

The prevalence of Salmonella germs in goats in some areas of Salahddin province

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Abstract: The study included isolating and diagnosing Salmonella germs from goats in four districts of Salahddin governorate (Balad, Duluiyah, Dujail and Samarra). The study included 334 goat heads from the massacres and fields that were subjected to clinical tests and data on the animal represented by age, and signs of disease, if any. (232) animals from the fields were collected (588) samples of feces, hair, milk and blood, and animals (102) collected (306) sample of feces, intestines and spleen for the purpose of bacterial isolation and serological diagnosis and characterization of antigens bacteria isolated.

Salmonella isolates were isolated from the stool (1.78%) and were due to two serotypes (*S.typhi*, *S.typhimurium*). This was confirmed by morphological traits, biochemical tests and serological profiling. Serotype *S.typhimurium* was the highest infection rate (66.6%) and the percentage of *S.typhi* was (33.3%).

Results showed that Salmonella germs (2.69%) were found in all animals (1.72%) and in carnivores (4.9%).

The distribution of salmonella germs patterns obtained from fields and slaughterhouses in the four areas showed significant differences below $P < 0.256$ in Salmonella germs isolates taken from carnivores, which had the highest rate of dilation in Duluiyah (7.69%) followed by Dujail and Samarra were the same (4%) and the lowest in a Balad (3.84%) in isolates of slaughter animals. Salmonella germs isolates were approximately equal in distribution to field animals in the study areas, the highest in a Balad (3.33%) followed by Samarra (1.78%), and Duluiyah (1.6%), and Dujail (0%). the distribution in the number of cases during the six months from the month (October to March) in the slaughterhouses and fields, where the highest in October and March by (5.35%) and was not in the month (11.76%) compared with the fields (5.12%).

Of the total (334) animals examined, 106 showed different symptoms (diarrhea, respiratory signs, heat and lethargy or more than one sign), of which only 4 were infected with salmonella bacteria and diarrhea was the highest (6.6%). While (228) animals showed no clinical signs of suspected Salmonella disease. However, (5) infected animals showed a positive result of Salmonella germs (2.19%). The infection between males and females showed no significant differences (2.65%), while in males it was 2.7%, and the ages were less than (6) months, the highest rate was (5.15%), and the age was (6) months (0.30%) the percentage of infection in the carnivores in the examined organs and feces was different. The percentage in the intestine (5.88%) was either in the spleen And (3.92%) and in the feces was (1.96%).

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I. Introduction

Salmonella germs remain one of the main causes of food-borne diseases and are also major epidemiological diseases that occur even in normal conditions (Rodrigue et al., 1990; Braden, 2006; Czerwinski et al., 2008).

Salmonella germs are the main cause of many diseases and pathogens, both in humans and in animals, which pose a danger to public health everywhere in the world (Begum et al., 2008). the disease affects people of all ages causing gastroenteritis, Diarrhea, nausea, fever and vomiting. Salmonella germs are important pathogens in infants, especially in developing countries. in some cases, infection with salmonella germs in children may develop into blood, causing bacteremia, Meningitis is sometimes caused and death (Hohmman, 2001; Khan et al., 2009; Anil et al., 2009) Salmonella effects humans and also affects field and non-field animals, including cattle, goats, sheep, horses, dogs, birds, fish and wild mammals (Radostitis et al., 2007). the severity of Salmonella infection depends on the dose of the bacteria taken and its severity, in addition to the immune state of the organism, which plays an important role in determining the severity of the injury as the immunity of the organism decreases in stress situations and also affected by age and the pattern of worship (Melling and Alder, 1998).

That the bacteria of Salmonella wide spread in the earth and there are many reasons behind the spread, including the frequent presence in the environment, the diversity of ways of transmission, and the multiplicity of serotypes, as well as increased resistance to antibiotic resistance., (Poppe et al., 1995) (Winokur et al.,2000) S. enterica is one of the most common pathogens of antimicrobial resistance (Rabsch et al., 2001). This advantage has led to an increase in infection rate and an increase in mortality due to infection of resistant serotypes (Tsai et al., 2010) that Salmonellosis is a health-related diseaseIt also causes the loss of weight of diseased animals, the reduction in the amount of milk produced, as well as abortions in pregnant females. it also causes significant economic losses resulting from health measures taken in case of food poisoning (Steinbach et al., 1997).

According to the World Health Organization (WHO) report 1997, salmonella disease was classified as a risk group III. Salmonella spread in goats in most countries of the world, with many researchers reporting various infections with salmonella bacteria (Molla et al., 2007; Chandra et al., 2005; Lemayehu et al., 2005). In the Arab world (Abdel, -ghani et al., 1997; Nabut et al., 1982) in Egypt and Saudi Arabia, respectively. In Iraq, there is a small number of studies showing that goats have been infected with Salmonella bacteria, such as by(AL-Sanjary ,1999).. Iraq is one of the countries where the spread of this disease significantly, and on the scope of human and animal injuries and for that many researchers went to study this disease, especially in the previous two decades and most of these studies focused on the axis of isolation and spread of the germ, and Iraq lacks studies on the spread of salmonella disease in goats and the identification of isolated salmonella germs pattern.

The aim of this study was to:

1. Isolation of Salmonella germs from goats in some areas of Salah al-Din governorate (Balad, Dujail, Duluiyah, Samarra).
- 2 - knowledge and diagnosis of species and serotypes that cause the infection of goats.
- 3- Studying the pathogenesis of Salmonella isolates from goats in experimentally programmed mice.

The method of work

Collection of samples

The study was carried out in the microbiology laboratory at the Faculty of Veterinary Medicine, Tikrit University and in the laboratories of the General Balad Hospital. the study included 334 goats and (1 day- 6 months) (6 months - 1 year) and more than 1 year in some areas of Salahddin province (Samarra, Balad, Dujail and Duluiyah) for the period from October 2017 to March 2018. The following information was provided on the samples (age, gender, area from which the sample was taken, date and signs) And it has been the use of veterinarians in this subject and collected sample samples of goats and as follows:

Field samples: -

Collected (588) samples of feces, milk, hair and goat's blood in the fields and from the areas mentioned above, according to scientific methods as follows:

Blood samples:-

A. Samples used for bacterial transplantation

A total of (163) blood samples were collected using EDTA tubes. They were planted directly on the media of the heart and brain diffusion broth and incubated at 37C for 24 hours. A drop of broth was then transferred using the metal carrier and planted on the media of Salmonella, mediaMacConkey and incubated 37 C for 24 hours.

B. Samples used for serological examination

A total of(69) blood samples were collected using non-container tubes on the EDTA inhibitor and left tilted at room temperature for 2-3 hours, then kept at 4 C for 24 hours and then centrifuged at 3000 cycles per minute 10 minutes to separate the serum and then collect the serum using a pipette and keep freezing at -20 C.

Stool samples

A total of (68) stool samples were collected using sterile cotton swabs, then planted directly on the mediaof tetrathionate broth, incubated 37C for 24 hours, and then transferred to the center of Salmonella - Shigella, mediaXLD and mediaMacConkey for the purpose of testing its fermentation or non-fermentation of lactose sugar, and its production of H₂S, and incubated at 37C for 24 hours (Quinn et al., 2007).

Milk samples

(56) milk samples were collected according to(Coles ,1986) as follows:

- 1- We start by examining the epilepsy clinically by the naked eye and by hand and making sure that it is free of bruises, bruises and palms.
2. The mud, dirt and dust deposited on the outside of the supplication should then be removed if present.
- 3 - Wash the udder and the surrounding areas with a dilute safflion solution 1: 1000 by using a clean and sterile cloth and leave to dry.

4. Raise the lid of the sterile test tube after teaching it to the location of the half of the sample to be taken from it and placed between the finger of the hand and diagonally under the nipple directly to avoid any contamination during the collection process.

5 - Get rid of the first bursts of milk and wiping teats with a piece of wet alcohol concentration of 70%.

6 - About 10 ml of milk was collected in sterile tubes as these tubes were sealed and then placed in a wooden box, which was stored on the ice for transport to the laboratory.

7 - In vitro and placed in the selective medium (selenite broth, or tetrathionate soup). then incubate at 37C for 24 hours, then take a drop by the loop carrier into the selective selector such as S.S agar, the media of the XLD and the media of the MacConkey.

Hair samples

(232) hair samples were collected using sterile medical scissors and placed in an eclectic selection medium (selenite broth, or tetrathionate). And then incubated 37C for 24 hours, then take a drop by the vector loop to selectively sold media such as (S.S agar) and the media of the XLD and the media of the MacConkey and spread it, and incubated 37 C for 24 hours for the purpose of testing fermentation or not fermentation For lactose sugar and its production of H₂S (Quinn et al., 2007).

Slaughter samples

A total of (306) different samples were collected from (102) head of goats in the massacres scattered in the areas mentioned above. The samples include:

Spleen

A total of 102 specimens were collected from the spleen and samples were taken in the selective eclectic medium (selenite broth, or tetrathionate). Then incubate 37C for 24 hours, then take a drop by the loop carrier into the selective selector such as S.S agar and spread it to 37C for 24 hours.

The colonies were then isolated from the selective (Brilliant green agar and XLD) for purification of the colonies (Quinn et al., 2004).

Intestine

A total of (102) intestinal samples were taken, which take part of the intestine and are placed in the selective enrichment media (selenite broth, or tetrathionate). The same steps as mentioned in the spleen samples are performed.

Feces

A total of (102) feces were collected from animals in slaughterhouses after slaughtering in the same manner as previously mentioned.

bacterial diagnosis

Salmonella germs were developed on solid agricultural communities, MacConkey media agar, S-S agar and media XLD after incubation at 37C for 24 hours initially, depending on phenotypic traits including colony size, color, shape, and ability to Fermentation of lactose sugar or non-fermentation in the center of the MacConkey as well as microscopic examination after dyeing chromium and the shape of the bacteria and color was examined by optical microscopy.

Biochemical Tests: -

That the process of biochemical diagnosis was based on the methods developed by (Jawetz et al., 2001; Quinn et al., 2007). If more than one pure bacterial colony was taken after the shape and nature of its pigmentation were determined by chromium and the following chemical tests were used:

1. Indol test 2-Methyl red test

3. Voges-Proskauer test 4. Citrate utilization test

5. H₂S production test 6. Urea hydrolysis test

7. Oxidase test 8. Catalase test

9. Gelatin hydrolysis test 10. Sugar fermentation test

11- Motility test

Results and discussion

Results of bacterial isolation

The results of the study showed 334 goats with 9 animals infected with Salmonella germs. The bacteria were isolated from (4) animals out of (232) animals in the fields and (1.72%). The animals infected with Salmonella germs in the slaughterhouses The number of (5) out of (102) animals and by (4.9%).

Table (1) shows the numbers of goats and the samples taken from fields and slaughterhouses

Position	NO. of goats	NO. of infected	Percentage of animals infected	NO. of samples	Positive	Percentage of positive samples
Field	232	4	%1.72	588	4	%0.68
Slaughter	102	5	%4.9	306	12	%3.92
Total	334	9	%2.69	894	6	%1.78

It was ascertained that the isolates belonged to Salmonella species and their serotypes through: The ratios were also in line with the results of CDC research (CDC, 2002) that S.typhimurium was one of the most common types of Salmonella. Table 2 shows isolated serotypes

Table(2) shows the serotypes that have been isolated

Serotype	No.of positive sample	%	No.ofinfected animals	%
S. typhimurium	11	%68.75	6	%66.6
S. typhi	5	%31.25	3	%33.3
Total	16	%100	9	%100

Percentage of goats infected with Salmonella bacteria in study areas:- in the fields

The number of infected animals and their percentages is shown in Table (5-4) distributed to four regions of a country where the infection rate was (3.33%), the highest rate in Samarra was (1.78%) and in Duluiyah (1.6%), While in Dujail it was (0%). Therefore, the percentage of total infection in the fields of the study areas reached (1.72%).

Table (3) shows the numbers of goats and the samples taken from fields

Field	NO.of goats tested	NO.of goats right	NO. of infected	Percentage of animals infected
Balad	60	58	2	%3.33
Duluiyah	60	59	1	%1.6
Dujail	56	56	0	% 0
Samarra	56	55	1	%1.78
Total	232	228	4	%1.72

in the Slaughte

Table shows the number of infected goats in the massacres and their percentages in the study areas themselves. The percentage of infection in the town of Balad (3.84%) and in Duluiyah (7,69%) and in Dujail and Samarra (4%). The percentage of total infection in the massacres (4.9%).

Table (4) shows the numbers of goats and the samples taken from Slaughte

Slaughter	NO.of goats tested	NO.of goats right	NO. of infected	NO. of sample	Percentage of animals infected
Balad	26	25	1	77	%3.84
Duluiyah	26	24	2	77	%7.69
Dujail	25	24	0	76	%4
Samarra	25	24	1	76	%4
Total	102	97	5	306	%4.9

Percentages of Salmonella isolates isolated from organs and feces.

The results of the samples taken from different organs and stool in the massacres (spleen, intestines, feces) showed different percentages of Salmonella infection, the highest was in the intestines (5.88%), followed by isolation rate in spleen (3.92%). (1.96%). this indicates that the body may not continuously release Salmonella germs with feces, especially the stocked and bactericidal animals, which may be sporadically released and may be associated with stressors (Neilson et al., 2010) The bacterial transplantation of feces should be repeated five times in succession and at different time intervals it was possible to exclude the animal's infection with Salmonella germs which he confirmed. (Smith et al., 2009).

Table(5) shows the percentage and percentage of Salmonella positive models of organs and feces

Position	Samples	NO. of sample	Positive	Percentage of positive samples
Slaughte	Spleen	102	4	%3.92
	Intestine	102	6	%5.88
	Feces	102	2	%1.96
Field	Feces	68	1	%1.47
	Hair	232	0	%0
	Milk	56	1	% 1.78
	Blood	232	2	%0.86
Total		894	16	%1.78

Percentage of injury by month of year

During the six months of the study, the goats tested in the massacres and fields showed a clear difference in the rate of infection. it is noted that the increase or decrease in temperature and humidity has a clear effect on isolating the Salmonella germs from goats, which was the highest rate of infection in the Slaughte in October, (11.76%) followed by the month of November, December and March (5.88%). In January and February there were no injuries (0%). In the fields, (5.12%), followed by October and November (2.56%) and the percentage (0% December, and February).

Table(6) shows the number of cases of infection distributed by months of study and percentage

Month of study	No.of tested			No.of Infected			%		
	Slaughte	Field	Total	Slaughte	Field	Total	Slaughte	Field	Total
October	17	39	56	2	1	3	%11.76	%2.56	%5.35
November	17	39	56	1	1	2	%5.88	%2.56	%3.57
December	17	38	55	1	0	1	%5.88	%0	%1.81
January	17	38	55	0	0	0	%0	%0	%0
February	17	39	56	0	0	0	%0	%0	%0
March	17	39	56	1	2	3	%5.88	%5.12	%5.35
Total	102	232	334	5	4	9	%4.90	%1.72	%2.69

Percentage of injury by sex

Table(7) shows the proportions of infected goats distributed by sex, where it was found that the percentage of infection in males was (2.7%) for the types of Salmonella S.typhimurium and S.typhi. In females, the incidence rate was (2.65%). Also . the results did not show significant differences between males and females for different types of Salmonella infection.

Table(7) shows percentages of *Salmonella* isolates isolated by sex

Sex of goats	NO.of Animal	NO.of Infected	%	S. typhimurium	S. typhi
Male	108	3	% 2.7	2	1
Female	226	6	%2.65	4	2
Total	334	9	%2.69	6	3

Percentage of injury by age group

Table(8) shows the percentage of goats infected with *Salmonella* germs by age. the goats (6 months) have a percentage of (5.15%) and the *Salmonella* species mentioned above, the highest rate of infection among the age groups. the percentage of infection (2.30%) and *Salmonella* species were also found in goats (6 months to 1 year). The age group of goats that had the lowest percentage of infection was the age group (0.93%) *S. typhimurium* only.

Table(8) shows the percentage of cases of isolated *Salmonella* types by age group

Age group of goats	NO.of Animal	NO.of Infected	%	Divide infected for serotype	
				S.typhimurium	S. typhi
Below 6 months	97	5	%5.15	4	1
Between 6 mth- 1 year	130	3	%2.30	1	2
More than 1 year	107	1	%0.93	1	0
Total	334	9	%2.69	6	3

الخلاصة

تضمنت هذه الدراسة عزل وتشخيص جراثيم السالمونيلا من الماعز في اربع مناطق من محافظة صلاح الدين (بلد ، الضلوعية ، الدجيل ، سامراء)، اشتملت الدراسة على (334) رأس ماعز من المجازر و الحقول أجريت عليها الفحوصات السريرية ودونت البيانات عن الحيوان متمثلة بالعمى والموسم والجنس والعلامات المرضية الظاهرة في حال وجودها . ثم وزعت الحيوانات بالشكل الآتي ، (232) حيواناً من الحقول جمعت منها (588) عينة من البراز والشعر والحليب و الدم ، و حيوانات المجازر عددها (102) جمع منها (306) عينة من براز و أمعاء و طحال لغرض العزل الجرثومي والتشخيص المصلي وتوصيف مستضدات الجراثيم المعزولة.

تم عزل (16) عذلة سالمونيلا من البراز و الاعضاء بنسبة (1.78%)، وكانت عائدة الى نمطين مصلية (S.typhi , S.typhimurium) وقد تم التأكد من ذلك من خلال الصفات الشكلية والاختبارات الكيمحيوية والتنميط المصلي ، وقد شكلت نسبة الاصابة بالنمط المصلي *S.typhimurium* اعلى نسبة إذ بلغت (66.6%) وكانت نسبة الاصابة بالنمط المصلي *S.typhi* بنسبة (33.3%) .

اظهرت النتائج نسبة اصابة بجراثيم السالمونيلا (2.69%) في جميع الحيوانات إذ بلغت في حيوانات الحقول (1.72%) اما في حيوانات المجازر فكانت النسبة (4.9%) .

اما توزيع انماط السالمونيلا التي تم الحصول عليها من الحقول والمجازر في المناطق الاربعة فقد اظهرت النتائج على وجود فروق غير معنوية تحت مستوى ($P>0.256$) ينسب عزلات السالمونيلا المأخوذة من حيوانات المجازر إذ كانت اعلى نسبة في الضلوعية (7.69%) ثم تلتها الدجيل وسامراء بنفس النسبة (4%) واقلها في بلد بنسبة (3.84%) في عزلات حيوانات المجازر ، وقد تساوت نسب عزلات السالمونيلا تقريباً عند توزيعها على حيوانات الحقول في مناطق الدراسة إذ بلغت اعلاها في بلد وكانت (3.33%) ، ثم سامراء (1.78%) ، ثم الضلوعية (1.6%) وكانت النسبة في الدجيل (0%) ، اما توزيع حالات و عدد الاصابات على اشهر الدراسة الستة (من شهر تشرين الأول -شهر اذار) في حيوانات المجازر والحقول إذ كانت اعلاها في شهري تشرين الأول واذار بنسبة (5.35%) وانعدمت في شهري كانون الثاني وشباط ، واكثرها في المجازر بنسبة (11.76%) مقارنة مع الحقول التي كانت بنسبة (5.12%) .

ومن مجموع (334) حيواناً مفحوصاً اظهر (106) اعراضاً مختلفة (اسهال ، علامات تنفسية ، حرارة وخمول او اكثر من علامة) إذ كان من بينها (4) حيوانات فقط مصابة بجراثيم السالمونيلا وشكل الاسهال اعلى نسبة (6.6%) ، في حين كان (228) حيواناً لم تظهر عليه اي علامات سريرية تعطي شك للإصابة بداء السالمونيلا ومع ذلك ظهر (5) حيوانات مصابة أعطت نتيجة موجبة لجراثيم السالمونيلا شكلت نسبة (2.19%) ولم تظهر الاصابة بين الذكور و الاناث فروق معنوية حيث سجلت في الاناث نسبة الاصابة (2.65%) بينما في الذكور كانت النسبة (2.7%) ، وقد سجلت الاعمار اقل من (6) اشهر اعلى نسبة اصابة (5.15%) واعمار بين (6) اشهر – سنة (2.30%) واقلها اكبر من سنة إذ شكلت النسبة (0.93%)، شكلت نسبة الاصابة في حيوانات المجازر في الاعضاء المفحوصة والبراز نسب مختلفة ، إذ كانت النسبة في الأمعاء (5.88%) اما في الطحال (3.92%) وفي البراز كانت النسبة (1.96%) .

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