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SOME INVESTIGATIONS ON BRUCELLA AND PSOROPTES MITES INFECTIONS AMONG BARKI SHEEP FLOCKS

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ABSTRACT

Two groups of Barki ewes in a private farm, aged 3-5 years, non-vaccinated against brucella infection and had history of reproductive disturbances, were examined for brucella infection by different serological tests. The first group (412 ewes) has skin lesions. Meanwhile, the second group (375 ewes) was clinically free from skin lesions. Blood samples were taken from both groups of ewes (4 times within one year) for serodiagnosis of brucella infection using tube agglutination test (TAT), Rose Bengal plate test (RBPT) and Rivanol (Riv. T) tests, in addition to collection of skin scraping samples from ewes suffering from skin lesions for detection of scab mites infection, at the time of blood sampling. The positive reactors of brucellosis by different serological tests were recorded in both groups. It was found also that 30.77% of brucella positive ewes had psoroptes mites infection. The suggested role of mites in opening portals of brucella infection via skin was mentioned. Guinea pigs used to assess this possible suggestive route and a slight rise in titre between scabbied brucella infected group than in brucella infected ones were observed. Treatment of scab mites infection after each examination was done using different specific drugs.

INTRODUCTION

Brucellosis is a highly contagious reproductive disease. There were no animals either domesticated or tame, even man have escaped the ravage and burden of brucellosis **(Mayer, 1974)**. Brucella infection causes abortion, weak born lambs, death of lambs, in addition to mastitis in infected ewes **(Fielden, 1986)**. Sheep brucellosis is usually caused by Brucella melitensis but occasionally resulted from Brucella abortus infection. It is an acute or chronic disease characterized by septicaemia followed by localization of infection in the lymph nodes and genital organs **(Fielden, 1986 and Marin et al., 1999)**. In Egypt, brucellosis in sheep was investigated by several authors **(Ismail, 1971; Shawkat, 1973; El-Bauomey, 1989)**. Meanwhile, sheep scab is

highly contagious and extremely unpleasant disease. It is usually caused by Psoroptes ovis **(hogg and Lohane, 1999)**. The mites live on the skin surface and pierce the epidermis to feed on lymph and serum exudates. Small pustules and hard yellow crusts develop at the puncture sites **(Asp and Tauni, 1988)**. There is extensive loss of weight gain, fleece and the exposed skin is thickened with raw patches **(Fatimah et al., 1994 and Nagal et al., 1995)**. The lesions observed are the result of combination of factors: damage caused by the mite during feeding, the hosts immune response to the mites, secondary bacterial infection and self inflicted trauma in response to severe irritation, weakness, loss of condition and death may occur (Mahmoud, 1960 and Bill and Harry, 1984). In Egypt, sheep mites were investigated by **Ashmawy (1977)**. Various studies were carried out on the role of bacteria infection and its association to sheep scab mites **Hogg and Lehane (1999) and Mahanta et al. (1997)**. Moreover, few studies were carried out on the role of blood sucking and biting insects (several kinds of ticks and culex spp.) as vectors of brucella infection either mechanically or biologically **(Mayer, 1977)** but no available data about such relation between sheep scab mites and brucella microbes transmission were found. So that, this study aimed to investigate the prevalence of brucellosis among scabbied sheep and the suggested role played by mites in transmission and persistence of brucella infection among such scabbed flocks.

MATERIALS AND METHODS

1. Animals.

Two groups of Barki ewes in a private farm, aged from 3-5 years, non vaccinated against brucella infection, and had a history of reproductive disturbances (abortions, weak born lambs and mastitis), were examined for brucella infection by some serological tests. The first group (412 ewes) had skin lesions on ears, nose, mouth and neck that may be extended to the back in some ewes. The second group (375 ewes) was clinically free from skin lesions.

2. Blood samples.

Blood samples were collected from the examined ewes, four times within a year, and sera were examined for serodiagnosis of brucella infection using the Tube Agglutination test (TAT), Rose Bengal plate (RBPT) and Rivanol (Riv. T) tests and the results were recorded as described by **Alton et al. (1988)**. The positive reaction was determined at a titer of ≥1/40 for TAT, ≥1/20 for Riv. T and agglutination reaction within one minute in RBPT.

3. Skin examination.

Skin lesions of suspected scabbed ewes were moistened with glycerine and scraped samples

were collected in clean petri dishes at the time of blood sampling. The samples were subjected to either directly or after heating not boiling with 10% potassium hydroxide (Alkali maceration technique) to microscopical examination using the methods adopted by **Tarry (1974) and Menazi (1976)**. The mites were identified according to **Ashmawy (1977) and Bill and harry (1984)**.

4. Bacteriological examination.

This was performed according to **Alton et al. (1988)**.

- Culture of specimens :

samples of lymph nodes, genital organs, spleen were taken from slaughtered ewes which gave positive serological tests. These samples were cultured on brucella medium base (Oxoid Co., USA) with brucella selective supplement (each vial contains: Polymixin B, Bacitriau, Cycloheximide, Nalidixic acid, Nystatin and Vancomycin). This media prevent growth of any bacteria or fungi rather than brucella. These plates were incubated at 37°C under 5-10% CO2 tension, periodically examined for suspected growth after 4 days and periodically examined till about 35 days before discarded as negative culture. Suspected brucella colony was confirmed by staining smears **(Gram and Modified Ziehl Neelsen stains).**

During this investigation, all positive ewes were segregated and slaughtered according to the test and slaughter programme applied by ministry of agriculture, Egypt.

5. Treatment :

Moreover, all scabbed sheep were treated by injection with Ivomec **(Merk & Sharp & Dohme, NJ, USA)** in a dose of 1 ml/50 kg body weight at first examination, then by local 10% sulphur ointment on non recovered lesions during the second examination and in the third testing by injection of Genesis (Abameetin, Ancare NewZealand Ltd. No. 247480) in a dose of 1 ml/50 kg body weight.

Experimental infection of guinea pigs:

An experiment was carried out on 15 guinea pigs in equal three groups for clarifying the possible role of sheep scab as an aid in transmission of brueella infection. The first group was infected by both positive sheep scabies scraping and Br. melitensis 16 M strain in a plastic cell adhered and bandaged on the back of guinea pigs.

The second group was infected by Br. melitensis 16 M strain only in a plastic cell adhered and bandaged on the back of guinea pigs. The third group was served as control negative. The clinical manifestation were recorded and the guinea pigs groups were scarified after 6 weeks and PM findings were registered and the blood was tested for brucellosis by TAT, RBPT and Riv. Tests.

RESULTS & DISCUSSION

In this study the prevalence of brucellosis and sheep scab mites (Psoroptes ovis) were illustrated in Table (1).

In the first examination, 78 (18.93%) of 412 ewes were positive in all studied serological tests for brucellosis, while 134 (32.52%) of them were positive for sheep scabs. In the second examination the percentages of brucellosis and scabs in 334 ewes were 12.27% and 10.18% respectively. In the third examination, the percentages were 1.02% and 0.68% in brucellosis and sheep scabs respectively. While in the fourth examination were 0.34% in brucellosis and negative results in the sheep scabs. The number of ewes had both infections were 24 (5.83%) in the first examination while was 12 (3.59%) in the second examination. However, in the third and fourth examinations were negative results.

Concerning the prevalence of brucellosis among examined ewes with different serological tests, the first group (412 ewes), the incidence of brucellosis was 17.47, 18.93 and 16.99 with RBPT, TAT and Rivanol tests, respectively. In the second examination, the percentages of brucella infection were 11.37, 12.27 and 11.67 with RBPT, TAT and Rivanol tests, respectively. In the third examination these percentages were 0.68, 1.02 and 0.34 with RBPT, TAT and Rivanol tests, respectively. The fourth examination showed only one positive case with TAT (0.34%). Concerning second group (375 ewes), the incidence of brucellosis was 5.06%, 5.86% and 4.8% with RBPT, TAT and Rivanol tests in the first examination.

The second examination was 0.84, 1.13 and 0.84 with RBPT, TAT and Rivanol tests, respectively. Meanwhile, it was 0.28, 0.57 and 0.28% with RBPT, TAT and Rivanol respectively. On the other hand, the incidence of brucellosis was 0% in the last examination. The examined second group of ewes proved to be negative to Psoroptes ovis infection during this study. Concerning the bacteriological examinations, total number of 123 slaughtered ewes during this study, which proved brucella positive tests, was subjected to bacteriological studies. The samples from lymph nodes, genital organs and spleen were cultured. Only two isolates of Brucella spp. were obtained from lymph nodes and uterus of infected ewes.

In guinea pigs, the first group showed all the clinical picture of scabies within one week and the blood sera showed positive result for brucellosis. The clinical symptoms of scabs in guinea pigs were developed as pastules at the site of infection, which ruptured and formed yellow crusts, severe irritation and skin was thickened and damaged. Moreover, crying and self-inflicted trauma in response to severe irritation was noticed along the period of study. The skin lesion was spread and extended over the back, abdomen, legs, ears and nose of guinea pigs.

The second group showed positive results of brucellosis. The PM findings of brucellosis in the

first and second groups were orchitis, enlarged lymph nodes, liver and spleen. The control group were negative for brucellosis and psoroptes mites infection.

Different serodiagnostic tests including tube agglutination, Rose Bengal plate and Rivanol tests were performed for diagnosis of brucellosis among sheep.

In the present study, the prevalence of brucella infection, in the first group, by the studied serological tests, declined from 18.93% to 12.28% then to 1.02% and finally 0.34%; in the first, second and third examinations and revealed negative result in the fourth examination. However, the seropositive incidences of brucella infection, in the second group, were 5.87%, 1.13%, 0.57% and 0% in the first, second, third and fourth examinations respectively.

The decline in the incidence of brucella positive reactors in our study resulted from the application of the periodical testing, slaughtering of positive animals, in addition to the application of the hygienic measures according to the rules for control of brucellosis, Ministry of Agriculture, Egypt.

In Egypt, the incidence of brucellosis was ranged from 2.68% to 18.5% **(Ismail, 1971; Shawkat, 1973 and El-Bauomey, 1989)**. The variations between our results and those obtained by other authors might be attributed to breed, sex, age locality, management and the stage of infection, in addition to the physiological and immune status of animals **(Tizard, 1987; Nada, 1990; Nada et al., 1992 and Ghazy et al., 2000)**.

The incidence of sheep scabies in the present study also declined from 32.52% to 0.68% along the four examinations. In this respect **Soll et al. (1992)** reported an incidence varied from 0.4% to 99% in four treated flocks previously affected with scabies (Psoroptes ovis). Moreover, **Marin et al , (1999)** found that the sero prevalence of brucella infection among sheep in four examinations were 80.9%, 27.7%, 66.7% and 78.5%.

The clinical symptoms and the presence of Psoroptes ovis mites in the current study was similar to findings of **Mahmoud (1960), Bill and Harry (1984) and Hogg and Lohane (1999)**.

Detection of Psoroptes ovis mites infection in 30.77% of the brucella seropositive ewes in the first and in 29.27% in the second examinations, payed the attention to suggest a probable relation between both infections. In this respect, various studies mentioned by **Mayer (1977)** described the role of insects (several kinds of ticks, culex spp. and flies) as transmitters for brucella microbes either mechanically or biologically to the animals.

Cotton et al. (1933), Cotton (1934), Corbel (1989), Refai and Kopec (1989) and Alton (1990) indicated the significant role of skin as portal for entry of brucella organisms to animal body. In our investigation the mites might play a role in the entry of brucella microorganism

through the brokened skin and increasing the incidence of brucella infection among the first group in comparison to results of second group. In addition, the scabies had an immunosuppressive effect on the host that might increase the susceptibility of the animal to brucella infection **(Tizard, 1987)**.

It was noticed that a considerable percentages of both infections were present in the second examination in spite of application of control measures following first examination. Such finding may be due to the presence of various stages of infection and latency. The results of the guinea pig experimental groups showed the appearance of clinical lesions of mites scab in the first group from the 1st to the 6th week which is similar to finding of **Wilson et al. (1977).** Meanwhile, the second group showed slight decrease in degrees of reaction of brucella antibodies than in the first group using the serological tests. This result coordinates with findings of **Cotton et al. (1934)**, who stated that brucella infection could be transmitted through broken, abraded and even a healthy skin of animals.

In conclusion, the Psoroptes ovis mites in sheep flock can affect the prevalence of brucellosis among them. So that in control programmes of brucellosis in sheep, the control of psoroptes mites infection must be taken in consideration and the scabbied cases should subjected to isolation, treatment and examination for brucellosis before readdition to the flock.

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Table (1). The prevalence of brucella and psoroptes mites infections among examined Barki ewes.

Examined groups	First group of ewes	Brucella positive ewes		Psoroptes mites infected ewes		Ewes had both infections		Ewes had both infections/total seropositive ewes	
		No.	Percentage %	No.	Percentage %	No.	Percentage %	No.	Percentage %
First exam.	412	78	18.93	134	32.52	24	5.83	24/78	30.77
Second exam.	334	41	12.28	34	10.18	12	3.59	12/41	29.27
Third exam	293	3	1.02	2	0.68	-	-		
Fourth exam.	290	1	0.34	-	-	-	-		

Table (2). The prevalence of brucellosis among examined Barki ewes with different serological tests.

Examined groups	1st examination						2nd examination						3rd examination						4th examination					
	RBPT (Positive)		TAT (Positive)		Rivanol (Positive)		RBPT (Positive)		TAT (Positive)		Rivanol (Positive)		RBPT (Positive)		TAT (Positive)		Rivanol (Positive)		RBPT (Positive)		TAT (Positive)		Rivanol (Positive)	
	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%
First group	72	17.43	78	18.93	70	16.99	38	11.37	41	12.27	39	11.67	2	0.68	3	1.02	1	0.34	-	-	1	0.34	-	-
Total number	412						334						293						290					
Second group	19	5.06	22	5.86	18	4.8	3	0.84	4	10.13	3	0.84	1	0.28	2	0.57	1	0.28	-	-	-	-	-	-
*Total number	375						353						349						347					

* The variation of the number of examined ewes was due to the application of test and slaughter program of brucellosis.

RBPT = Rose Bengal plate test.

TAT = Tube agglutination test.

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الملخص العربى

بعض الاستيضاحات على مرض البروسيلا والإصابة بحلم الجرب فى قطعان الأغنام البرقى

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تم فحص مجموعتين من إناث الأغنام البرقى - التى تتراوح أعمارها من ثلاثة إلى خمسة أعوام تربى فى مزرعة خاصة - وذلك لتشخيص مرض البروسيلا باستخدام الطرق السيرولوچية المختلفة وكانت هذه الأغنام غير محصنة بلقاح البروسيلا ولديها بعض الإضطرابات التناسلية، المجموعة الأولى والمكونة من ٤١٢ نعجة تعانى من أعراض جلدية ظاهرية والمجموعة الثانية مكونة من ٣٧٥ نعجة خالية من أى أمراض جلدية تم أخذ ٤ عينات دم دورية خلال عام واحد من هذه الأغنام وذلك للتشخيص السيرولوچى لمرض البروسيلا باستخدام إختبار التلزن الأنبوبى (TAT) واختبار الروزبينجال (RBPT) واختبار الريفانول (Rivanol-T) بالإضافة إلى تجميع مسحات جلدية من الأغنام التى تعانى من الأعراض الجلدية وذلك بغرض العزل والتعرف على حلم الجرب فى نفس توقيت أخذ عينات الدم للتشخيص السيرولوچى لهذه الأغنام، وأظهرت النتائج أن ٧٧ر٣٠٪ من النعاج الإيجابية لمرض البروسيلا مصابة بحلم الجرب (Psoroptes ovis).

وقد تم كذلك إستخدام خنازير غينيا فى دراسة الدور المحتمل الذى يمكن أن تسببه الإصابة بحلم الجرب كعامل مساعد للإصابة بمرض البروسيلا.

وقد تم أيضاً دراسة مقارنة تأثير الإصابة بحلم الجرب على مستوى الأجسام المناعية للبروسيلا فى كلا المجموعتين.

تم علاج الإصابة بالجرب فى هذه الأغنام بعد كل فحص باستخدام العلاجات المتخصصة المختلفة.