INNSPUB

International Journal of Biosciences | IJB |
ISSN: 2220-6655 (Print), 2222-5234 (Online)
http://www.innspub.net
Vol. 15, No. 2, p. 348-354, 2019

RESEARCH PAPER OPEN ACCESS

Detection of bovine tuberculosis in cow carcasses in Baghdad

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Key words: Tuberculosis, Bovine, Mycobacteria, Carcasses, Lymph node.

http://dx.doi.org/10.12692/ijb/15.2.348-354 Article published on August 24, 2019

Abstract

Bovine Tuberculosis is a contagious disease ,both animals and humans are highly susceptible to infection, infected animal act as source for infected farmers, veterinarians, slaughterhouse workers, butchers so the importance of our study is to stand on the extent of tuberculosis infection in cow carcasses in three abattoris of Baghdad (Al-shula,Al-karkh and Al-dura.), Thousands carcasses were examined, six hundreds and fourty two samples of lymph nodes and organs were collected from (128) carcasses these involved (10) retropharyngeal lymphnode, (8) portal lymph nodes,(110)mesenteric lymph node(85)spleen samples, (4) supramammary lymph node, and (4)uterus samples, the samples were transported to laboratory for bacteriological culturing The were processed for isolation of mycobacteria following standard procedures for homogenization, suspension, centrifugation and decontamination with 4 % of NaOH and were inoculated on Lowenstein-Jensen (L-J) media with and without pyruvate and incubated at 37°C incubator for culture for a maximum up to 8 wk. after growth appear direct microscopic examination and biochemical test for mycobacteria (growth on mac-conckey without crystal violet ,urease test, catalas test,nitrate reduction,(thiophen2 carboxylic acid hydroxide(TCH),sensitivity to antimycobacterial drug ethambutol (EMB), Isonicotinic hydrazide (INH), Streptomycin(SM), Rimfampicin (RMP), Para-aminosalicylic acid(PAS). the results indicated that the percentage of infection(2.8% and0.6%)in Al-shula and Al-karkh abattoirs respectively while AL-dura abattoir was found (0%)and (8.1%) of lymphnode ,(17.6%)of lung,(3.8%)of livers,(3.5%)of spleen and (25%) for each supramammary lymph node and uterus infected with bovine tuberculosis twenty bacterial isolated belonged to mycobacterium were isolated from carcasses,(17) of them were *Mycobacterium bovis*.this is the first study in isolation of *Mycobacterium bovis* from cow carcasses in Iraq.

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Introduction

Bovine tuberculosis is considered to be the most frequent infection in cattle, it is estimated that *M. bovis*, the etiological organism of TB in bovines is also responsible for 5% of all TB infections in humans**,** Tuberculosis is a respiratory disease and transmission of infection is mainly by the airborne route. Susceptible species to bovine tuberculosis which cause by *M.bovis*, include cattle, humans, goats, cats dogs, buffalo and other type of animals, *M. bovis* infection was recognised as a major public health problem when this organism was transmitted to man via milk from infected cows. Human working with cattle infected with bovine tuberculosis, on the farm or in the slaughter house, are more likely to develop pulmonary disease than alimentary disease. (O'Reilly and Daborn, 1995). Cattle shed *M. bovis* in respiratory secretions, feces and milk, urine, vaginal secretions or semen. *M. bovis* is transmitted between cattle by inhlation or ingestion, the organism can cause Cutaneous, genital, and congenital infections. Due to behavioral changes badgers and possums are most likely to transmit *M. bovis* to cattle during the late stages of disease (Menzies and Neill,2000).The fact that *M. bovis* has been isolated from various animal products such as fresh and sour milk, from lesions in the lung and lymph nodes at slaughterhouses, as well as from sputum and biopsy samples of humans, indicates that the disease spreads through both direct and indirect modes of transmission (okolo, 1992) infection of the uterus with abortion, the economic loss from infected the cow is characterized by decreasing in production of milk and meat and sterility and the infected cow will loss (10-25%) from production, the infection in human accrued through inhalation contaminated air with mycobacteria or ingestion of contaminated milk ,or meat from infectd animal and worker in farm and slaughterhouse and veterinarian were most infected (Hernandez and Baca, 1998). So the aim of our study to stand on the extent of bovine tuberculosis infection in cow carcasses in the three main abattoris of Baghdad government (Al-shula,Al-karkh and Al-dura.) In Iraq.

Material and methods

Samples collected from cow carcasses in the three main abattoris of Baghdad government (Al-shula, Al-karkh and Al-dura). Thousands carcasses were examined, six hundreds and fourty two samples of different lymph nodes and organs were collected from (128) suspected carcasses these involved (10)retro pharyngeal lymph node, (8) portal lymph nodes, (110) mesenteric lymph node(85)spleen samples, (4) supramammary lymph node, and (4)uterus samples the sample were transported to laboratory.

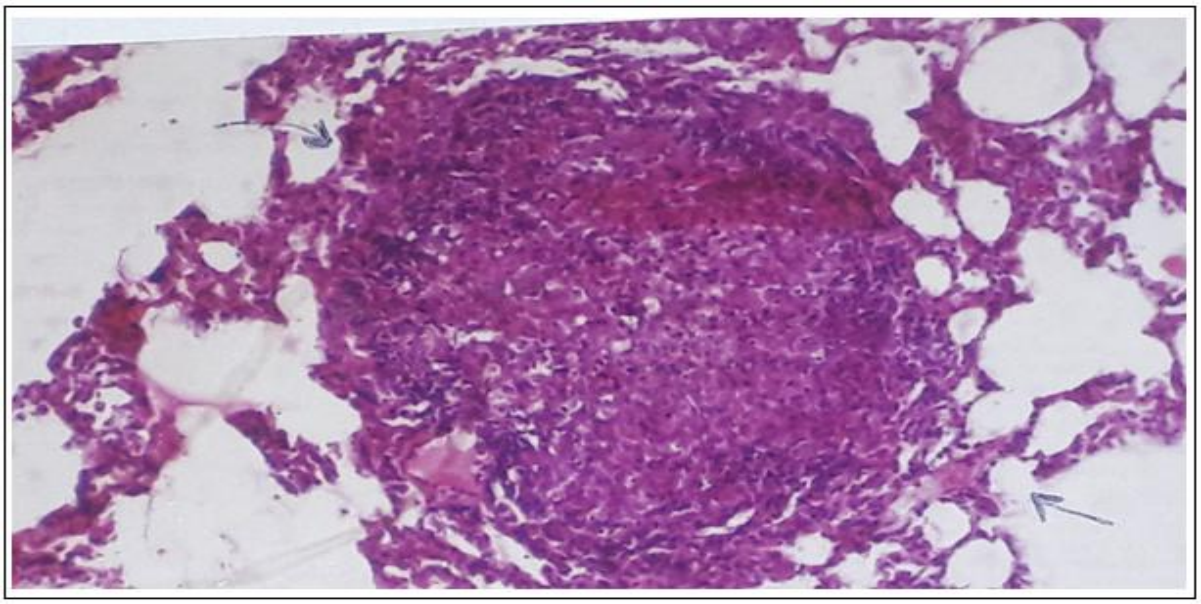

Fig. 1. Histopathological section in lung with tubercle and emphysema stained by Hematoxylin and eosin stain(x125).

Bacteriological culturing

The samples were processed for isolation of mycobacteria following standard procedures for homogenization, suspension, centrifugation and decontamination with (4%) NaOH (Claxton *et al.*, 1979). about 50 µl of processed sample were inoculated on Lowenstein-Jensen (L-J) media with and without pyruvate and incubated at 37°C incubator for culture for a maximum period up to 8 wk. direct microscopic examination after growth appear on culture.

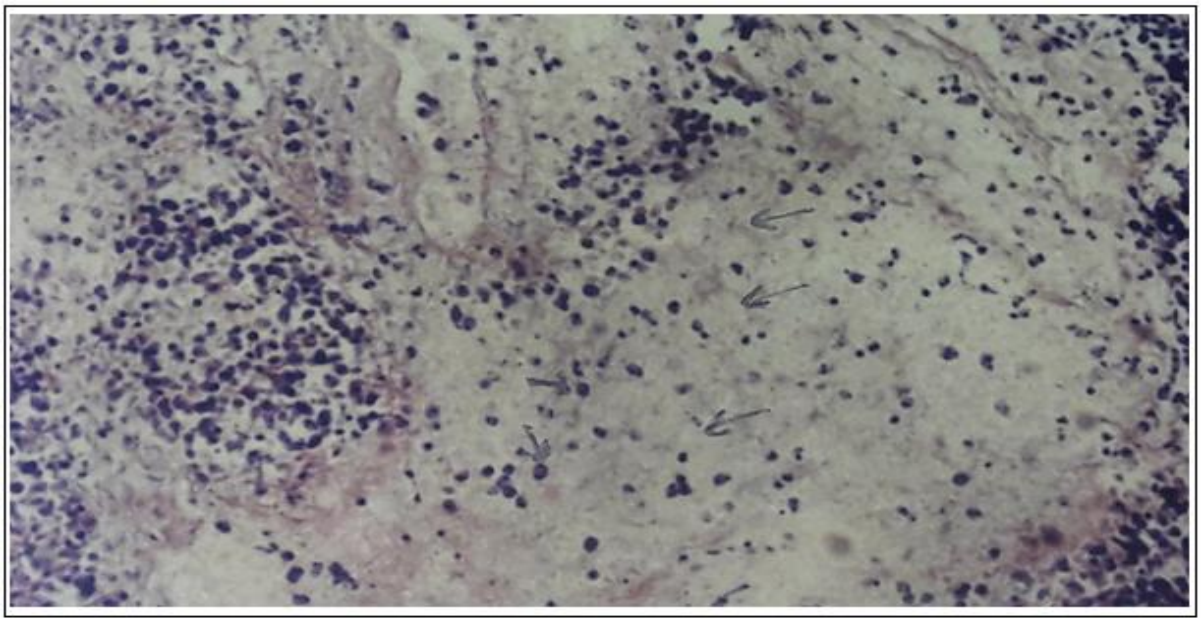

Fig. 2. Histopathological section in in lymph node with casesous necrosis and infiltration of monocyte stained by Hematoxylin and eosin stain (x250).

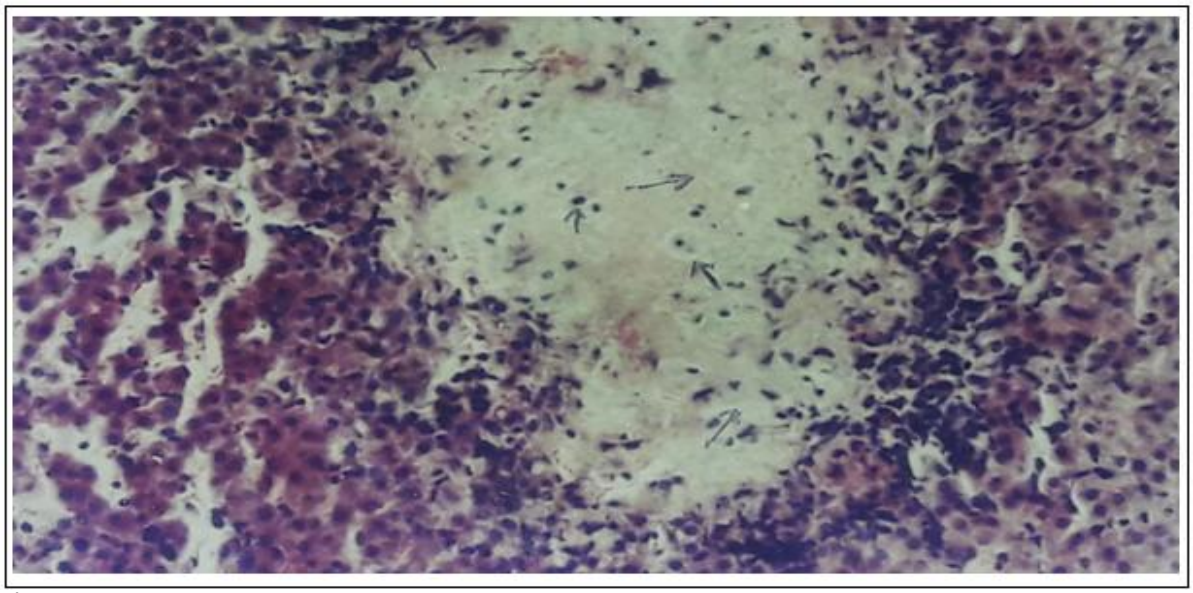

Fig. 3. Histopathological section in liver with caseous necrosis and monocyte, giant cell,and macrophage stained by Hematoxylin and eosin stain(x250).

Biochemical test

Growth on mac-conckey without crystal violet ,urease test ,nitrate reduction ,catalase test ,thiophen -2-carboxylic acid hydroxide(TCH), and sensitivity test for antimycobacterial drug ethambutol(EMB), Isonicotinic hydrazide (INH),Streptomycin(SM), Rimfampicin (RMP),Para- aminosalicylic acid (PAS).(Robert *et al.*,1991).

Histopathological examination

The sample put in (10%) formalin solution and pathological section were staining with heamatoxlin and eosin stain (Luna, 1968).

Results

The results indicated that the percentage of infection in cow carcasses were(2.8% and0.6%)in Al-shula and Al-karkh abattoirs respectively while AL-dura abattoir was found free from infection ,(8.1%) of lymphnode ,(17.6)of lung,(3.8)of livers,(3.5%)of spleen and (25%)for each supramammary lymphnode and uterus were infected with bovine tuberculosis twenty bacterial isolated belonged to mycobacterium species were isolated from carcasses,(17) of them were diagnosed as *M.bovis* according to biochmecial test and sensitivity to drug(Table1,2,3).and results of histopathological examination indicate the positive results in (8.4%) of samples Figs(1,2,3).

Table 1. Isolation rate of *M.bovis* from cow samples.

Samples	Number of samples	Direct macroscopic examination		Positive culture		Positive in histopathology	
Lymph node	384	29	7.6%	31	8.1%	31	8.1%
Lung	85	15	17.6%	15	17.6%	15	17.6%
Liver	80	3	3.8%	3	3.8%	3	3.8%
Spleen	85	3	3.5 %	3	3.5%	3	3.5%
Supramammary L.N	4	1	25%	1	25%	1	25%
Uterus	4	1	25%	1	25%	1	25%
Total	642	52	8%	54	8.4%	54	8.4%

Discussion

The results indicated that the percentage of infection in cow carcasses were(2.8% and0.6%)in AL-shula and AL-karkh abattoirs respectively while AL-dura abattoir was found free from infection while in study of kustandi, (1984) found that in AL-dura abattoir the infection rate was(%9.23) as well as ALazwi *et al.*,(1992) found that the infection rate (%1)in cow, in other study in USA Hernandez *et al.*, (1997)reported that the infection rate (%0.46) in cow and Brown *et al.*,(1998) found the infection was(%0.5)in cow, Mycobacteria were isolated from 15 (68.2%) caseous lesions and eleven isolates were identified as *M. bovis*,in Brazil(Clarice *et al.*,2003) in Mali the infection rate in cattle was 19 (24.1 %) were identified as *M. bovis* (Mamadou *et al.*,2016), in our study infection of lung was (%17.6) and(%8.9) in lymph node and this finding is similar to the study of francis (1971) and other study in Nigeria the prevalence of bovine tuberculosis in slaughtered cattle, three Zonal abattoirs. (8.3%) were confirmed positive for M. bovis, majority of the lesions 54.2% were from lungs. (Sa'idu *et al.*, 2015). Nolwazi *et al.*, (2017) was isolated from lymph nodes (3.1%) had *M. bovis*. and (%3.6) of *M. bovis* in mesenteric lymph node (Hernandez *et al.*, 1998), *M. bovis* was isolated from 3 cattle that had no gross lesions of tuberculosis. One animal had lesions only in the subiliac lymph node, which is not routinely examined during slaughter surveillance.

These findings are important because detection of gross lesions of tuberculosis during inspection of carcasses at slaughter is the primary method for detection of tuberculous cattle and herds in the United States.(Diana *et al* .,1996) Design Typical tuberculous lesions were most pronounced in the lungs and tracheobronchial and mediastinal lymph nodes(Palmer *et al.*,2002). (47.5%) of *M.bovis* isolates were grown from prescapular lymph gland biopsy (Srivastava *et al.*,2008) Abdul Basit *et al.*,(2015)found *M. bovis*(6.5%) in cattle, buffalos, Goats and sheep respectively.

In our study in liver the (%3.8) and (%3.6)in spleen and (%25) in suprammary lymph node and Hernandez *et al.*, (1998) isolated mycobacteria from the same L.N and infection rate, in uterus in our

study was (%25) and Hein and tomosovic (1979)isolated mycobacteria (%1.6) from three sample of uterus in baffalow. the histological examination in our study confirm the infection with mycobacteria and this agree with the result of Brown *et al.*,(1998).and other study lesions manifest typical granulomas with a necrotic center surrounded by inflammatlory cells and a fibrous capsule (Joseph and Kuria 2019).

Table 2. Biochemical test for *M.bovis*.

Number of isolates	Nitrate reduction	Urea hydrolysis	Tween80 hydrolysis	Catalase test	TCH	Type of mycobacteria
1	_	+	_	_	S	*M.bovis*
2	_	+	_	_	S	*M.bovis*
3	_	+	_	_	S	*M.bovis*
4	_	+	_	_	S	*M.bovis*
5	_	+	_	_	S	*M.bovis*
6	_	+	_	_	S	*M.bovis*
7	_	+	_	_	S	*M,bovis*
8	_	+	_	_	S	*M.bovis*
9	_	+	_	_	S	*M.bovis*
10	_	+	_	_	S	*M.bovis*
11	_	+	_	_	S	*M.bovis*
12	_	+	_	_	S	*M.bovis*
13	_	+	_	_	S	*M.bovis*
14	_	+	_	_	S	*M.bovis*
15	_	+	_	_	S	*M.bovis*
16	_	+	_	_	S	*M.bovis*
17	_	+	_	_	S	*M.bovis*

Table 3. Sensitivity of *M.bovis* to antimycobacterial drug.

number	INH	PAS	RMP	STR	ETB	Type of isolate
1	*R*	*S*	*S*	*R*	*R*	*M.bovis*
2	*R*	*S*	*S*	*R*	*R*	*M,bovis*
3	*R*	*S*	*S*	*R*	*R*	*M,bovis*
4	*R*	*S*	*S*	*R*	*R*	*M.bovis*
5	*R*	*S*	*S*	*R*	*R*	*M.bovis*
6	*R*	*S*	*S*	*R*	*R*	*M.bovis*
7	*R*	*S*	*S*	*R*	*R*	*M.bovis*
8	*R*	*S*	*S*	*R*	*R*	*M,bovis*
9	*R*	*S*	*S*	*R*	*R*	*M.bovis*
10	*R*	*S*	*S*	*R*	*R*	*M.bovis*
11	*R*	*S*	*S*	*R*	*R*	*M.bovis*
12	*R*	*S*	*S*	*R*	*R*	*M.bovis*
13	*R*	*S*	*S*	*R*	*R*	*M.bovis*
14	*R*	*S*	*S*	*R*	*R*	*M.bovis*
15	*R*	*S*	*S*	*R*	*R*	*M.bovis*
16	*R*	*S*	*S*	*R*	*R*	*M.bovis*
17	*R*	*S*	*S*	*R*	*R*	*M.bovis*

Acknowledgements

The authors are grateful to the staff of *Institute of Chest Disease laboratory* and all workers in baghdad abattoris who assisted in this study.

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