Al-Qadisiyah Journal of Veterinary Medicine Sciences
(P-ISSN 1818-5746/ E-ISSN 2313-4429)
www.qu.edu.iq/journalvm

Research article

Serological investigation of Brucella spp. and *Toxoplasma gondii* in horses by using rose Bengal and toxoplasma latex tests in Dhi-Qar Province, Iraq

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(Received 25/1/2017, Accepted 24/4/2017)

Abstract

To detect the infection by Brucella spp. and Toxoplasma gondii. This study was conducted using a screening Rose Bengal and Toxoplasma latex tests on 100 blood samples collected from horses (18 male +82 female) average of age (1month-25 years) living in different areas in the districts of Dhi-Qar province in southern Iraq, where results showed a spread of Brucella spp. antibodies germs by 8% and the spread of the parasite Toxoplasma gondii antibodies 21%. This study showed the spread of infection by bacteria Brucella and Toxoplasma parasite in horses in Dhi-Qar. Province-southern Iraq.

Key words: Brucella, horses, latex, toxoplasma, serological investigation.

Introduction:

Brucellosis is one of most worldwide zoonosis' diseases. Human infection is usually by animal contact. also transmitted to human due to occupational exposure and it's cause significant losses in farm animals through abortions, arthritis and fistula manifest immediately add to the cost of treatment and control to influence male and female fertility (1) *Brucella spp*.is Gram negative, an intracellular pathogen. There are six known species of the genus Brucella bacterium *is B. abortus, B. melitensis, B. suis, B. ovis, B. canis,* and *B. neotomae.* (2). In Iraq there was a study on Brucellosis in horses and its confirmed presence of Brucella antibodies by using the ELISA test. (3) Usually the horses infected by *Toxoplasma gondii* with sub clinical condition. This Parasite lives inside cells and spreads to infects many tissues, and it's also infected large number of mammals and birds species and it's an important disease because it's globally and zoonosis' disease in addition to it's importance in veterinary medicine (4). This parasite infected all warm-blooded animals and estimates the number of people around the world infected with Toxoplasma about one third of the Earth's population (5). The important clinical sings are meningitis and abortion in addition to fever and unbalanced motion, but often without clinical symptoms (6). This study was designed to investigate of brucellosis and toxoplasmosis in horses in the province of Dhi- Qar, which cause abortion in mares by investigating of antibodies which caused by the infection and studying of some epidemiology effects on horses and how to prevent prevention and treatment, because this horses lives the different ruminants which were recorded infected with both diseases and some mares exposure to arthritis and abortion so must recorded brucellosis and toxoplasmosis in differential diagnosis list.

Materials and Methods:

One hundred blood samples were collected randomly from horses (18 males +82 females) average of age(1month-25 years) living in different areas in the districts(Al-nassiryia, Al-batha, Al-aslah, A-refaie and Al-shatrah) of Dhi- Qar province

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some lived with and some with out and all farms were open farms and they divided to two age A-(1-6 months) last age of weaning 10 heads B-(7Months-25 years) 90 heads. Blood sample was collected from jugular vein of each animal without EDTA and allowed it is to coagulant and spirited the serum. The serum was stored at -20˚C until use. by using Rose Bengal(Cromatest®) test according to procedure (after 4min. from mix one drop of serum and one drop of antigen= rough granules (agglutination) mean positive) and Toxoplasma latex (Spinreact®) tests according to procedure (after 4min. from mix one drop of serum and one drop of antigen(rough granules (agglutination) mean positive). The (SPSS Version 17, L.S.D., ($P \leq 0.05$) program was used to analyze the results.

Results:

Results showed a spread of *Brucella spp*. antibodies in 8% and the spread of the parasite *Toxoplasma gondii* antibodies 21% from 100 samples. There were a significant variations ($p>0.05$) between male and female. Males were significantly lower ($P<0.05$) than females.as in table1. Results showed high levels of antibodies to each pathogen in females than males.

Table (1) antibodies of Brucella spp. and Toxoplasma gondii according to sex.

sex	No.	*Brucella spp.*				*Toxoplasma gondii*			
		+ve	%	mean	SD	+ve	%	mean	SD
Male	18	1	5.55	0.055 [a]	0.236	1	5.55	0.055 [a]	0.236
Female	82	7	8.53	0.085 [b]	0.281	20	24.39	0.244 [b]	0.432
Total	100	8				21			

([a, b])denotes significant different at $P \leq 0.05$

There were significant different between age groups. Level of antibodies of *Toxoplasma gondii* was higher in group 7Months-25 years than group 1-6 months but there were 20 samples positive in group (7Months-25 years) but one sample is positive in group 1-6 months. Level of antibodies of *Brucella spp*. was higher in-group 1-6 months than group7Months-25 years as in table 2.

Table (2) antibodies of Brucella spp. and Toxoplasma gondii according to age.

Age	No.	*Brucella spp.*					*Toxoplasma gondii*				
		+ve	mean	SD	female	male	+ve	mean	SD	female	male
1-6 months	10	1	0.1 [a]	0.316	1	0	3	0.3	0.483	2	1
7Months-25 years	90	7	0.077 [b]	0.269	6	1	18	0.2	0.402	18	0
Total	100	8			7	1	21			20	1

([a, b])denotes significant different at $P \leq 0.05$

The results showed also a significant different between Toxoplasmosis and Brucellosis. Toxoplasmosis is higher than Brucellosis as in table 3.

Table (3): Significant different between Toxoplasmosis and Brucellosis

Disease	No.	+ve	%	mean	SD
Brucellosis	100	8	8	0.08 [a]	0.273
Toxoplasmosis	100	21	21	0.21 [b]	0.409

([a, b])denotes significant different at $P \leq 0.05$

Discussion:

In this study, two serological tests were used based on the agglutination on the glass slide between antibodies and antigens. They can be administered in all laboratories and require simple and available equipment. Rose Bengal and Toxoplasma latex tests are rapid tests but the results given the same distribution of Brucellosis and Toxoplasmosis which the female more prevalence to Brucellosis and Toxoplasmosis (2) conclusion the Sensitivity of Rose Bengal is 89.9% when indirect ELISA test is 96.8% and complement fixation is 94%.also specificity of Rose Bengal is 84.5%, indirect

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ELISA 96.3% and complement fixation is 88.5% (2). (7) in Nigeria recorded 7.8% Prevalence of Brucella antibodies in horses (7) and it near to our result, when (8) recorded 16% in Nigeria by use Rose Bengal test. In Southeast Turkey (1) recorded 13.68 %, also in Turkey (9) reported 13.29%.in north of Iran the result of Prevalence was 2.5% (10).when in mid of Iran reported 0.05% also by use Rose Bengal test (11). In Iraq in Mosul city there were no recorded of brucella antibodies in equine by using Rose Bengal test (3) so the study is first recorded of brucella antibodies in horses by using Rose Bengal test in Iraq. In addition, brucella antibodies are randomly found horses according to sex and age, and founded *Brucella spp*. antibodies in young foals came from maternal immunity but the age after weaning came from infection (16). The results of our study recorded prevalence of antibodies to *Toxoplasma gondii* in horses in Dhi-Qar and it's same recorded in northwestern China 31.4% (5),when in japan recorded 4.24%(12) and in Pakistan 23.5% (13) and all recent studies by using latex agglutination test. In Iraq in Baghdad city reported 67.39% (14) and in Mosul city reported 72.2% (15) also by using latex agglutination test. The number of horses in Dhi-Qar was decrease between the years 2000-2010 then increased after that. Their numbers increased as decorative animals living between ruminants, dogs and cats, where abortion was recorded in all ruminants in the province. *Toxoplasma gondii* antibodies in young foals may be came from maternal immunity and the type of breeding (14). The significant increase of Toxoplasmosis against Brucellosis give an indicator that prevalence *Toxoplasma gondii* more higher than *Brucella Spp.*, so the risk of abortion in mare from *Toxoplasma gondii* more higher than *Brucella Spp*., and this same prevalence *of Toxoplasma gondii* in Mosul city witch wasn't recorded *Brucella Spp* antibodies in horses so this is the first study in Iraq record antibodies of *Brucella Spp* by using rose Bengal test (3) (15). This difference may be due to the fact that the transport of *Toxoplasma gondii* is carried out by rodents and cats which are many and close to horses and may reach the horses through water and food contaminated with parasite. There is a significant difference between the incidence of males and females and explained this because of the tendency of *Brucella Spp* to the uterus as well as parasite *Toxoplasma gondii* during pregnancy and the uterus is the main goal of both pathogens .

Conclusions:

This study reported of antibodies of *Brucella spp*. and *Toxoplasma gondii* in horses in Dhi-Qar province in different sexes and ages. Rose Bengal test and latex agglutination test are rapid testes but give good results and can use them in survey programs to Toxoplasmosis and Brucellosis in horses also can use them to detect the response to treatment. The prevalence of *Toxoplasma gondii* is higher than *Brucella spp*.in horses.

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