10.21608/avmj.2025.309429.1335 Assiut University web-site: www.aun.edu.eg

SEROLOGICAL ASSESSMENT OF THE OCCURRENCE OF *SALMONELLA ABORTUSOVIS*, *BRUCELLA* SPP., AND *LISTERIA MONOCYTOGENES* AMONG EWES IN BABYLON PROVINCE, IRAQ

ALI DHIAA MARZA [1]; ALI HAMID RABEEA [1] AND ABDULAMIR ALI ALZAHID [2]

[1] Department of Veterinary Internal Medicine, College of Veterinary Medicine, Al-Qasim Green University, Iraq

[2] Department of Medical Laboratory Technologies, College of Medical Sciences, Al-Amal University, Iraq

Received: 24 September 2024; **Accepted:** 21 November 2024

ABSTRACT

The current study aimed to determine the infection rate of *Salmonella abortusovis, Brucella* spp*,* and *Listeria monocytogenes* among ewes in Babylon Province. For this purpose blood samples were collected from 50 aborted ewes, 15- 30 days after abortion, and 10 normal deliveries of ewes during the period from September 2021 to June 2022. The collected samples were evaluated by using ELISA. The results showed that from the aborted cases, 78% (39/50 cases) were positive for one or more of the diseases examined, while regarding the 11 normal deliveries, only 27% (3 out of 11) were found positive. Estimation of infection rate among the aborted ewes using Anti-*Salmonella abortusovis* antibody*,* anti-*Brucella* spp antibody and anti-*Listeria monocytogenes* antibody detected positive cases at rates of 66%, 22% and 12% respectively, and 25.6% of cases gave antibody titre against more than one etiology that was subjected in the current study. The current study reveals that 78.7% of abortions caused by *Brucella* spp. took place during the third trimester.

Key words*:* Abortion, Brucellosis, Salmonellosis, Listeriosis

INTRODUCTION

Abortion is defined as the expulsion of an immature fetus (one or more) before the completion of the normal pregnancy period, which is either dead or remains alive for a period of less than 24 hours (Clothier *et al*., 2020 and Deresa *et al*., 2020). Abortion can be caused by either infectious or noninfectious such as nutritional stress, which leads to decrease in the size or lysis of the corpus luteum and a decrease in progesterone level. The consumption of certain types of toxic plants leads to the closure of the binding site of Oesterodiol – B17 or has a direct toxic effect on the fetus (Radostits *et al*., 2007).

Infectious abortion in ewes may be associated with viral, parasitic, fungal, or bacterial infections, which are the most important causes. *Brucella* spp*., Salmonella* spp*., Listeria* spp*., Campylobacter* spp*., and*

Corresponding author: ALI HAMID RABEEA
E-mail address: alirabee@vet.uoqasim.edu.iq
Present address: Department of Veterinary Internal Medicine, College of Veterinary Medicine, Al-Qasim Green University, Iraq. 07823112420

Chlamydia spp.,are the main bacterial causes of ewe abortion (Blood *et al.,* 1989).

Many methods are used for diagnosis of bacterial abortions in ewes, including direct methods (smear staining, culture methods and genetic methods) or indirect methods (detection of humeral and cellular immune response against causative agents). ELISA consider the sensitive and specific test used for detection of humeral immune response (Latimer *et al.*,2004). The current study is designed to examine the infection rate of *Salmonella abortusovis*, *Brucella* spp., and *Listeria monocytogenes* among ewes in Babylon province and investigating some epidemiological aspects related to these infections.

MATERIALS AND METHODS

The study was carried out during the period from September 2021 to June 2022, blood samples were collected under aseptic conditions into 5 ml sterile tubes from 60 ewes (50 from aborted ewes in a period from 15- 30 days after the abortion and 10 normal deliveries ewes) of the Babylon province then samples were transported in isothermal containers with ice (4 °C ± 2) for less than an hour to the Internal and Preventive Medicine Branch, College of Veterinary Medicine/Al-Qasim Green University, Babylon, Iraq. The serum was obtained by centrifugation of blood samples at 3000 rmp for 15 min within 24 h of collection. It was then stored at -20°C to be used for detection of antibodies.

Serodiagnosis was done using the following ELISA kits:-

a- Anti *Salmonella abortusovis* antibody: detected by use of ELISA kit (Creative Diagnostics- USA) and according to the manufacturer's instructions

b- Anti *Brucella* Antibody: detected by use of ELISA kit (SVANOVIR®Brucella-Ab I-ELISA- Spain) (captures antibodies to *B. abortus and B. melitensis.*) and according to the manufacturer's instructions.

c- Anti *Listeria monocytogenes* antibody: detected by use of ELISA kit (Mybiosource- UK) and according to the manufacturer's instructions.

RESULTS

Out of 50 aborted ewes, 39 (78%) tested positive for one or more diseases, while 11 (22%) were ELISA negative for any of the investigated diseases. In contrast, among 10 ewes with normal deliveries, 3 (30%) tested positive, and 7 (70%) were negative (Table 1).

Table 1: Prevalence of examined diseases among aborted and normal deliveries ewes

Animals	Total No.	+ve for one or more examined diseases		-ve for any of examined diseases	
		No.	%	No.	%
Aborted	**50**	**39**	**78**	**11**	**22**
Normal deliveries	**10**	**3**	**30**	**7**	**70**
Total	**60**	**42**	**70**	**18**	**30**

Infection details of each disease separately was illustrated in Table (2). The results demonstrate that *Brucella* spp. was the most commonly detected pathogen, followed by *Salmonella abortusovis* and *Listeria monocytogenes*.

Table 2: ELISA results for aborted and normal deliveries ewes.

Animals	Total No.	Anti *Salmonella abortusovis*		Anti *Brucella* spp. Antibody		Anti *Listeria Monocytogenes* antibody	
		No.	%	No.	%	No.	%
Aborted	50	11	22	33	66	6	12
Normal deliveries	10	1	10	2	20	0	0
Total	60	12	20	35	58.3	6	10

Results from Table (3) indicate that mixed infections were identified in 10 cases (25.6%), whereas 29 (74.3%) cases were confirmed to have a single pathogen.

Table 3: Frequency of mixed versus single infection among aborted ewes

Types of antibodies	Number of positive samples	Rate of positive samples (out of cases)
Anti *Brucella spp.* Antibody+ Anti *Salmonella abortusovis*	7	17.9%
Anti Brucella Antibody+ Anti *Listeria Monocytogenes* antibody	2	5.1%
Anti *Salmonella abortusovis* + Anti *Listeria Monocytogenes* antibody	1	2.5%
Total of mixed infection	10	25.6%
Single infection	29	74.3%
Total	39	100%

The relationship between abortion and the stage of pregnancy (Table 4)

- **Brucellosis**: Most cases (78.7%) occurred in the third trimester.
- **Salmonellosis**: Cases were distributed across the second (36.3%) and third trimesters (45.4%).
- **Listeriosis**: Primarily occurred in the first trimester (66.6%).

Table 4: The relationship between causative agents and the semester of abortion.

Type of infection	No.	cases in each Semester					
		1st		2nd		3rd	
		No	%	No	%	No	%
Brucellosis	33	2	6	5	15.1	26	78.7
Salmonellosis	11	2	18.1	4	36.3	5	45.4
Listeriosis	6	4	66.6	0	0	2	33.3

The results indicate that recurrent abortions were observed in cases of Brucellosis and Salmonellosis, but not in Listeriosis.

Table 5: The relationship between abortions recurrence with infection type

Infection type	No	abortion recurrency	
		No	%
Brucellosis	33	4	12.5%
Salmonellosis	11	1	9.0%
Listeriosis	6	0	0%

DISCUSSION

These results indicate *Brucella spp*. induce abortion in ewes, shown by its greater detection among isolates in comparison with Salmonella abortusovis and Listeria monocytogenes. This is in agreement with earlier studies that always reported Brucella spp. the most common cause of abortion in ruminants (Clothier et al., 2020; Deresa et al., 2020; Radostits et al., 2007; Hamzah et al., 2020).

Brucellosis was the leading infection diagnosed as 66% of the aborted ewes were positive for Anti-*Brucella spp*. antibodies. Such a high prevalence rate of Brucella spp. is in line with other reports. in abortive ewes (Arif et al., 2020; Al-Dabagh et al., 2014).

Our findings support the notion that *Brucella spp.* induce most abortions in ewes in the late stage of pregnancy (third semester) (Blood et al., 1989; Menzies, 2012; Mohmood et al., 2020). A recent study suggested that this could be a result of an interaction with a chemotactic factor, reythritol, for Brucella spp. in the placenta (Roop et al., 2012).

About one quarter of the studied ewes (22%) were positive for *Salmonella abortusovis*. This percentage was lower than that reported by Havrun et al. (2006) and Wirz-Dittus et al. (2010). This difference in prevalence rate could be affected by different sorts of diagnosis, sample size, or geographic differences. This indicates the impact of this pathogen and the necessity of continuous monitoring and control of it (Roshan et al., 2018; Giannati-Stefanou et al., 1997)

In the current study, only 12% of aborted ewes were caused by *Listeria monocytogenes,* which is lower than a previous study (Ayoub et al., 2020). Abortion caused by Listeriosis mostly occurred in the first trimester (Gojam & Tulu, 2020; Mikaeel & Al-Saeed, 2020), as we also noted. The disparity in the detection rate could be attributed to the epidemiologic setting of the studied populations and the sensitivity of the diagnostic assays.

Our survey identified that 25.6% of the cases had antibodies to many pathogens, referring to either coinfection or remaining antibodies from previous infection or even vaccination. Therefore, a correct diagnosis utilizing the serological test is not enough alone Nielsen & Yu, 2010; Tizard, 1988). In this study, the indirect ELISA test was unable to distinguish the current infection from the past exposure, and may overestimate an active infection.

Taken the association of the trimester of abortion and the aetiological agents together indicated the following distribution: *Brucella spp.* was more frequent in the third trimester, and *Listeria monocytogenes* in the first one. This agreed with previous studies (Nooruldeen et al., 2021 and Aghwan et al., 2021).

Repeated abortion was observed in cases of *Brucella spp.* and *Salmonella abortusovis,* suggesting repeated exposure or insufficient immunity to former infections. This should draw the attention of veterinarians to ensure effective vaccination and revise the control measures to avoid a possible future outbreak (Blasco, 1997; Ismaeili et al., 2021).

In conclusion, the current study confirms the necessity of improving diagnosis for better identifying and controlling the causative pathogen of ovine abortion to maintain the health of animal livestock and achieve economic growth and higher production to face poverty and end hunger to achieve the Sustainable Development Goals (SDGs).

REFERENCES

Aghwan, S.S.; Al-Bakri, H.S. and Albaqqal, S.M. (2021): Comparison the efficiency of different techniques for the diagnosis of Toxoplasma gondii infection in slaughtered ewes. *Iraqi Journal of Veterinary Sciences*, *35*, 19-23.

Al-Dabagh, I.I.; Jasim, B.M. and Jarjees, M.T. (2014): Seroprevalence of antibodies to toxoplasmosis, brucellosis and chlamydiosis in abortive sheep in Nineveh governorate, Iraq. *Iraqi Journal of Veterinary Sciences*, *28*(1), 21-25.

Arif, E.D.; Saeed, N.M. and Rachid, S.K. (2020): Isolation and Identification of from Aborted Ewes in Sulaimani Province, Northern Iraq. *Polish Journal of Microbiology*, *69*(1), 65-71.

Ayoub, M.; Abu-Rawash, A.; Ibrahim, S. and Aragan, A. (2020): Hygienic Studies on Microbial Causes of Abortion in Sheep. *Damanhour Journal of Veterinary Sciences*, *5*(1), 11-13.

Blasco, J.M. (1997): A review of the use of *Br. melitensis* Rev.1 vaccine in adult sheep and goats. Prev. Vet. Med. 31: 275-283.

Blood, D.C.; Radostits, O.M. and Henderson, J.A. (1989): Veterinary medicine. A textbook of the diseases of cattle, sheep, pig, goats and horses 5th ed. Bailliere tindall. London. P: 677-696.

Clothier, G.; Wapenaar, W.; Kenny, E. and Windham, E. (2020): Farmers' and veterinary surgeons' knowledge, perceptions and attitudes towards cattle abortion investigations in the UK. *Veterinary Record*, *187*(11), 447-447

Deresa, B.; Tulu, D. and Deressa, F.B. (2020): Epidemiological investigation of cattle abortion and its association with brucellosis in Jimma zone, Ethiopia. *Veterinary Medicine: Research and Reports*, *11*, 87.

Esmaeili, H.; Rad, Z.N. and Hamedi, M. (2021): Identification of the Excretion of Brucella Melitensis Vaccine Strain Rev. 1 in Lactating Ewes and the Assessment of Antibody Response in their Lambs. *Journal of Medical Bacteriology*, 48-52.

Giannati-Stefanou, A.; Bourtzi-Hatzopoulou, E.; Sarris, K. and Xenos, G. (1997). Epizootiological. Study of sheep and goat abortion by Salmonella abortus ovis in Greece. *Journal of the hellenic veterinary medical Society*, *48*(2), 93-98.

Gojam, A. and Tulu, D. (2020): Infectious causes of abortion and its associated risk factor in sheep and goat in Ethiopia. *Int. J. Vet. Sci. Technol*, *4*, 008-012

Habrun, B.; Listes, E.; Spicic, S.; Cvetnic, Z.; Lukacevic, D.; Jemersic, L. and Kompes, G. (2006): An outbreak of Salmonella Abortusovis abortions in sheep in south Croatia. *Journal of Veterinary Medicine, Series B*, *53*(6), 286-290.

Hamzah, K.J.; Alhtheal, E.D. and Ibraheem, A.K. (2020): Pathological changes induced by *klebsiella* pneumonia in immunized rats wit whole sonicated klebsiella pneumonia antigens mixed with certain adjuvants. Biochemical and Cellular Archives, 20(2).

Latimer, K.S.; Mahaffey, E.A. and Prasse, K.I. (2004): Clinical pathology: veterinary laboratory medicine.

Mahmood, A.K.; Hamzah, K.J.; Dirwal, A.R. and Salh, A.H. (2020): Isolation of Escherichia coli from skin wounds in cow. *Plant Archives*, *20*(1), 3108-3110.

Menzies, P.I. (2012, September): Diagnosis and control of small ruminant abortion. In *American Association of Bovine Practitioners Proceedings of the Annual Conference* (pp. 129-137).

Mikaeel, F.B. and Al-Saeed, A.T.M. (2020): Serological and molecular diagnosis of Toxoplasma gondii among ewes and horses in Duhok province-Iraq. *The Iraqi Journal of Agricultural Science*, *51*(4), 1212-1219.

Nielsen, K. and Yu, W. L. (2010): Serological diagnosis of brucellosis Contributions, Sec. Biol. Med. Sci; MASA, XXXI, 1, p. 65–89.

Nooruldeen, M.Y.; Jaafar, S.E. and Salih, A.I. (2021): Seroprevalence of Neospora caninum infections in cattle in Kirkuk province. *Iraqi J Vet Sci*, *35*(2), 331-334.

Radostits, O.M.; Henderson, J.A.; Blood, D.C.; Arundel, J.T. and Gay, C.C. (2007): "Veterinary Medicine: A Textbook of the Diseases of Cattle, Sheep, Pigs, Goats, and Horses". 11th Ed; W.B. Saunders Elsevier. UK. Chapter 18: 966-993.

Roop, R.M.; Barton, I.S.; Hopersberger, D. and Martin, D.W. (2021): Uncovering the hidden credentials of Brucella virulence. *Microbiology and Molecular Biology Reviews*, *85*(1), e00021-19.

Roshan, H.M.; Saadati, D. and Najimi, M. (2018): Molecular detection of Brucella melitensis, Coxiella burnetii and Salmonella abortusovis in aborted fetuses of Baluchi sheep in Sistan region, south-eastern Iran. *Iranian journal of veterinary research*, *19*(2), 128

Tizard, I. (1988): An Introduction to Veterinary Immunology. PA, W.B. Saunders Co. Philadelphia

Wirz-Dittus, S.; Belloy, L.; Doherr, M.G.; Hüssy, D.; Sting, R.; Gabioud, P. and Waldvogel, A.S. (2010): Use of an indirect enzyme-linked immunosorbent assay for detection of antibodies in sheep naturally infected with Salmonella Abortusovis. *Journal of veterinary diagnostic investigation*, *22*(4), 531-536.

التقييم المصلى لحدوث عدوى السالمونيلا ابورتوس أوفيس والبروسيلا وليستيريا مونوسيتوجينس في النعاج بمحافظة بابل العراق

على ضياء مرزه ، على حامد ربيع ، عبد الامير على

Email: alirabee@vet.uoqasim.edu.iq Assiut University web-site: www.aun.edu.eg

هدفت الدراسة الحالية إلى معرفة معدل انتشار السالمونيلا ابورتوسوفيس وبروسيلا spp وليستيريا مونوسيتوجينس في النعاج في محافظة بابل، ولهذا الغرض تم جمع عينات الدم من 50 نعجة مجهضة و10 نعجات ولادة طبيعية في الفترة من 15-30 يوم بعد الإجهاض خلال الفترة من سبتمبر 2021 إلى يونيو 2022 وتم إجراء اختبارات ELISA. أظهرت النتائج أن 22% (11 من 50) من حالات الإجهاض كانت سلبية لثلاثة اختبارات استخدمت في الدراسة الحالية بينما كانت 78% (39 من 50) إيجابية. تم الكشف عن الأجسام المضادة لـ Salmonella abortusovis و Brucella spp و Listeria monocytogenes بنسب 66% و 22% و 12% على التوالي، و 25.6% من الحالات تعطي عيار الأجسام المضادة ضد أكثر من مسبب واحد من الأمراض التي خضعت للدراسة الحالية. تكشف الدراسة الحالية أن 78.7% من حالات الإجهاض الناجمة عن Brucella spp حدثت في الثلث الثالث من الحمل. ختاما, تؤكد هذه الدراسة على ضرورة تحسين وتطوير التشخيص لتحديد والسيطرة على مسببات الاجهاض في الاغنام للحفاظ علة صحة الثروة الحيوانية وتحقيق النمو الاقتصادي لمواجهة الفقر والقضاء على الجوع لتحقيق أهداف التنمية المستدامة.