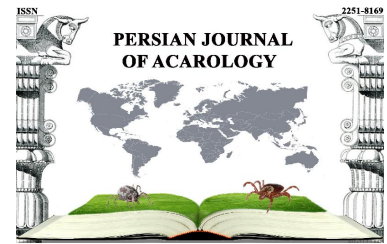




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Historical collection of ticks (Acari: Argasidae) from Iran and other regions of central Asia (Museogenomics research)

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In the work was examined historical collection of ticks (Acari: Argasidae), which were collected by E. Pavlovsky. Purposes of the work are to assess the state of the tick collection, to determine the possibility of using it for molecular genetic examinations, to attract interested researchers to scientific cooperation.

Academician Yevgeny Pavlovsky (1884–1965) is a Russian parasitologist, a well-known researcher, who studied ticks and tick-borne infections. A lot of parasitological expeditions to Central Asia (1934–1955) were organized by him. A large collection of ticks was collected during these expeditions. Ticks were collected in the territories of Iran, Uzbekistan, Tajikistan, Kyrgyzstan, Turkmenistan and Armenia. The collection includes 580 lots, containing more than 15000 ticks (Acari: Argasidae). Museum samples are in satisfied condition. Now the tick collection is stored in the Department of biology of the Military Medical Academy (St. Petersburg, Russia).

A trial examination of ticks was conducted with the aim of identifying genetic markers for tick-borne infections. The examination was carried out by a real-time polymerase chain reaction (Real-Time PCR) with the help of special reagent kits (Bondarenko *et al.* 2012; Timofeev *et al.* 2012) (Table 1).

Table 1. Reagent kits used for research.

Reagent kits	Pathogenes (DNA/RNA)	Nosological forms
«RealBest DNA <i>Borrelia burgdorferi</i> s.l.» («Vector-Best», Russia)	<i>Borrelia burgdorferi</i> (Johnson <i>et al.</i> 1984; Baranton <i>et al.</i> 1992) (23S rRNA)	Lyme disease
«RealBest DNA <i>Borrelia miyamotoi</i> » («Vector-Best», Russia)	<i>Borrelia miyamotoi</i> (Pritt <i>et al.</i> 2016) (glpQ)	Lyme disease
«RealBest DNA <i>Rickettsia species</i> » («Vector-Best», Russia)	<i>Rickettsia</i> (da Rocha-Lima 1916) (gltA)	Tick-borne recurrent fever
«RealBest DNA <i>Coxiella burnetii</i> » («Vector-Best», Russia)	<i>Coxiella burnetii</i> (Derrick 1939; Philip 1948)	Q-fever
«RealBest DNA <i>Francisella tularensis</i> » («Vector-Best», Russia)	<i>Francisella tularensis</i> (McCoy and Chapin 1912; Dorofe'ev 1947)	Tularemia

In general, 50 ticks were examined. Studies performed with adults of ticks *Ornithodoros papillipes* (Birula 1895; Guglielmone *et al.* 2010) without differentiation of sex (Table 2).

Table 2. Examined ticks.

No. lot	Total ticks	Place	Region
1	6	Outbuildings	Davalu, Armenia, 1939
2	5	Outbuildings	Bojnurd, Iran, 1942
3	4	Outbuildings	Shahrud, Iran, 1942
4	7	Outbuildings	Kermanshah, Iran, 1943
5	3	House	Kyzyl-Kim, Kyrgyzstan, 1939
6	4	Outbuildings	Osh, Kyrgyzstan, 1945
7	6	Mouse hole	Kara-Kula, Turkmenistan, 1948
8	6	Porcupine cave	Kara-Kula, Turkmenistan, 1948
9	5	Hedgehog hole	Beruni, Uzbekistan, 1936
10	4	Outbuildings	Beruni, Uzbekistan, 1936

Genetic markers of tick-borne infections were found in 10 of the 50 samples. Genetic markers of *Rickettsia species* were found in 8 samples. Genetic markers of *Borrelia burgdorferi s.l.* were found in one sample. *Coxiella burnetii* markers were detected in one case. *Borrelia miyamotoi* and *Francisella tularensis* markers were negative in all cases (Table 3).

Table 3. Detection of genetic markers of tick-borne infections.

No. lot	Region	Season	Total ticks	Positive samples		
				<i>Rickettsia species</i>	<i>Borrelia burgdorferi s.l.</i>	<i>Coxiella burnetii</i>
4	Iran	March 1943	7	1		
5	Kyrgyzstan	June 1939	3			1
7	Turkmenistan	March 1948	6	4		
8	Turkmenistan	June 1948	6	2		
10	Uzbekistan	July 1936	4	1	1	

Among the positive samples to *Rickettsia species*, the most of them are ticks, collected in the vicinity of Kara-Kala (lots No. 7 and 8). Also, genetic markers of *Rickettsia species* were found in ticks, collected in one of the outbuildings of the city of Beruni (lot No. 10), as well as in ticks, collected in Kermanshah (lot No. 4). Genetic markers of *Borrelia burgdorferi s.l.* were discovered in the body of ticks, caught in one of the outbuildings of the city Beruni (lot No. 10). *Coxiella burnetii* markers were found in tissues of ticks, caught in one of the houses of the city Kyzyl-Kim (lot No. 5).

Analysis of the results of real-time PCR for markers of DNA of the bacteria showed that in all cases the values of threshold cycle of amplification (C_t) ranged from 32 to 44 cycles of amplification. This may indicate a partial degradation of bacterial DNA in museum samples. However, it is suitable for further studies of the bacterial genome (Table 4).

Table 4. C_t values for positive samples.

No. sample	C_t - threshold cycle	Genetic markers of pathogens
4	37	
12	39	
33	44	
34	34	
35	37	<i>Borrelia burgdorferi s.l.</i>
36	37	
37	39	
42	32	
3	39	<i>Rickettsia species</i>
19	39	<i>Coxiella burnetii</i>

Thus, it is shown that the historical collection of ticks is relevant. The biological material has been preserved in satisfied condition and is suitable for molecular genetic research. The samples contain fragments of bacterial nucleic acids, which can be identified by a real-time polymerase chain reaction. It is advisable to conduct further researches. We invite interested researchers to discuss joint investigations.

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