

## *Seroprevalence of Babesia ovis in small ruminants in Siwa Oasis, Egypt*

**H. I. Hosein<sup>1\*</sup>, Samia A. Ahmed<sup>2</sup>, F. A. Ibrahim<sup>3</sup>, T. R. Abou-Elnaga<sup>3</sup>,  
M. A. Gebely<sup>3</sup>, Mona A. Mahmoud<sup>3</sup>**

<sup>1</sup> Department of Veterinary Medicine, Faculty of Veterinary Medicine, Beni-Suef University, Beni-Suef, Egypt

<sup>2</sup> Department of Veterinary Medicine, Infectious Diseases and Fish, Faculty of Veterinary Medicine, Cairo University, Cairo, Egypt

<sup>3</sup> Department of Animal and Poultry Health, Desert Research Center, Cairo, Egypt

Seroprevalence of *Babesia ovis* in sheep and goats was studied in Siwa Oasis between January 2002 and January 2003. A total of 240 blood samples were collected from 108 sheep and 132 goats for preparation of blood smears and for separation of serum samples and tested against *B. ovis* by using IFAT. *B. ovis* was detected in 55 (50.92%) and 59 (44.69%) blood smears examined in sheep and goats, respectively. The overall prevalence of *B. ovis* infection was 71.3% in sheep and 68.2% in goat using IFAT. The seasonal prevalence of *B. ovis* peaked in both spring and summer as revealed by blood smear examination and IFAT. A total of 143 ticks were collected from 62 sheep and 81 goats during the study. The ticks examined were *Rhipicephalus turanicus* (75.52%) and *Hyalomma anatolicum* (24.48%).

*Babesia ovis* is an intraerythrocytic protozoan parasite that causes babesiosis in sheep and goats. The disease has economic importance in subtropical and tropical regions of the world, Ristic, (1981) and Levine, (1985). It leads to significant losses among small ruminants due to its drastic effect on hemobiotic system, Radostits *et al.* (2000). Since the indirect immunofluorescence antibody technique (IFAT) has become available for a serological diagnosis and survey of babesiosis, it is possible to test serum samples for babesiosis with high sensitivity.

A number of epidemiological surveys on ovine babesiosis have been reported using IFAT technique in Egypt, (Mahmoud, 1992; Abou-Elnaga, 2000,2002) however there no literature on ovine babesiosis in Siwa Oasis.

The purpose of this study is to document the presence of *B. ovis* in Siwa Oasis in order to provide information on the prevalence and intensity of infection in sheep and goats.

### **Materials and Methods**

**Sampling.** Blood samples were collected from 108 sheep and 132 goat (aged from three months

and up to three years) in Siwa Oasis for a period of one year from January 2002 to January 2003. The animals sampled from each flock were selected randomly. Sera were separated by centrifugation and stored at -20°C.

**Blood smears.** Thin blood smears were prepared from the blood of 108 sheep and 132 goats and examined according to (Kruse and Prichard, 1982).

**IFAT.** 240 serum samples (108 from sheep and 132 from goats) were tested for antibodies against *B. ovis* antigen. The antigen was prepared according to (Leeflang and Perie, 1972). Whole blood (6% parasitaemia) was drawn into phosphate buffer saline (PBS pH 7.4) in such a proportion that coagulation was prevented (8ml blood/100 ml PBS). The mixture was centrifuged and the sediment was washed three times in PBS to free the antigen, the final sediment was restored to the initial blood volume, adjusted by PBS containing 1% bovine serum albumen to aid in adherence of cells to the microscopic slides. Uniform thin smears, covering the entire surface of a clean microscopic slide, were made and air dried. Masking tap was applied to the dried cell surface, wrapped in groups of five sides in aluminum foil and stored at -70 °C until used by

\* Corresponding author. Tel.: +20 10194646;  
fax: +20 0822327982. (H. I. Hosein).

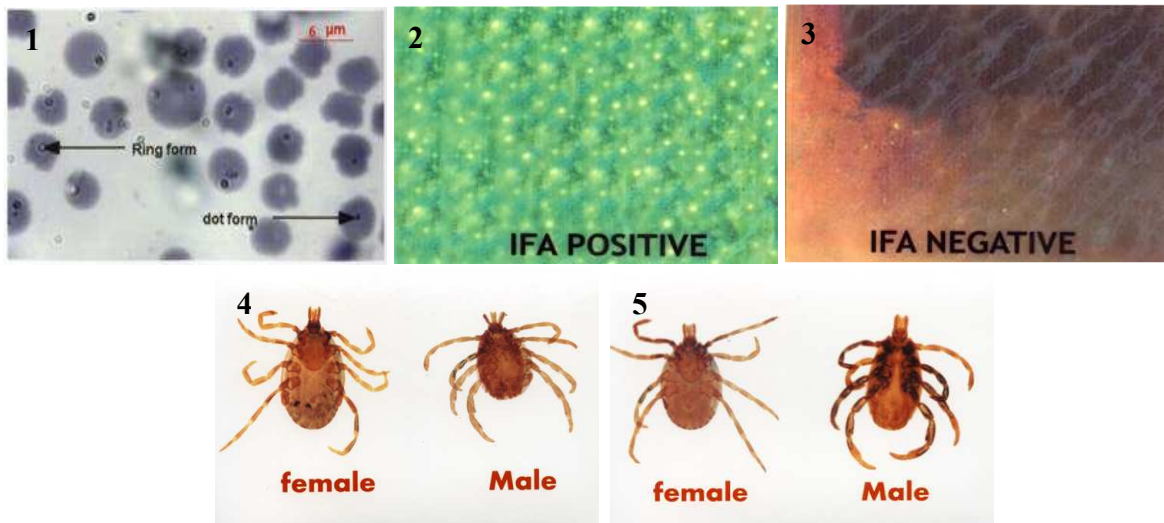


Photo (1): Ring form and dot form of *B. ovis* in blood film. Photo (2): Negative IFA for *B. ovis* Photo (3): Positive IFA for *B. ovis*. Photo (4): *Rhipicephalus turanicus* female and male ticks collected from sheep and goats. Photo (5): *Hyalomma anatolicum* female and male ticks collected from sheep and goats.

Table (1): Prevalence of *B. ovis* infection among sheep using blood film examination.

Season	No. of Examined sheep	Blood film examination	
		Positive cases	
		No.	%
Summer	41	21	(51.21%)
Autumn	11	5	(45.45%)
Winter	31	15	(48.38%)
Spring	25	14	(56.00%)
Total	108	55	(50.92%)

Table (2): Prevalence of *B. ovis* infection among goats using blood film examination

Season	No. of Examined Goats	Blood film examination	
		Positive cases	
		No.	%
Summer	19	10	(52.63%)
Autumn	49	17	(34.69%)
Winter	29	13	(44.82%)
Spring	35	19	(54.28%) *
Total	132	59	(44.69%)

\* Significant  $p \leq 0.05$

**Table (3): Prevalence of *Babesia ovis* infection among sheep using indirect fluorescent antibody test (IFA) in Siwa Oasis.**

Season	No. of Examined sheep No.	No. of positive cases	
		No.	%
Summer	41	31	(75.6%)
Autumn	11	7	(63.6%)
Winter	31	20	(64.5%)
Spring	25	19	(76%)
Total	108	77	(71.3%)

**Table (4): Prevalence of *Babesia ovis* infection among goats using indirect fluorescent antibody test (IFA) in Siwa Oasis.**

Season	No. of Examined Goats	No. of positive cases	
		No.	%
Summer	19	14	(73.7%)
Autumn	49	32	(65.3%)
Winter	29	18	(62.1%)
Spring	35	26	(74.3%)
Total	132	90	(68.2%)

**Table (5): Identification of tick species collected from sheep and goats in Siwa Oasis**

Total Number of collected tick samples	Sheep				Goats			
	<i>Rhipicephalus turanicus</i>		<i>Hyalomma anatolicum</i>		<i>Rhipicephalus turanicus</i>		<i>Hyalomma anatolicum</i>	
	Male	Female	Male	Female	Male	Female	Male	Female
143	16	26	7	13	21	45	4	11
	11.18%	18.18%	4.89%	9.09%	14.68%	31.46	2.79%	7.69%

IFAT using methods previously described by (Goff *et al.*, 1982; Tender and Friedhoff, 1988).

**Ticks.** One hundred and forty three ticks (62 from sheep and 81 from goats) were collected from sheep and goats. Ticks were identified in Parasitology Department, Faculty of Veterinary Medicine, Cairo University using stereozoom microscope (Olympus) and Hoogstraal keys according to (Hoogstraal, 1956).

### Results

The examination of Giemsa stained blood smears revealed the presence of *B. ovis* piroplasms

in 55 sheep (50.92%) and 59 goats (44.69%). (Table 1, 2, Photo 1). The seasonal distribution of *B. ovis* infection peaked in spring (56%) and (54.28%) in sheep and goats respectively followed by summer (51.21%) and (52.63%) in sheep and goats respectively. The prevalence of infection between seasons was found to be statistically significant ( $p < 0.05$ ) according to (Snedecor and Cochran, 1980).

A total of 240 ovine serum samples (108 from sheep and 132 from goats) collected from Siwa Oasis were tested for the presence of antibodies

against *B. ovis*; 167 samples (77 from sheep and 90 from goats) were found to be positive and the overall prevalence was estimated to be 69.58% (71.3% in sheep and 68.2% in goats). Photo (2) shows negative IFA to *B. ovis*. Photo (3) shows positive IFAT to *B. ovis* (Table 3) shows the number of sheep tested and the proportions reacting positively to the IFAT of *B. ovis* antibody in different seasons. Table 4 shows the number of goats tested and the proportions reacting positively to the IFAT of *B. ovis* antibody in different seasons. The seasonal distribution of *B. ovis* antibodies peaked in spring (76% and 74.3%) in sheep and goats respectively followed by summer (75.6% and 73.7%) in sheep and goats, respectively.

One hundred and forty three ticks (62 from sheep and 81 from goats) (Table 5) were examined for the determination of sex and species. Ninety five (39 from sheep and 56 from goats) were found to be female. In terms of species, 75.52% were *Rhipicephalus turanicus* (67.74% in sheep and 81.48% in goats), and 24.48% were *Hyalomma anatolicum* (32.26% in sheep and 18.52% in goats) photo (4, 5). *Rhipicephalus turanicus* was the predominant species infested both sheep and goats, male *Rhipicephalus* represented (11.18% and 14.68%) in sheep and goats respectively while female ticks represented 18.18% in sheep and 31.46% in goats.

### Discussion

In this study, 50.92% of sheep and 44.69 % of goats were infected with *B. ovis* using blood film examination. A higher and lower prevalences were recorded by many authors. Jurasek, (1986) recorded that 48% of goats were positive for *Babesia ovis*. Inci *et al.*, (2002) detected *Babesia ovis* in 17.7% of sheep and 6.38% of goats. Razmi *et al.*, (2003) recorded *Babesia ovis* in 23.5% from sheep and in 0.5% in goats. Such variation in the prevalence may be attributed to several factors including difference in localities and consequently difference in climatic conditions which affect the vector activity.

Regarding seasonal variation of prevalence of *Babesia ovis* using blood film examination in sheep and goats. In the spring, the highest prevalence rate (56%) and (54.28%) in sheep and goats, respectively was recorded, followed by summer (51.21%) and (52.63%) in sheep and goats respectively. Many authors reported that the

highest prevalence of *Babesia ovis* was observed during spring and summer which are considered the seasons of high activity of tick vector, (Rodriguez *et al.*, 1989; Trifonov and Ruseve 1989; Pipano, 1991; Yeruham *et al.*, 1992).

Concerning the serological prevalence of *Babesia ovis* infection among sheep and goats using IFAT, 71.3% of the examined sheep and 68.2% of the examined goats were proved to be positive reactors. Seasonal variation of prevalence of seroreactors of *Babesia ovis* in sheep and goats showed the same pattern observed by blood smear examination as spring represented the highest prevalence followed by summer, winter and autumn in sheep. In goats spring represented the highest prevalence followed by summer, autumn and winter. IFA has been used by many authors for detection of *B. ovis* among sheep and goats. In Egypt, Mahmoud, (1992) recorded the prevalence of *Babesia ovis* in sheep in Beni-Suef, El-Sharkia, Kafr El-Shikh and El-Fayoum governorates as 90.9%, 88.54%, 87.5% and 86.17% respectively. Papadopoulos *et al.* (1996a) recorded *Babesia ovis* in 52.1% sheep and 36.4% goats. Friedhoff (1997) detected *Babesia ovis* in 50% sheep and 40% goats. Karatepe *et al.* (2003) reported the prevalence of *Babesia ovis* 34.78% in goats and 23.63% in sheep. Variation in the seroprevalence of *Babesia ovis* infection obtained by different authors and those obtained in this study may be attributed to the geographical and climatic conditions, activity of the vector, parasitic status of the investigated area and immune status of the examined animals (Ferrer *et al.*, 1998).

Concerning the identification of ticks collected from blood parasite infected sheep and goats, the identified tick species were *Rhipicephalus turanicus* and *Hyalomma anatolicum*. Similar results were obtained by Trifanov and Ruseve, (1989), Al-Asgah, (1990), Papadopoulos *et al.* (1996b), Friedhoff, (1997), Yeruham *et al.* (2000), Abd-El Baky, (2001), Mazyad and Khalaf (2002) and Inci *et al.* (2003). *Babesia ovis* has been reported to be transmitted by *Rhipicephalus turanicus* and *Hyalomma anatolicum* Yeruham *et al.* (1992) and Friedhoff, (1997). Climatic conditions particularly temperature, rainfall, 23 iver humidity and the length of the rainy .....on have been demonstrated as the important factors affecting the life cycle and abundance of ticks, (Mushi *et al.*, 1996 ; Yeruham *et al.*, 1996;

Yakhchali and Hosseine, 2006).

Concerning the locality of the investigated area, it represents the most suitable conditions favoring the activity of ticks due to the high temperature in such desert area, the presence of cracks in walls and ground and the lack of hygienic conditions necessary for control of ticks. Such conditions clarify the high prevalence of blood parasites among investigated sheep and goats in Siwa Oasis.

In conclusion, the results obtained in this study clarifies that *B. ovis* is important prevalent pathogen among sheep and goats in Siwa Oasis due to the abundance of the ticks vector as a result of the suitable climatic conditions, poor buildings and lack of sanitations.

### References

- Abd El-Baky, S. M. M. (2001):** Prevalence of external parasites in the South eastern desert of Egypt. J. Egypt. Soc. Parasitol., 31(1): 223-232.
- Abou-Elnaga, T. R. (2002):** Effects of babesiosis on some serum biochemical parameters in sheep and goats in Matrouh governorate. 10<sup>th</sup> Sci. Cong. 2002. Fac. Vet. Med, Assiut Univ., Egypt. 235-247.
- Abou-Elnaga, T. R. (2000):** Some diseases transmitted by ticks in small ruminants Ph.D. Thesis Vet. Sci. Cairo Univ., Egypt.
- Al-Asgah, N. A. (1990):** Seasonal dynamics of *Rhipicephalus turanicus* (Acari: Ixodidae) on sheep and goats in the Al-Sarawat Mountains of Makkah province, Saudi Arabia. Bulletin of the Institute of Zoology Academic Sinica. 29 (2): 113-119.
- Ferrer, D.; Castella, J. and Gutierrez, J. F. (1998):** Seroprevalence of *Babesia ovis* in sheep in Catalonia, Northeastern Spain. Vet. Parasitol., 79: 275-281.
- Friedhoff, K. T. (1997):** Tick-borne diseases of sheep and goats caused by *Babesia*, *Theileria* or *Anaplasma* spp. Parasitologia 639 (2): 99-109.
- Goff, W. L.; Wager, G. G.; Ciaic, T. M. and Long, R. F. (1982):** The bovine immunoresponse to tick derived *Babesia ovis* infection. Serological studies of isolated immunoglobulins. Vet. Parasitol., 11 (2-3): 109-120.
- Hoogstraal, H. (1956):** reference to Equatoria province and with preliminary reviews of the genera *Boophilus* African Ixodidae 1-Ticks of the Sudan (with special, Margaropus and Hyalomma). Vol. I. U.S. Naval medical researches unit No.3, Cairo, Egypt.
- Inci, A.; Karaer, Z. and Ica, A.(2002):** Babesiosis in sheep and goats around Kayseri.Saglik – Bilimleri Dergisi, Firat Univ. Vet., 16 (1): 79-83.
- Inci, A.; Nalbantoglu, S.; Cam, Y.; Atasever, A.; Karaer, Z.; Cakmak, A.; Sayin, F.; Yukari, B. A.; Ica, A. and Deniz, A. (2003):** Theileriosis and tick infestations in sheep and goats around Kayseri.Turk Veterinerlik-ve-Hayvancilik-Dergisi., 27:(1)57-60.
- Jurasek, V. (1986):** Results of the laboratory examinations of parasitoses in the animals of Mozambique. Folia Veterinaria. 30 (1): 93- 97.
- Karatepe, M.; Karatepe, B.; Cakmak, A. and Nalbantoglu, S. (2003):** Prevalence of *Babesia ovis* in sheep and goats in the region of Nigde. Turkiye Parazitoloji dergisi. 27 (1): 18- 20.
- Kruse, G. O. W. and Prichard, M. H. (1982):** The collection and preservation of animal parasites. HwMI, University of Nebraska press, Lincoln, U.S.A.
- Leefflang, P. and Perie, N. M. (1972):** Comparative immunofluorescent studies on four *Babesia* species of cattle. Res. Vet. Sci., 13: 342-346.
- Levine, D. N.(1985):** Veterinary Protozoology. Iowa State University Press, Ames. pp. 413.
- Mahmoud, M. S. (1992):** Some serological studies on *Babesia* species infecting sheep in Egypt. M.V.Sc. Thesis, Fac. Vet. Med. Cairo Univ., Egypt.
- Mazyad, S. A. M. and Khalaf, S. A. A. (2002):** Studies on *Theileria* and *Babesia* infecting live and slaughtered animals in Al-Arish and El-Hasanah, North Sinai governorate, Egypt. J. Egypt. Soc. Parasitol., 32 (2): 601-610.
- Mushi, E. Z.; Isa, J. F. W. and Sakia, R. M. (1996):** Monthly fluctuation of Ixodid tick infestation on indigenous goats in Botswana. Small Rum. Res., 21: 277-279.
- Papadopoulos, B.; Michel, B and Perie. N. M. (1996a):** Piroplasmosis of domestic animals in the Macedonia region of Greece. Vet. Parasitol., 63: 67-74.
- Papadopoulos, B.; Morel, P. C. and Aeschlimann, A. (1996b):** Ticks of domestic animals in the Macedonia region of Greece. Vet. Parasitol., 5 (63): 1-2, 25- 40.
- Pipano, E. (1991):** Observation on the seasonal distribution of blood parasites in sheep in Israel. Israel. J. Vet. Med. , 46 (1): 37-38.
- Radostits, O. M.; Gay, C. C.; Blood, D. C. and Hincheliff, K. M. (2000):** Veterinary Medicine. 9<sup>th</sup> ed WH Saunders Co LTD London New York Philadelphia Sydney.
- Razmi, G. R.; Naghibi, A.; Aslani, M. R.; Dastjerdi, K. and Hossieni, H. (2003):** An epidemiological study on *Babesia* infection in small ruminants in Mashhad suburb, Khorasan province, Iran. Small Rum. Res., 50: 39-44.
- Ristic, M.(1981):** Babesiosis. In: Diseases of cattle in the tropics. Ed.: M. Ristic. Martinus Nijhoff Publishers, The Hague, Netherlands,; pp. 443-468.
- Rodriguez, O. N.; Rodriguez, P.; Espaine, L. and Rivas, A. (1989):** Frequency of haemoparasites in sheep in Cuba. Revista-Cubana de Ciencias Vet., 20 (1): 57-70.
- Snedecor, G. W. and Cochran, W. G. (1980):** Statistical methods. Oxford & J.BH Publising com. 7<sup>th</sup>. ed.
- Tender, A-M. and Friedhoff, K. T. (1988):** Serodiagnosis of experimental and natural *B. equi* and *B. caballi* infections. Vet. Parasitol., 20 (1/3): 49-61.
- Trifonov, T. and Ruseve,V. (1989):** Epizootiologicalstudy of piroplasmosis of cattle, sheep and its ticks vectors in Stangia region of Bulgaria. Veterinarnya Sbirka, 87 (4):43-46.
- Yakhchali, M. and Hossein A. (2006):** Prevalence and ectoparasites fauna of sheep and goats flocks in Urmia suburb, Iran. Vet. Arhiv., 76: 431-442.
- Yeruham, I.; Hadani, A.; Gafker, F. and Rosen, S. H. (1996):** The seasonal occurrence of ticks (Acari: Ixodidae) on sheep and in the field in the Judean area of Israel. Exp. App. Acarol., 20: 47 – 65.
- Yeruham, I.; Hadani, A.; Gafker, F. and Rosen, S. H. (2000):** Ticks on two flocks of sheep in Israel: prevalence of infestation and corporeal distribution. Ann. Trop. Med.

Parasitol., 94 (7): 735 – 738.

Yeruham, I.; Hadani, A.; Gafker, F.; Rosen, S. H. and

Schlien, J. (1992): A field study of haemoparasites in two flocks of sheep in Israel. Isr. J. Vet. Med., 47 (3): 107-111.

### مسح سيرولوجي لطفيال البابينزيا اوفيز في المجترات الصغيرة بواحة سيوة – مصر

تم عمل مسح سيرولوجي لطفيال البابينزيا اوفيز في قطعان الأغنام والماعز بواحة سيوة في الفترة من يناير ٢٠٠٢ إلى يناير ٢٠٠٣ وجدت نسبة الإصابة باستخدام شرائح الدم ٥٠,٨% ، ٤٤,٦% في الأغنام والماعز على التوالي، بينما ارتفعت هذه النسبة إلى ٧١,٣% ، ٦٨,٢% في الأغنام والماعز على التوالي باستخدام اختبار التشعع الفلوريسنتي الغير مباشر . تم تصنيف القراد الجامد إلى نوعين هما: ريبيسفلس ترونكس ٧٥,٥% وهيالوما انتوليكم ٢٤,٤% بمنطقة البحث.