



A review on *Babesia* spp. and tick vectors in animals in Iran

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ABSTRACT

Babesiosis is an important tick-borne disease that affects a wide range of domestic and wild animals and occasionally humans in tropical and subtropical countries. So far, More than 100 different *Babesia* species have been identified in animals. Iran is one of the largest countries in the Middle East. The existence of two chains of mountains namely Zagros and Alborz has provided a number of climatic variations with different flora and fauna. These different climatic zones in Iran are potentially favorable for a large variety of tick vectors transmitting blood pathogen protozoa like *Babesia* spp. and *Theileria* spp. in animals. In the last decade, many molecular studies have been performed to identify *Babesia* spp. and tick vectors in different parts of Iran. This review article aims to provide useful information about the history, characters, geographical distribution, and prevalence of *Babesia* species and their related tick vectors in animals in Iran.

Keywords

Babesia spp., tick, animal, Iran

Number of Figures: 1
Number of Tables: 5
Number of References: 87
Number of Pages: 10

Introduction

Babesiosis is an important tick-borne disease in domestic and wild animals. The causative agent and vectors for this disease are *Babesia* spp. and Ixodid ticks, respectively [1]. *Babesia* species live and multiply as 'piroplasms' in erythrocytes of the vertebrate host, and in contrast to erythrocytic stages of Plasmodium do not contain pigment [2]. The female tick usually is infected by ingesting protozoa in the blood meal and transmitting them to new hosts as sporozoites in the tick's saliva of the next generation, larvae, nymphs, and adults. The main clinical signs of babesiosis are fever, hemoglobinuria, anemia, and icterus [2]. Babesiosis has a wide geographical distribution in temperate, tropical, and subtropical regions. Iran is located in the western Palearctic countries with a high diversity of hard ticks [3] that could act as a vector for *Babesia* spp. in animals [3]. Advances in molecular biology methods have led to changes in the identification of *Babesia* species and their vectors in the world. Many molecular studies have been conducted on *Babesia* spp. identification in domestic animals in Iran from 1998 to 2015. Based on these results, two systematic reviews were also published about babesiosis in sheep, goats, cattle, and horses [4, 5]. This review provides useful information about the history, characters, geographical distribution, and prevalence of *Babesia* species, and their related tick vectors in animals in Iran.

History of Babesia

Babesia, also called *Nuttallia*, is an apicomplexan parasite that infects red blood cells and is transmitted by hard ticks. It was discovered in the red blood cells of cattle by the Romanian bacteriologist Victor Babes in 1888. He later observed a similar organism in sheep blood [6]. Five years later, Smith and Kilbourne showed the presence of an intraerythrocytic parasite in dairy cattle with Texas cattle fever, a disease that had long stricken cattle ranchers in the southern USA [7]. They were given the name *Pyrosoma bygeminum*, and showed that ticks play a major role in the transmission of this disease. This was the first description of an arthropod-transmitted, pathogen of vertebrates. Starcovici chose the name *Babesia* for these organisms in 1893 [8]. Lignieres described two forms of *Babesia* as *B. bigemina* and *B. bovis* in cattle in Argentina in 1903 [9]. In Iran, Delpy identified *B. ovis* in sheep for the first time in 1936 [10]. The first human babesiosis was reported in a splenectomized Yugoslavian farmer in 1957. After the initial case in Europe, a case caused by *B. microti* was diagnosed in a splenectomized patient from California, USA, in 1966. *Babesia crassa* as a large *Babesia* species was isolated for the first time in the world from an Iranian sheep in 1981 [13].

Taxonomy, transmission, and morphology

The genus *Babesia* belongs to the phylum *Apicomplexa*, and the family *Babesiidae*. *Babesia* is a relatively pear-shaped, round, or oval parasite; the apical complex contains a polar ring, rhoptries, and subpellicular tubules. Micronemes and conoids are present in some stages and in some species [14]. Based on the merozoite size and comparison with erythrocyte radius, *Babesia* spp. are divided into large and small groups. The lengths of small and large *Babesia* are 1.0 to 2.5 μm and 2.5 to 5.0 μm , respectively. The morphometric method has no clear genetic basis, because the size and morphology of *Babesia* spp. may be changed during the asexual stage within red blood cells or when infects a non-specific host [15, 16]. So far, over 100 *Babesia* species have been identified in vertebrate hosts. Of those, eighteen species have been found to cause babesiosis in domestic mammals, including pigs, horses, cattle, sheep, goats, cats, and dogs. Most *Babesia* species have been reported in rodents, cattle, and carnivores (Table 1).

Life cycle

The life cycle of *Babesia* spp. consists of at least the asexual and sexual stages of reproduction that occur within the vertebrate host and tick vector, respectively. The sporozoites of the tick's salivary glands are generally transmitted to the vertebrate host 2-3 days after tick attachment. The sporozoites change to merozoites and enter red blood cells and divide by binary fission into new merozoites. Infected erythrocytes eventually rupture and release organisms that invade and multiply within other red blood cells. Some of the merozoites become pre-gametocytes that cannot be distinguished by a light microscope. When the tick vectors ingest the infected blood of the vertebrate host, the merozoites are microscopically detectable in the tick's gut after 10 hours [16]. The pre-gametocytes develop into gametocytes and begin to form ray bodies at the anterior of the piroplasm. The ray bodies form gametes and fuse to produce a motile zygote termed ookinete, which enters the gut epithelium cells. The ookinete starts meiotic division, resulting in many kinetes productions. At this stage, the kinetes migrate via hemolymph to different tick tissues such as ovarian cells. The infection of eggs leads to transovarial transmission. Some kinetes enter the salivary gland cells where a large multinuclear sporont is finally formed, giving rise to thousands of small sporozoites, which are injected during the feeding act and lead to transstadial transmission (Figure 1) [17, 18].

Babesia spp. infection in domestic animals in Iran

Cattle

Table 1.
Different *Babesia* species and tick vectors with geographical distribution in domestic animals [1]

Host	Species	Morphology	Tick vector	Distribution
Cattle	<i>B. bovis</i>	Small	Boophilus,Rhipicephalus	Africa, America, Asia, Australia, Europe
	<i>B. Bigemina</i>	Large	Boophilus,Rhipicephalus	Africa, America, Asia, Australia, Europe
	<i>B. major</i>	Large	Haemaphysalis	Asia, Europe
	<i>B. occultans</i>	Large	Hyalomma	Africa
	<i>B. ovata</i>	Large	Haemaphysalis	Asia
	<i>B. divergens</i>	Large	Ixodes,	Europe
	<i>B. sp. Kashi</i>	Large	Hyalomma	China
Buffalo	<i>B. orientalis</i>	Small	Rhipicephalus	Asia
	<i>B. bovis</i>	Small	Boophilus,Rhipicephalus	Asia, America
	<i>B. bigemina</i>	Large	Boophilus,Rhipicephalus	Asia, America
Horse, Donkey	<i>B. equi</i>	Small		Asia, Europe, America
	<i>B. caballi</i>	Large	Dermacentor, Hyalomma, Rhipicephalus	Asia, Europe, America
Pig	<i>B. trautammani</i>		Rhipicephalus	Africa, Europe
Sheep, Goat	<i>B. ovis</i>	Small	Rhipicephalus	Africa, Asia, Europe
	<i>B. motasi</i>	Large	Haemaphysalis	Africa, Asia, Europe
	<i>B. crassa</i>	Small	Unknown	Asia
Dog	<i>B. vogeli</i>	Large	Rhipicephalus sanguineus	Africa, America, Asia, Australia, Europe
	<i>B. conradae</i>	Small		America
	<i>B. gibsoni</i>	Small	Haemaphysalis longicornis, Rhipicephalus	Africa, America, Asia, Australia, Europe
	<i>B. rossi</i>	Large	Haemaphysalis	South Africa
	<i>B. canis</i>	Large	Dermacentor	Europe
Cat	<i>B. felis</i>	Small	Unknow	South Africa
	<i>B. cati</i>		Unknow	India

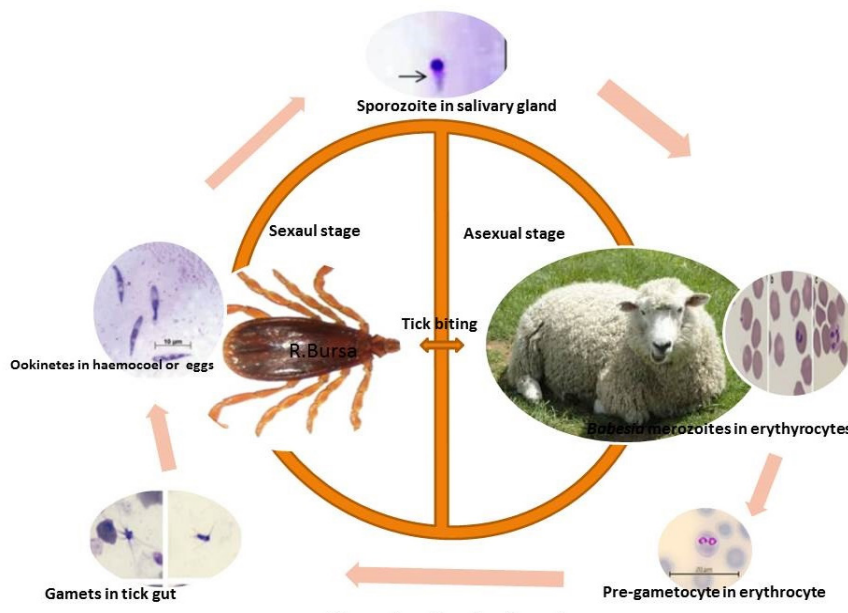


Figure 1.
The life cycle of *Babesia ovis* [1].

In Iran, the main species of *Babesia* in cattle are *B. bigemina* and *B. bovis* [19]. The first outbreak of babesiosis due to *B. bovis* was reported from a dairy farm in the Rasht area [20]. *B. bovis* localizes near the margin of the erythrocyte and is clearly smaller than *B. bigemina* and larger than *Theileria annulata*. The shape of *B. bovis* is ring form or ovoid. The erythrocytic stage of *B. bigemina* is large, round, oval, or pear-shaped and fills the whole erythrocyte when divided [17]. Few

studies have been performed on bovine babesiosis compared to ovine babesiosis in Iran. It seems that *B. bovis* and *B. bigemina* are more common in cattle in western and northwestern Iran (Table 2). Among Ixodid ticks, *B. annulatus*, *R. Bursa*, and *R. sanguineus* could be the vector for *Babesia* spp. in dairy cattle [23, 24]. The large *B. occultans* was recently reported from two cattle in the Miandoab area by molecular methods [27].

Table 2.
Reported *Babesia* species and related tick vectors in cattle in Iran

Year	<i>Babesia</i> species	Tick vector	Province or Area	Prevalence	Diagnostic methods	References
1977	<i>B. bovis</i>	<i>B. annulatus</i>	Rasht	-	Microscopic examination	20
2012	<i>B. bigemina</i>	-	Tabriz	-	Microscopic examination, PCR	21
2012	<i>B. bigemina</i>	-	Kurdistan	21%	Microscopic examination	22
2012	<i>Babesia</i> spp.	<i>R. sanguineus</i> and <i>R. bursa</i>	Kurdistan and West Azarbijan	-	PCR	23
2017	<i>B. bigemina</i> <i>B. bovis</i>	<i>B.annulatus</i> <i>R.sanguineus</i> <i>R.bursa</i>	Urmia	42%	Microscopic examination, PCR	24
2020	<i>B. bigemina</i> <i>B. bovis</i>	-	East and West Azarbijan	25.49%	Microscopic examination, PCR	25
2020	<i>B. bigemina</i> <i>B. bovis</i>	-	Mazandaran	33.33%	PCR	26
2021	<i>B. occultans</i>	-	Miandoab	-	Microscopic examination, PCR	27

Sheep and goats

Three *Babesia* species including *B. ovis*, *B. motasi*, and *B. crassa* have been reported in infected sheep and goats in Iran (Table 3). *Babesia ovis* is a small round piroplasm, situated usually at the periphery of the red blood cells of infected sheep [17]. This species is widespread in almost all parts of Iran [28, 29]. *Babesia motasi*, as a large species is less prevalent in Iran [30]. *Babesia ovis* is high pathogenic and causes anemia and hemoglobinuria, while *B. motasi* appears moderately virulent [31]. *Babesia crassa* is a large species that was isolated from an Iranian sheep. It is characterized by an oval tetrad form in infected erythrocytes. The protozoon appears to be nonpathogenic to intact sheep and goats [13]. The outbreaks of ovine babesiosis are recorded in sheep and goats at the age of 6- 12 months each year [32-33]. Potential vectors for *B. ovis* could be *R. bursa*, *R. sanguineus*, and *R. turanicus* [35, 36]

Horse and donkey

Babesia equi and *B. caballi* were reported from horses in different areas of Iran (Table 4). Studies

have shown that *B. equi* is more prevalent than *B. caballi* in Iranian horses and donkeys. The presence of *B. caballi* and *T. equi* was confirmed in 1940 by microscopic and molecular examination [54]. A few case reports of babesiosis due to *B. caballi* and *B. equi* have been published on horses in different parts of Iran from 1994 to 2000 [55-58]. In a study, *B. equi* infection was determined in donkeys of North Khorasan province by microscopic and molecular methods [67]. The name of *B. equi* has recently been changed to *Theileria equi*, because the sporozoites of *T. equi* first evade the lymphocytes and multiply by schizogony. After rupture of infected lymphocytes, the released merozoites enter the red blood cells and change to rounded, amoeboid, and a Maltese cross-shaped phenotype [2]. The vectors of *T. equi* could be *Hyalomma* spp. and also *Rhipicephalus* spp.. The eggs of these ticks were not infected with ookinetes to indicate transovarial transmission [17]. The merozoites of *B. caballi* are large and pear-shaped. They are produced by binary fission. *Babesia* infections are always detected in the eggs of tick vectors that could be transmitted to larvae in the next generation [17].

Table 3.
Reported Babesia species and related tick vectors in sheep and goats in Iran

Year	<i>Babesia</i> species	Tick vector	Province or Area	Prevalence	Diagnostic methods	References
1936	<i>B. ovis</i>	-	-	-	Microscopic examination	10
1966	<i>B. motasi</i>	-	West of Iran	-	Microscopic examination	32
1981	<i>B. crassa</i>	-	-	-	Microscopic examination	13
1998	<i>B. ovis</i>	-	Caspian sea,	15.93%	Serology	34
			Mountainous	58.81%		
			Persian gulf	12.04%		
			Desert climates	13.22%		
2002	<i>B. ovis</i>	<i>R. sanguineus</i>	Mashhad	24.6%	Microscopic examination	35
	<i>B. motasi</i>	<i>Hy. marginatum</i>		0.5%		
2003	<i>B. ovis</i>	-	Mashhad	14%, 0.5%	Microscopic examination	36
	<i>B. motasi</i>	-				
2006	<i>B. ovis</i>	-	Khouzestan	47.5%	Serology	37
2006	<i>B. ovis</i>	-	-	-	Microscopic examination, PCR	38
	<i>B. motasi</i>	-				
2007	<i>B. ovis</i>	<i>R. bursa</i>			PCR	39
		<i>R. sanguineus</i> ,				
		<i>R. turanicus</i>				
2008	<i>B. ovis</i>	-			PCR	40
2010	<i>B. ovis</i>	-	Different areas of Iran	24.6%	Microscopic examination, PCR	41
2012	<i>B. ovis</i>	-	-	-	Reverse line blot	42
2013	<i>B. ovis</i>	-	Tabriz	14%	PCR	43
2013	<i>B. ovis</i>	-	Dargaz, Kalat	0.99%	Microscopic examination, PCR	44
2013	<i>B. ovis</i>	-	Mazandarn province	5%	Microscopic examination, PCR	45
	<i>B. motasi</i>	-				
2014	<i>B. ovis</i>	<i>R. turanicus</i> , <i>Hya. marginatum</i>	North khorsan province		Microscopic examination, PCR	46
2014	<i>B. ovis</i>	<i>R. bursa</i>	West Azarbijana	16.7%	Microscopic examination, PCR	47
2017	<i>B. ovis</i>	<i>D. niveus</i> <i>D. marginatus</i>	Ardabil		PCR	48
2017	<i>B. ovis</i>	-	Lorestan		PCR	49
2017	<i>B. ovis</i>	<i>R. sanguineus</i> <i>Hya. suspense</i>	Gonbad Kavoos, Marvaeh tapaeh	-	PCR	50
2018	<i>B. ovis</i>	-	East azerbaijan	11.04%	PCR	51
2020	<i>B. ovis</i>	-	Baneh	86%	PCR	52
2020	<i>B. ovis</i>	-	Tonkabon, Ramsar	6%	PCR	53
	<i>B. motasi</i>	-				

Table 4.
Reported *Babesia* species and related tick vectors in horses and donkeys in Iran

Year	<i>Babesia</i> species	Tick vector	Province or Area	Prevalence	Diagnostic methods	References
2000	<i>B. cabali</i>	-	Fars province	-	Microscopic examination	57
2000	<i>B. cabali</i> , <i>B. equi</i>	-	Mashhad	-	Microscopic examination	58
2013	<i>B. cabali</i> , <i>B. equi</i>		North Khorasan Province	2%, 48%	Seology	59
2014	<i>B. equi</i>	<i>Hya. excavatum</i> , <i>Rh. bursa</i>	North Khorasan Province	45%	Microscopic examination, serology, PCR	60
2014	<i>B. cabali</i>		North Khorasan Province	4.8%	Microscopic examination, Serology	61
2014	<i>B. cabali</i> , <i>B. equi</i>		Urmia area	2.08%, 26%	Microscopic examination, PCR	62
2014	<i>B. equi</i>	-	Khuzestan Province	28.5%	PCR	63
2014	<i>B. equi</i>		Yazd area	4.7%, 22.8%	Microscopic examination, PCR	64
2014	<i>B. equi</i>	-	Mianeh area	4.1%	Microscopic examination	65
2015	<i>B. equi</i>	-	Ahvaz area		Microscopic examination, PCR	66
2015	<i>B. equi</i>	-	North Khorasan Province	3.77%, 50.94%	Microscopic examination, PCR	67
2016	<i>B. equi</i>		Piranshar area	9.6%, 96%	Microscopic examination, PCR	68
2017	<i>B. equi</i> , <i>B. caballi</i>	-	Isfhan, Sharekord	-	PCR	69
2017	<i>B. equi</i>	-	Kurdistan	1.61%	Microscopic examination, PCR	70
2018	<i>B. equi</i>	-	West Azarbijan	3.2%, 27.7%	Microscopic examination, PCR	71

Dog

For the first time, *Babesia canis*, and *B. gibsoni* were reported in the blood smear of splenectomized dogs and foxes from the north of Iran in 1973 [72]. Further study was shown that the isolated strain is mild and does not produce clinical signs in experi-

mentally infected dogs. Many studies have reported the large *Babesia* spp. in dogs of different parts of Iran (Table 5). *Babesia canis* as a large *Babesia* has three subspecies, *B. canis vogeli*, *B. canis rossi*, and *B. canis canis*. They are different in genotype, geographic distribution, pathogenicity, and vector-specificity [73].

Table 5.
Reported *Babesia* species in dogs in Iran

Year	<i>Babesia</i> species	Tick vector	Province or Area	Prevalence	Diagnostic methods	References
1973	<i>Babesia canis</i> <i>Babesia gibsoni</i>	-	Mazandaran Province	0.64%	Microscopic examination	72
2012	<i>B. canis</i>		Shiraz	-	PCR	74
2013	<i>B. canis</i>		Khousestan Province	3.75%	Microscopic examination	75
2014	<i>B. canis</i>		Charmahal Bakhtiri	7.5%	PCR	76
2016	<i>B. gibsoni</i>		Kerman province	5%	PCR	77
2020	<i>B. canis vogeli</i>		Shariar	-	PCR	78
2021	<i>B. canis vogeli</i>		Hamadan	4%	PCR	79
2022	<i>B. canis canis</i>		Tehran	-	PCR	80

Babesia gibsoni, *B. conradae*, and *Theileria annae* are termed small canine *Babesia*. Among different hard tick species, it has been reported that *R. sanguineus* could act as a vector for *B. vogeli* and *B. canis*, and *Haemaphysalis spp.* as a vector for *B. rossi* and *B. gibsoni* [73].

Camel

So far, a specific *Babesia* species has not been reported in camels worldwide. Based on molecular methods, *Babesia* species related to cattle and horses have been found in camels [81]. *Babesia caballi* and *T. equi* have been detected in camels in Iran [82-84].

Rodents

Babesia microti, a species of rodent origin, has been recognized as an agent of human babesiosis in the world [1]. There are a few reports about the presence of *Babesia microti* in Iran [85-87].

Conclusion

This review presented a comprehensive summary of research findings on the identification, prevalence, and distribution of *Babesia* species and their related vectors in domestic animals in Iran. In the last decade, many molecular studies have been performed to identify *Babesia* spp. and tick vectors in different parts of Iran. However, there is no information about *Babesia* infection in cats and wild animals. Further molecular and experimental methods will be needed to better understand the epidemiology of *Babesia* species and their related tick vectors in domestic and wild animals.

Competing Interests

The authors declare that they have no conflict of interest.

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How to cite this article

Razmi Gh. A review on *Babesia* spp. and tick vectors in animals in Iran. Iran J Vet Sci Technol. 2022; 14(2): 1-10.
 DOI: <https://doi.org/10.22067/ijvst.2022.76034.1131>
 URL: https://ijvst.um.ac.ir/article_42489.html