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Parasitological, biochemical and hematological studies in equines infected with piroplasmosis

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Abstract

Three hundred five blood samples were collected from horses individually aged from 2 to 8 years old of both sex, from different locality in Cairo and Giza governorates of Egypt. The results showed that the prevalence rate of *Theileria equi* in examined horses was 10.4%, 16.1% and 12.5% by using microscopic examination, cELISA and PCR, respectively. *Babesia caballi* were not detected with three diagnostic methods used. Clinical, hematological and biochemical studies were evaluated in examined horses. Significant changes in hematological and some biochemical parameters were observed within infected animals. The obtained results high lights the importance of PCR analysis for *T.equi* in clinically healthy carrier horses.

Key words: *Equine piroplasmosis, Theileria equi, cELISA, PCR.*

Introduction

Equine piroplasmosis is a tick-borne haemoprotozo an disease of equine that is caused by two species of apicomplexan protozoa, *Babesia caballi* and *Theileria equi* (*Babesia equi*) **(Mehlhorn and Schein1998)**. *Theileria equi* was previously designated as *Babesia equi* but compelling evolutionary, morphological, biochmemical, genetic evidence and finding an extra-erythrocytic stage within equine-peripheral blood mononuclear cells PBMC supports its reclassification as a *Theileria* **(OIE 2009 & 2014).**

Theileria equi (B. equi) was considered more virulent than *B. caballi* that caused a more persistent fever and anemia **(Radostitis *et al.*, 2000)** because infection in carrier animals can be sterilised by suitable drug treatment, whereas *T. equi* infection may be suppressed by chemotherapy but didn't eliminated **(De Waal and Van Heerden 2004).**

Equine theileriosis, an OIE list disease, caused by *Theileria equi* a small piroplasm, that is very pathogenic in horses. It is transmitted by various genera of ixodid ticks which include *Dermacentor spp., Hyaloma spp. and Rhipicephalus spp.* **(Urquhart *et al.*, 1996).** The disease is endemic in tropical and subtropical areas **(Hailat *et al.*, 1997).** Equine piroplasmosis has caused important economic losses in the horse industry, being a serious threat to the horse raising industry and international movement of horses **(Friedhoff *et al.*, 1990).**

The clinical signs of piroplasmosis were variable and often nonspecific **(Bashiruddin *et al.*, 1999).** Equine theileriosis occur in acute, subacute and chronic form and hence the clinical manifestation is variable and could manifest as fever, anemia, jaundice and haemglobinuria in acute form and sometimes in chronic infection icterus and oedema of the ventral abdomen may be seen **(Urquhart *et al.*, 1996)**. In chronic and subclinical cases, despite the absence of obvious clinical signs, infected horses can carry the piroplasms in their blood for many years. Carriers of the infection are usually asymptomatic and are responsible for the

propagation and maintenance of the infection.

Babesiosis could be diagnosed by a number of different methods, including Giemsa-stained blood smears; enzyme- linked immunosorbent assays (ELISA) and polymerase chain reaction (PCR) **(Bose *et al*., 1995).** The examination of the stained blood smears was simple but insufficient for accurate detection and identification of *B. caballi* and *T. equi* during mixed infections and in particular in carrier cases or subclinical infections with low parasitemia **(Krause 2003** and **Quintao and Ribeiro, 2003)**. Serological methods detect both past and current infections simultaneously and thus used for screening the equids in international trades **(OIE 2008).** The goal of serological testing is to identify seropositive animals whose migration is restricted **(Wise *et al.* 2013).**

Among all the serological tests, cELISA is the test of choice and is recommended by the World Organization for Animal Health **(OIE 2008).** Moreover, other studies have shown that PCR is highly sensitive and specific test for detection of *T. equi*, even in chonically infected horses **(Bashiruddin *et al*. 1999, Farah *et al*. 2003, Rampersad *et al.* 2003 and Ribeiro *et al*. 2013a)**

Hematological and sero-biochemical alterations are indicators to disease severity and are considered to be good tools for the diagnosis and prognosis for effective therapy**.**

The aim of the present work was to (1) study the prevalence of *T. equi* and *B. caballi* in the collected blood samples from examined horses to (2) compare the efficacy of conventional diagnostic methods (microscopic examination of stained blood smears, serological method (cELISA)) and to polymerase chain reaction (PCR) for the diagnosis of equine piroplasmosis with samples from spontaneously infected horses and (3) through light on diagnosing of the changes of blood picture and blood biochemical components associated with equintheileriosis in infected horses**.**

Materials and Methods

1- Animals:

A total number of 305 horses aged from 2 to 8 years old of both sex, from different locality in Cairo and Giza governorates of Egypt. This study was conducted all over one year extended from January till the December 2014. Investigated horses were clinically and laboratory examined for the presence of *T. equi* and *B. caballi* infection. All the animals were clinically healthy at the moment of sample collection except 57 cases of horses have had contact with ticks, followed by clinical signs manifested by: walking disorders, fever, weakness, exhaustion, anaemia, jaundice and haemoglobinuria.

2- Blood samples:

Blood samples were collected by *jugular venipuncture* with a vacutainer needle at presentation. Blood samples were collected in 2 vacuum–sealed sterile tubes. The first tube containing Ethylene diamine tetra-acetic acid (EDTA) at 1mg/ml concentration as anticoagulant, this part of blood sample is used for microscopical blood smears examination, hematological analysis and DNA extraction for PCR assay. The second tube contained no anticoagulant for serum separation and stored at -20° C until used for ELISA and biochemical analysis.

Parasitological examination:

Blood smears were prepared at the time of sampling and stained with Giemsa stain then microscopically examined for *T. equi* and *B. caballi* evidence **(Zweygarth *et al*., 2002).** The horse was considered Theileria free after three negative successive blood smears.

Haematological tests:

It were performed according to **Feldman *et al*., (2000)** including, the estimation of white differential count, total leucocytic count (TLC), red blood cell (RBCs) count and the hemoglobin concentration (Hb).

Biochemical tests:
Serum samples were tested spectrophotometrically to determine the following parameters; alanin aminotransferase (ALT), aspartate aminotransferase (AST) activities according to **Reitman and Frankel (1957)**. Creatinine was measured according to **Fabiny and Ertingshausen (1971).** Blood urea was measured according to **Tabacco *et al.* (1979).** Total proteinswere determined according to **Gornal*et al*. (1949).** Albumin was measured according to **Doumas *et al*. (1971)** and Protein electrophoresis was performed according to **Keyser and Watkins (1972).**

Serological tests:
Competitive Enzyme- Linked Immunosorbent Assays (cELISA):
The competitive enzyme linked immunosorbant assay (c-ELISA) to detect antibodies against *T. equi* and *B. caballi* in tested sera by using a commercially available kit distributed by (VMRD, USA). The test was performed following the instructions of manufacturer **(USDA, 2005).** The mean optical density (OD) at 620 nm was determined for all wells using a microplate reader (Expert plus UV-G020 151-Austria). A strong color development indicated little or no inhibition of primary monoclonal antibody binding and therefore the absence of *T. equi* or *B. caballi* antibodies in the sample.

Molecular analysis:
DNA extraction:
Equine ETDA blood samples were used for DNA extraction, which was accomplished using a commercial kit (Gen Jet Genomic DNA purification kit K0271, Fermentas) in accordance with the manufacturer's instructions

b- PCR technique (Danielle *et al*., 2011):
Primers designed based on the sequences of the gene 18SrRNA of *Theileria equi* (Teq) was used. The *T. equi* sequences were TeqF forward 5'-ATGCCCTTCAGTTCG- 3' and TeqR reverse 5'-ACCTCCCTGTGTCAGGATTG-3'. The *B. caballi* sequences were BcF forward5'-GAGGGACTTTGGGTCATAA-3'and BcR reverse q5'-GGTACTCGATCGGTAGGA-3'. PCR reaction was performed in an Gradient Thermal cycler(1000 S Thermal cycler Bio-RAD USA). The reaction mixture (total volume of 50 µl) was 25 µl Dream green PCR Mix (DreamTaq Green PCR Master Mix (2X) Ferment as Company, cat., No. K1080, USA.), 5µl target DNA, 2µl of each primers (containing 10 p mole/µl) and the mixture was completed by sterile D. W. to 50µl .The reaction was carried out as 94°C for 2 minutes, followed by 35 cycles of 94°C for 30 sec, 60°C for 30 sec and 72°C for 30 sec, with a final extension of 72°C for 5 minutes. The products from the PCR were subjected to a horizontal electrophoresis run on 1.5% agarose gel, in Tris-borate EDTA (TBE) buffer, together with the 100 bp molecular weight marker (Thermo). The results were viewed in a trans - illuminator and were documented by photographing by canon camera.

Statistical analysis: Both hematological and biochemical results were statistically evaluated using T. test **(Petrie and Watson 1999)**.

Results
Clinical examination:
The clinical investigation of examined horses revealed that all animals were apparently health except 57 horses (18.7%) suffered from fever, anorexia, weakness and icterus.

Parasitological finding:
By examination of the Giemsa stained blood smears from 305 investigated horses out 32 horses (10.4%) were infected with *T. equi*. Different intra-erythrocytic stages of *T. equi* were detected as spherical, oval, ring form (Fig. 1A&B),Four *T. equi* merozoites in characteristic arrangement called Maltese cross (Fig.1C). The schizonts were detected in the lymphocytes with two forms, macroschizonts and microschizonts. (Fig.1D). No *B. caballi* was detected on any smears.

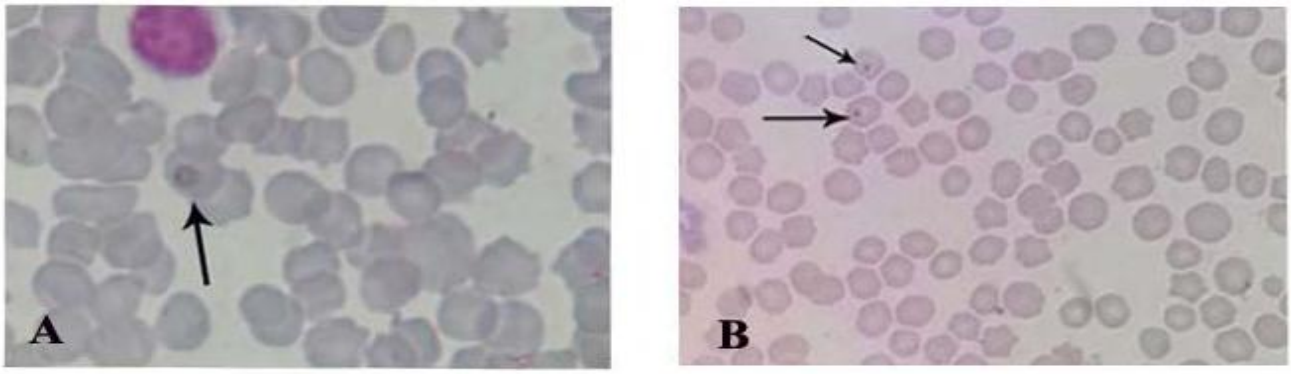
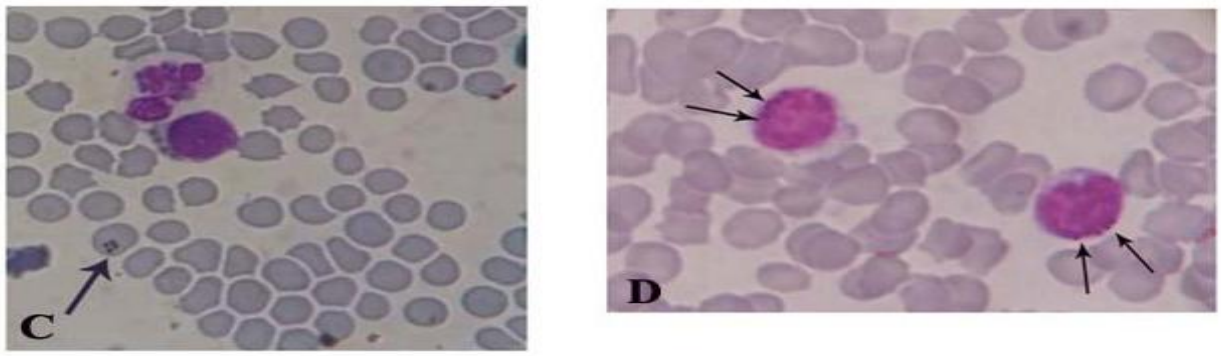


Fig. 1
A-Intra-erythocytic small piroplasms of *T. equi* forming a ring form.
B-Blood smear showing erythrocytes abnormalities with *T. equi* merozoites.



C-Intra-erythocytic small piroplasms of *T. equi* forming a Maltese cross.
D-Intra-lymphocytic schizont stages of *T. equi*.

Haematological findings:

Haematololgical aspects were studied in an attempt to provide information on the disease status. The mean values of total RBCs count and Hb concentration (Table 1) showed asignificant decrease in diseased animal group when compared to apparent healthy control animal group. In the present study a significant increase in (TLC) total leuckocytic count were observed in infected animal groups (Table 1).

Serum biochemistry profile:

Results of biochemical changes indicated significant decrease in total proteins in diseased animal group, while there was noticed a significant increase in gamma globulin fraction value (Table 2).

Concern the liver enzymes (Table 3) reflect there were significant increases in the serum hepatic enzymes (ALT and AST), whereas increases of serum Creatinin and Urea values above its normal ranges in diseased horses group compared to control group.

Table (1). Changes in mean values of hematological parameters of diseased animal group and in comparison to control healthy animal group.

Exam. horses	Hb (g/dl)	RBCs ($x10^6/mm^3$)	WBCs ($x10^3/mm^3$)	neutrophil (%)	Lymphcytes (%)	Monocytes (%)	Eosinophil (%)	Basophil (%)
Control N = 3	12.9 ± 0.2	4.795 ±0.045	6.925 ± 225	50.5 ± 2.5	42±2	4.5±0.5	2.5 ± 0.5	0.5 ± 0.5
Diseased N = 6	10.44*** ± 2.46	3.672** ± 1.23	9.870 ± 294	42.4*** ± 0.67	44.6*** ± 4.6	3.8*** ± 0.7	8.2*** ±2.7	0.4 ± 0.1

*Significant at $p< 0.05$; Hb conc., Hemoglobin concentration; RBCs, Red blood cells; WBCs, white blood cells count. g/dl, gram per deciliter, mm^3 cubic millimeter.

Table (2). Changes in mean values of biochemical parameters (Total protein and different serum protein fractions) of diseased animal group in comparison to control healthy animal group.

Exam. Horses	Total protein (g/dl)	Albumen (g/dl)	Globulin (g/dl)					
			α1	α2	β1	β2	ʁ1	ʁ2
Control N=3	6.88 ± 0.08	2.87 ±0.06	0.726 ± 0.037	0.493 ± 0.017	0.676± 0.043	0.616 ± 0.035	0.953 ± 0.014	0.543 ± 0.012
Diseased N = 6	5.91*** ± 0.072	2.066 ± 0.038	0.713 ± 0.025	0.435 ± 0.321	0.578 ± 0.026	0.521 ± 0.04	1.015 ±0.021	0.603* ± 0.024

g/dl, gram per deciliter

Table (3). Changes in mean values of biochemical parameters of diseased animal group and in comparison to control healthy animal group.

Exam. Horses	ALT (U/L)	AST (U/L)	Creatinin (mg/dl)	Urea (mg/dl)
Control N = 3	12.666 ± 0.881	49.666 ± 1.202	1.363 ± 0.023	29.33 ± 0.881
Diseased N = 6	23.666 ± 3.326***	70.833 ± 6.431***	1.78 ± 0.050	39.5 ± 1.87***

Significance *P >0.05, **P >0.01, ***P >0.001 ALT, Alanine amino-tranferase; AST, Aspartate amino-transferase; IU/L, Internationnal unit per liter; mg/dl, milligram per deciliter.

cELISA for detecting of *T. equi* antibodies in examined horses:-

This diagnostic method was revealed that 53 (15.7%) of examined horses were seropositive to *T. equi*. cELISA was recorded the highest prevalence rate of *T. equi* in examined horses in compared with other diagnostic methods (microscopical examination and PCR) (Table 4). *B. caballi* antibodies weren't detected in examined serum samples.

Table (4). Prevalence of *T. equi* in examined horses by using microscopic examination of stained blood smears, cELISA and PCR.

Test	Samples No.	Positive		Negative	
		No.	%	No.	%
Blood film	305	32	10.4	273	89.5
cELISA	305	49	16.1	256	83.9
PCR	24	3	12.5	16	64

cELISA, competitive Enzyme linked immunosorbent assay; PCR, Polymerase chain reaction.

Molecular detection of *T. equi* DNA in blood of examined horses:

PCR technique was carried out on 24 horse blood samples (Table 4). These samples were amplified with a prevalence12.5% and the target bands were detected in between 200 to 300bp (nearly about 283bp) (Fig 2**).** No positive reaction was observed by PCR for *B. caballi* infection.

Detection of *T. equi* DNA in horses blood samples by PCR assay.

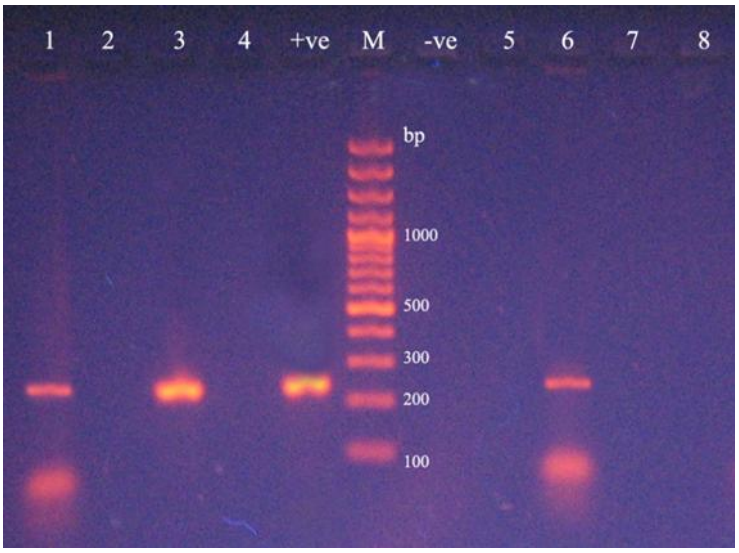


Fig (2): Single PCR performed with genomic DNA of *T. equi*
M:100 bp DNA ladder., +ve : positive control., -ve : negative control., Lane1,3& 6: positive sample., Lane 2, 4,5,7&8 : negative sample.

Discussion

Equine theileriosis is a notifiable disease with a wide distribution. Equine piroplasmosis is of great importance because of the international movement of horses in connection with equine sport competitions, and some countries restrict the entrance of horses that are serologically positive for piroplasma species. For this reason, specific and sensitive tests to detect piroplasma infections are needed. The clinical picture of piroplasmosis is variable and often non-specific and gives only a hint for further investigation. Fever, icterus, anorexia and depression are the most frequent symptoms with low erythrocyte counts, low hemoglobin **(Zobba *et al*., 2008).**

In present study the results of the clinical investigation of examined horses were in agreement with those reported by **Rashid *et al*. (2009), Takeet *et al*. (2009), Ibrahim *et al.* (2011), Salib *et al*. (2013), OIE (2014)**and **Salem and El-Sherif (2015)** who described the *Theileria* infection in equines in relation to the history of fever, anaemia, anorexia and weakness.

The clinical signs observed in clinically affected horses of the present study might be attributed to the infection of the RBCs by the theilleria merozites which destructed the cell wall of the RBCs causing hemolytic anemia and Icterus. Also parasites spread through the lymphoid system and other organs rapidly could cause disturbance of the immune and endocrine systems.

By microscopic examination of Gemsia stained blood smears revealed the presence of small intra-erythrocytic polymorphic stages of *T. equi* meroziotes (Fig. 1A, B&C) in addition to other two schizont forms in lymphocytes (Fig. 1D) in 32 out of 305 horses in screened horses with a prevalence rate of 10.4%. The present results of microscopic findings were agreement with **Rashid *et al.* (2009), Adaszek *et al.* (2011), Ibrahim *et al*. (2011) and Salib *et al.* (2013)**.

Concern The current recorded prevalence rate nearly agreement with those by **Katie (2010) and Ibrahim *et al.* (2011)** who reported the prevalence of *T. equi* was 14% and 18% in Egypt. In other country also **Ribeiro *et al.* (2013a)** stated that 12% in Brazil.

The high prevalence mentioned by **Farah *et al.***

(2003) was 38.8% in Egypt and **Salim *et al.* (2008)** 63.5% in Sudan. These rates were higher than results reported that in the current study.

The low prevalence recorded by **Ribeiro *et al.* (2013b)** 3.1% in Portugal**, Malekifard *et al.* (2014)** 6.25% in Iran and **Sumbria *et al.* (2015)** 4.17% in India.

The variation in the prevalence of *T. equi* may be attributed to different factors such as immunological naivety, accuracy of screening tests especially in asymptomatic piroplasm carrier horses, geographical areas differences including import/export restriction, density of infected ticks and horses, inappropriate management practices and differing climatic factors such as temperature and humidity which affect the tick vector proliferation.

In the present study, the hematological parameters showed a significant decrease in the means values of total RBCs count and Hb concentration (Table1) of diseased animals group when compared to healthy animals control group. This present hematological findings were in agreement with all those mentioned by **Zobba *et al.*(2008), Al-Saad (2009), Rashid *et al.* (2009), Ibrahim *et al.* (2011), Adaszek *et al.* (2011), Salib *et al.* (2013) and Salem and El-Sherif (2015).**

The reduction of erythrogram in infected horses with *T. equi* in the present work can be explained in light of several reasons, this might be due to anemia, three mechanisms of hemolysis have been described in canine babesiosis to explain the pathogenesis of anemia, but the same causes could ascribed to equine piroplasmosis **(Zobba *et al.*, 2008)**: mechanical mechanism by trophozoite intra-erythrocyte binary fission, immunomediated mechanism by autoantibodies hepatobiliary directed against components of the membranes of infected and uninfected erythrocytes, and toxic mechanism by hemolytic factor produced by the parasite **(De Gopegui *et al.*, 2007).**

In the present study a significant increase in TLC, lymphocytes and eosinophils count (Table1) were observed in infected animal groups. These changes in leukogram were in agreement with the results of **Ibrahim *et al.* (2011)** and **Salem and El-Sherif (2015)** who proposed that the parasitic infestations usually induce eosinophilic response. **Ike *et al.* (2005)** indicated that in leukocytic findings, TLC increases remarkably with the rise of parasitized erythrocytes rate and the decrease in RBCs count. Also, **Bashiruddin *et al.* (1999)** clarify the increase in TLC may be due to the stimulation of lymphoid system and bone marrow as immune response against the parasite or their toxins resulting in proliferation of lymphocytes.

The present biochemical findings revealed that there were significant reduction seen in total proteins values (Table 2), which agree with **Hailat *et al.* (1997) and Al-Saad (2009)** who stated that decrease protein levels during blood parasitic infection may occur due to digestive disturbance, destruction of protein due to fever as well as less production from liver. Concerning the globulins level, an increase was observed. The results are in accordance with findings by other investigators **Takeet *et al.* (2009), Ibrahim *et al.* (2011) and Salib *et al.* (2013).** The observed hyperglobulinemia can be attributed to an increase in the globulin concentration in response to parasitic antigen and released hemoglobin from destructed erythrocytes.

In the present study significant increase in the activities of ALT and AST (Table 3) was recorded in the diseased animal group when compared to control group. The increase of serum hepatic enzymes could be the result of centrilobular degeneration and necrosis of hepatocytes which generally express as liver damage. These results corroborated by **Takeet *et al.* (2009), Ibrahim *et al.* (2011) and Salib *et al.* (2013).**

Urea and creatinine level were higher for infected horses than for control (Table 3).The difference was statistically significant for urea but not for creatinine levels. Dehydration of infected animals could explain the elevation of serum urea concentrations. These agree with **Takeet *et al.* (2009) and Ibrahim *et al.***

(2011). It can be concluded that theileriosis hasa deleterious effect on the health of horses, as indicated by the altered haematological and biochemical parameters of infected animals.

The clinical picture of piroplasmosis is variable and often non-specific and gives only a hint for further investigation. Despite being highly specific, blood smear analysis is of limited sensitivity to detect the agents of piroplasmoses during the acute phase of the infection **(Butler *et al.*, 2008).** This is especially true in apparently healthy carrier hosts, during the latent phase; the parasitaemia becomes too low to detect positive chronic cases.

Serological tests are the method of selection to detect latent infections. The detection of specific antibodies by various serological methods has been recognized as the method of choice for the identification of persistently infected animals, which were subject to restrictions in international commerce. Serological methods also facilitated the testing of large numbers of animals for epidemiological studies **(Bose and Peymann 1994).**

Evidence was provided that good crude antigenic preparations of intracellular parasites were difficult to produce, particularly due to the presence of host contaminant components, such as red blood fragments which made the standardization of immunological assays a crucial step by increasing the occurrence of non-specific reactions **(Mahoney and Goodger 1981).**

The competitive inhibition enzyme-linked immunoassay (cELISA) was later improved by the use of a recombinant protein instead of culture – derived whole parasites **(Cunha *et al.*, 2002).** This test overcomes the problem of antigen purity because specificity depends only on the monoclonal antibody used.

The prevalence of *T. equi* in the tested samples with the three diagnostic methods was illustrated in Table (4) and showed that the highest prevalence was recorded by cELISA compared with microscopic examination and PCR.

It was found that, 49 (16.1%) of serum sample were positive for *T. equi* by cELISA. This result was in agreement with **Ibrhaem *et al*. (2011), Baptista *et al*. (2013) and Sumbria *et al*. (2016).** The high prevalence obtained by the serological assays in this study may be related to various factors such as false-positivity or cross reaction.

By comparing the direct and serodiagnosis results with PCR results revealed false positive and false negative results.

False positive did not show positive amplification in PCR for *T. equi*. This may be attributed to the parasites clearance from the circulating blood in the treated host **(Baptista *et al.*, 2013)**. In addition, parasites DNA in our study blood samples may have been present in levels below the sensitivity threshold of conventional PCR which was used **(Ribeiro *et al.*, 2013a)**. Serologically the antibodies could be detected even 4 years after the parasites had been eliminated from the blood **(Bruning 1996).**

While false negative results especially in the early infection period or in asymptomatic chronic carriers horses which have very low parasitaemia **(Alhassan *et al.*, 2007a and Franzén *et al*., 2009).** As the parasite can't detected by microscopic examination of blood smears. Despite being highly specific, blood smear analysis is of limited sensitivity to detect the agents of piroplasmoses and anaplasmosis **(Butler *et al.*, 2008).** On other side, serologically the antibody titers had not reached the detectable levels **(Baptista *et al*., 2013)**.

DNA amplification for the diagnostic detection of equine piroplasmosis has been known as a powerful tool both in the early phase of infection and in carrier animals **(Rampersad *et al*., 2003)**. In the present study, a total of 24 samples (which were positive with blood film examination and ELISA), were amplified with aprevalence 12.5, the target bands were detected at 283bp by using a universal primer pair (Fig. 2).

The importance of PCR as highly diagnostic tools for piroplasm which could be undetected in the blood of asymptomatic carrier horses that could infect ticks and transmit the disease to other horses **(Holman *et al*., 1997).** The transport of these carrier horses may result in the dissemination of the parasites in *T. equi*-

free countries. Like these asymptomatic carrier horses are known sometimes to adapt to infection and may appear only in certain circumstances, such as under stress or severe immune suppression resulting in relapse of the infection **(Abutarbush *et al.*, 2012).**

The parasites may reach several tissues of the host like liver, spleen, groin lymph node, lungs, heart, bone marrow, and brain **(Alhassan *et al.*, 2007b).** Recent studies have been shown that bone marrow may be a potential reservoir site of *T. equi* and *B. caballi* in clinically healthy horses (**Alhassan *et al.*, 2007b and Pitel *et al.*, 2010).**

In international trading, cELISA is considered as official test because it shows both current and past exposures to the parasite; on other hand, molecular test targets only the DNA of parasite, so for epidemiological studies, combination of both should be used **(Munkhjargal *et al.*, 2013)** because PCR shows the presence of parasite and cELISA explores the history of animal regarding the disease **Sumbria *et al.* (2016).**

Conclusion

To remain infection free, necessary tools include accurate screening diagnostic tests. serology alone cannot prove a state of activeor established infection, unless seroconversion is observed. PCR has been described as a more sensitive and specific method than microscopical examination and ELISA to confirming, characterizing, and differentiating detect equine piroplasms infections.

Preventive measures and Control strategies should be put into practice in order to reduce exposure to and infection of horses by equine tick-borne pathogens, which should include reducing host exposure to ticks, spraying horses with acaricides and treating positive animals. Further studies which investigate on the potential tick vectors will be useful for promoting an integrated and comprehensive control of these equine pathogens.

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دراسات طفيلية و بيوكيميائية وهيماتولوجية فى الخيول المصابة بمرض البيروبلازموزيس

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الملخص العربى

نظراً لأهمية مرض البيروبلازموزيس فى الخيول وما يسببه من خسارة هائلة عند ادخال حالات حاملة لهذا الطفيل فى المناطق الخالية منه وذلك عند عدم تشخيص الطفيل بالطرق التقليدية.أجريت هذه الدراسة لتشخيص مرض البيروبلازموزيس فى الخيول من مناطق متفرقة بمحافظتى الجيزة والقاهرة بمصر ، جمعت ٣٠٥ عينة دم من الخيول تتراوح أعمارها ما بين ٢- ٨ سنوات ومن كلا الجنسين ، حيث كانت جميع الخيول تحت الدراسة سليمة ظاهرياً ماعدا ٥٧ حيوان والتى كانت تعانى من أعراض ظاهرية والتى تتمثل فى فقدان الشهية – إرتفاع فى درجة الحرارة- شحوب بالأغشية المخاطية – تضخم بالغدد اليمفاوية. تم دراسة مدى انتشار مرض البيروبلازموزيس على مدار سنة كاملة.حيث أظهرت الدراسة أن معدل الإصابة بطفيل الثيليريا إكواى باستخدام الفحص المجهرى، اختبار الاليزا التنافسى كانت ١٠.٤٪، ١٦.١٪ على التوالى، ولم يتم تشخيص طفيل البابيزيا كابلى. كما أظهرت الدراسة التغيرات المعبرة ذات الأهمية فى كل من المعايير الهيماتولوجية والكيميائية فى دم الخيول المصابة عن الخيول السليمة. بمقارنة نتائج كل من الفحص المجهرى واختبار الاليزا التنافسى بنتائج تفاعل البلمرة المتسلسل (١٢.٥٪) حيث تميز الأخير بدرجة عالية من الدقة والخصوصية العالية ، أثبتت الدراسة ضرورة استخدام تفاعل البلمرة المتسلسل جانباً الى جنب مع الاختبارت التقليدية وخصوصاً فى حالات الحيوانات الحملة لطفيل والسليمة ظاهرياً.