization “Avian mycoplasmosis in Manual of Standards for Diagnostic Tests and Vaccines chapter pp 482-496.
9. Rana Abdulla Salih, & Zuhair Abdulkareem. Dawah. (2022). LD50 and affective dose of Eruca sativa mill (gergeer) ethanolic extract. *University of Thi-Qar Journal of Agricultural Research*, 11(2), 11–24. <https://doi.org/10.54174/utjagr.v11i2.176>
10. Prof. Dr. Kamel K. Fahd, & Zaman M. Abdalwahad. (2022). Study of the physical and chemical characteristics of the Euphrates River in the Al-Fadhliyah district in Nasiriya governorate. *University of Thi-Qar Journal of Agricultural Research*, 11(2), 83–88. <https://doi.org/10.54174/utjagr.v11i2.184>
11. Muhammad Jodi Shahid, & Zahra’a Razzaq Hussain. (2022). Effect of feed dilution the with date kernels treated in different ways on some blood traits of broiler. *University of Thi-Qar Journal of Agricultural Research*, 11(2), 89–94. <https://doi.org/10.54174/utjagr.v11i2.185>
12. Ali Ahmed Abdul-Kareem, & Abdul-Kadhun Dakhil Raysan. (2022). Effect of CAPN1 gene polymorphism on some physical and sensory traits in broiler meat . *University of Thi-Qar Journal of Agricultural Research*, 11(2), 102–111. <https://doi.org/10.54174/utjagr.v11i2.188>

