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Hyalomma aegyptium as dominant tick in tortoises of the genus *Testudo* in Balkan countries, with notes on its host preferences

**Pavel Široký · Klára J. Petrželková · Martin Kamler
Andrei D. Mihalca · David Modrý**

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Abstract Collection of 1327 ticks sampled throughout Greece, Bulgaria, Romania and Croatia, from 211 tortoises belonging to three species, *Testudo marginata* Schoepff, *T. graeca* Linnaeus, and *T. hermanni* Gmelin, revealed the presence of four species of ixodid ticks, namely *Hyalomma aegyptium* (Linnaeus), *Haemaphysalis sulcata* Canestrini and Fanzago, *H. inermis* Birula and *Rhipicephalus sanguineus* (Latreille). Study confirmed the strong dominance of all life stages of *H. aegyptium* among ticks parasitizing west Palaearctic tortoises of genus *Testudo* Linnaeus. Furthermore, a considerable portion of ticks collected from tortoises in southwestern Bulgaria represent larvae and nymphs of *H. sulcata*. At the same area we collected as exception one larva and one nymph of *H. inermis* from a single specimen of *T. hermanni*. Our findings of four adults of *R. sanguineus* is the first record of this species from reptilian host. According to our results achieved on localities with syntopic occurrence of two tortoise species, *T. marginata* and *T. graeca* represent in

P. Široký (✉)
Department of Biology and Wildlife Diseases, University of Veterinary and Pharmaceutical Sciences, Palackého 1-3, Brno CZ-612 42, Czech Republic
e-mail: sirokyp@vfu.cz

K. J. Petrželková
Department of Mammal Ecology, Institute of Vertebrate Biology, Academy of Sciences of the Czech Republic, Květná 8, Brno CZ-603 65, Czech Republic

M. Kamler · D. Modrý
Department of Parasitology, University of Veterinary and Pharmaceutical Sciences, Palackého 1-3, Brno CZ-612 42, Czech Republic

A. D. Mihalca
Department of Parasitology and Parasitic Diseases, Faculty of Veterinary Medicine, Calea Manastur 3-5, Cluj-Napoca 400372, Romania

D. Modrý
Institute of Parasitology, Biology Center, Academy of Sciences of the Czech Republic, Branišovská 31, České Budějovice CZ-370 05, Czech Republic

the Balkans the principal hosts of *H. aegyptium*, whereas *T. hermanni* serves only as an alternative host in the areas close to range of either *T. marginata* or *T. graeca*.

Keywords *Hyalomma aegyptium* host preferences · *Haemaphysalis sulcata* · *Haemaphysalis inermis* · *Rhipicephalus sanguineus* · Balkan · *Testudo* spp

Abbreviations

GR Greece
BG Bulgaria
RO Romania
CR Croatia

Introduction

The genus *Hyalomma* Koch, 1844, distributed in Africa, southern Europe and Asia, contains 24 in part polytypic species of mostly large ticks with inornate scuta (Hoogstraal 1956; Kolonin 1983; Horak et al. 2002). The majority of them are difficult to identify, either due to morphological variability or to a tendency toward hybridization (Rees et al. 2003). The tortoise tick *Hyalomma aegyptium* (Linnaeus, 1758) is among the easily recognizable species. It possesses a set of morphological characters (for instance two equal, well separated spurs of coxa I in both sexes), justifying its classification within a separate subgenus *Hyalomma* (formerly *Hyalommasta* Schultze, 1930). All other species fall either into the subgenus *Hyalommina* Schulze, 1919 or *Euhyalomma* Filippova, 1984 (Apanaskevich 2003). *Hyalomma aegyptium* has a typical three-host life cycle and infests tortoises, other reptiles and mammals, but tortoises of the genus *Testudo* Linnaeus, 1758 are the principal hosts of adult stages (Hoogstraal and Kaiser 1960; Apanaskevich 2003, 2004).

Hyalomma aegyptium occurs in the Mediterranean region and in the Middle East, eastward up to Central Asia, Afghanistan and Pakistan (for reviews see Kolonin 1983; Apanaskevich 2003). On tortoises, it is most frequently reported from *Testudo graeca* Linnaeus, 1758 (e.g. Hoogstraal and Kaiser 1960; Petney and Al-Yaman 1985; Robbins et al. 1998; Leontyeva and Kolonin 2002) followed by *T. horsfieldii* Gray, 1844 (e.g. Kaiser and Hoogstraal 1963; Leontyeva and Kolonin 2002; Apanaskevich 2003), whereas other *Testudo* species are mentioned exceptionally, if ever (Zlatanova 1991; Burridge and Simmons 2003). Sweatman (1968) mentioned, certainly erroneously, that *H. aegyptium* parasitized *T. kleinmanni* Lortet, 1883 in Lebanon, where this tortoise species is not believed to occur. *H. aegyptium* is a dominant tortoise tick in the above-mentioned geographical region, despite the fact that the developmental stages of some other tick species can also feed on tortoises (Zlatanova 1991; Barnard and Durden 2000).

Small vertebrates play an important role in maintaining the natural focuses of many important diseases. Small mammals and birds are well-studied reservoirs of many tick-transmitted disease agents (e.g. Randolph et al. 2002). On the other hand, the potential of reptiles to serve as reservoirs of infectious agents is generally neglected, despite the fact that many of them belong to the most abundant vertebrates. In many areas of the Balkan Peninsula, three species of tortoise of the genus *Testudo* are still abundant and frequently share the pastures with domestic ungulates. Our study in selected localities of the Balkan is aimed at (i) elucidating the spectrum of tick species feeding on tortoises, and (ii) evaluating host preferences of adult *H. aegyptium* at localities where different *Testudo* species live together.

Materials and methods

Study areas

One field trip (30th May–14th June 2004) was carried out to Greek localities with both *T. marginata* Schoepff, 1792 and *T. hermanni* Gmelin, 1789 populations. Three study sites were situated between Platamonas and Leptokaria, at the periphery of Volos, and between Kardamili and Sparti. Another series of samples were obtained from populations of *T. hermanni* and *T. graeca* in the vicinity of Melnik in south-western Bulgaria (16–31st July 2002) and during a transect over the southern slopes of Rodopi Mountains (30th April–4th May 2005) in southern Bulgaria. Furthermore, 18 *T. hermanni* were checked for ticks during May 2003 at Žuljana, Pelješac Peninsula, Croatia, and 73 *T. graeca* were examined at localities Histria, Canaraua Fetei, and Greci, in easternmost Romania (15–20th August 2005, 25–26th April 2006) (for collecting sites see Fig. 1).

Tick collecting, developmental stage and species determination

Tortoises were found by walking through the habitat. All three *Testudo* species living in the Balkans are easily recognizable by their morphological traits (for the keys to the species of the genus *Testudo* see e.g. Ernst and Barbour 1989; Iverson 1992; Fritz and Cheylan 2001). Ticks were collected from tortoises using tweezers,

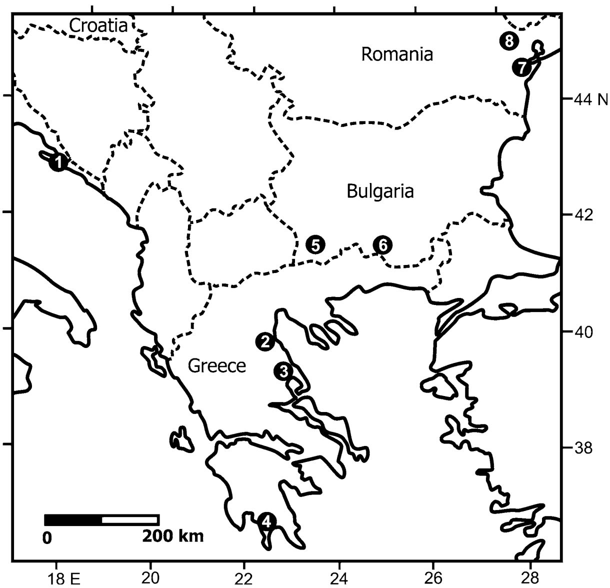

Fig. 1 Sampling areas; Croatia: 1—Žuljana, Pelješac Peninsula; Greece: 2—area between Platamonas and Leptokaria, 3—vicinity of Volos, 4—area between Kardamili and Sparti; Bulgaria: 5—vicinity of Melnik, 6—southern slopes of Rodopi Mountains; Romania: 7—Histria and Canaraua Fetei, 8—Greci

immediately put into plastic tubes containing 70% ethanol, and labeled with a field number of the tortoise specimen. Each sampled tortoise was measured in the field with a Vernier caliper (maximum straight carapace length, maximum straight carapace width) to the nearest milimetre (mm), weighed using pesola scales (g), and then sexed using the usual morphological criteria for *Testudo* spp. (Fritz and Cheylan 2001). Small specimens without expressed secondary sexual traits were classified as juveniles. Each tortoise was individually marked by temporary paint (retained 1–2 months) on its carapace, to prevent repeated recording and was released immediately afterwards at the point of capture. Ticks were transported to the laboratory and then determined to species level using morphological characters and available keys (e.g. Feldman-Muehsam 1948; Pomerancev 1950; Hoogstraal 1956; Nosek and Sixl 1972; Apanaskevich 2003).

Data analysis

Prevalence (%) and intensity of infestation by ticks (mean ± SD, range) were counted, where appropriate, for each tortoise and tick species and sex separately. Tortoise body measurements and numbers of males and females of *H. aegyptium* were compared between *T. marginata* and *T. hermanni* by Mann–Whitney test. Further analyses were run only on *T. marginata*, because of low prevalence (%) and intensity of infestation by ticks in other sampled tortoise species. By using Mann–Whitney test we tested differences in body measurements and condition index between males and females of *T. marginata*. Using the same tests we compared the numbers of males, females and nymphs of *H. aegyptium* and between males and females of *T. marginata.* Bonfferoni corrections (with added mean correlation between variables as a parameter) were used for *P*-values (Sankoh et al. 1997). We performed Spearman's correlations between numbers of males, females and nymphs of *H. aegyptium* and tortoise body measurements including condition index (Hailey 2000; Willemsen and Hailey 2002). We ran principle component analysis on body measurements including condition index of tortoises and the factor codes were used as covariate in ANCOVA by which we tested the differences in numbers of males, females and nymphs of *H. aegyptium* and between males and females of *T. marginata*.

Comparisons were not made between *T. hermanni* and *T. graeca* on Bulgarian localities, because of insufficient number of sampled *T. graeca* adults. Also statistical comparisons of different areas were not conducted.

Results

Tick species associated with *Testudo* tortoises

In total, 1327 ticks, belonging to four species, namely *Hyalomma aegyptium*, *Haemaphysalis sulcata* Canestrini and Fanzago, 1878, *Haemaphysalis inermis* Birula, 1895 and *Rhipicephalus sanguineus* (Latreille 1806), were collected from 211 tortoises (Table 1). *H. aegyptium* was found to be the most frequent tick species, constituting a clear majority of ticks found on both *T. hermanni* and *T. marginata* in Greek localities and *T. graeca* sampled in Romania (all Romanian ticks were found exclusively on tortoises at locality Greci). Interestingly, both *T. hermanni* and

Table 1 Total number of ticks collected during our survey

	Total no. larval/nymphal/adult tick recovered			
	H. aegyptium	*H. sulcata*	*H. inermis*	*R. sanguineus*
T. marginata (GR) (n = 27)*	205/105/448	0/0/0	0/0/0	0/0/4
T. graeca (BG) (n = 15)	4/10/4	24/1/0	0/0/0	0/0/0
T. graeca (RO) (n = 73)	35/26/64	0/0/0	0/0/0	0/0/0
T. hermanni (BG) (n = 55)	14/59/0	47/23/0	1/1/0	0/0/0
T. hermanni (GR) (n = 23)	221/18/13	0/0/0	0/0/0	0/0/0
T. hermanni (CR) (n = 18)	0/0/0	0/0/0	0/0/0	0/0/0

* collection of larvae from *T. marginata* is not complete, because of huge numbers of parasitizing larvae and limited time

T. graeca from the vicinity of Melnik, Bulgaria, were infested by pre-imaginal stages of *H. aegyptium* and *H. sulcata* (Table 2). Another two species of ticks were found as exceptions—one larva and one nymph of *H. inermis* were collected from a single *T. hermanni* at Melnik, Bulgaria, and two males and two females of *R. sanguineus* were collected from two *T. marginata* at Kardamili, south of Peloponnese Peninsula, Greece. No ticks were found on 18 *T. hermanni* inspected in Croatia or on 15 *T. hermanni* collected along southern slopes of Rodopi Mts. in Bulgaria.

Prevalence and intensity of tick infestation

Hyalomma aegyptium

All adult *T. marginata* (n = 26) were infested with males, females and larvae of *Hyalomma aegyptium*, with a mean male/female sex ratio of 2.93 (range 1.09–12), whereas nymphs infested 23 adult tortoises (88%). Intensity of infestation of *T. marginata* is given in Table 3. Larvae of ticks that parasitized *T. marginata* were not analyzed because of the strong intensity of infestation and the limited time spent in the field (more than a hundred larvae infested some tortoises).

In total, six of 21 adult Greek *T. hermanni* were infested with adult *H. aegyptium*, four tortoises exclusively by males and a single tortoise by a female tick only. One single specimen of Greek *T. hermanni* was simultaneously parasitized by both males (4) and females (1) of this tick species. Sixteen of 21 adult *T. hermanni* carried larvae and seven were parasitized by nymphs. Larvae and nymphs parasitized simultaneously six *T. hermanni*. The intensity of infestation on Greek *T. hermanni* is shown in Table 3 (except larvae: 13 ± 6.7, range 1–23). No *H. aegyptium* adults were collected from any Bulgarian *T. hermanni* (n = 55). However, of 40 tortoises examined at Melnik (Table 2), *H. aegyptium* larvae and nymphs infested eight and twelve of *T. hermanni*, respectively. Six *T. hermanni* were simultaneously infested by both immature stages.

Six of 15 *T. graeca* examined in Bulgaria were infested with *H. aegyptium*. Adult ticks (exclusively males) were found on two tortoises, three tortoises were parasitized by nymphs and three tortoises by larvae (one tortoise simultaneously by both immature stages and one tortoise by nymphs and adult ticks).

In Romania, during the 2005 field trip, five of 28 *T. graeca* were infested with adult ticks, none by both sexes simultaneously, two tortoises by a male, three by a female only, the intensity of infestation being only 1 adult tick per tortoise. Nymphs

Table 2 Tortoise infestation by ticks in area where *T. hermanni* and *T. graeca* occur together in SW Bulgaria. Two specimens of *Haemaphysalis inermis* are not included

Tick species	*Hyalomma aegyptium*						*Haemaphysalis sulcata*					
Host species	*T. graeca* (n = 15)			*T. hermanni* (n = 40)			*T. graeca* (n = 15)			*T. hermanni* (n = 40)		
Ticks life-stage	Larvae	Nymphs	Adults	Larvae	Nymphs	Adults	Larvae	Nymphs	Adults	Larvae	Nymphs	Adults
Prevalence (%)	20	20	13	20	30	0	43	7	0	35	25	0
Intensity of infestation												
Mean	1.3	3.3	2	1.8	4.9	0	4	1	0	3.4	2.3	0
SD	0.6	3.2	1.4	1.8	5.5	–	2.8	0	–	2.3	1.3	–
Range	1–2	1–7	1–3	1–6	1–21	–	1–8	1	–	1–7	1–4	–

Table 3 Comparison of the occurrence and intensity of infestation by adults and nymphs of *H. aegyptium* on *T. marginata* and *T. hermanni* in Greek localities with occurrence of both *Testudo* species. Larvae are not included, because of incomplete collecting from *T. marginata*

	H. aegyptium–males			*H. aegyptium*–females			*H. aegyptium*–adults total			*H. aegyptium*–nymphs		
	∑	mean ± SD	range	∑	mean ± SD	range	∑	mean ± SD	range	∑	mean ± SD	range
Testudo marginata												
males (n = 16)	218	13.6 ± 5.6	3–22	66	4.1 ± 3.2	1–13	284	17.8 ± 8.1	4–33	66	4.7 ± 4.8	1–17
females (n = 10)	116	11.6 ± 5.6	5–22	48	4.8 ± 3.1	1–11	164	16.4 ± 7.4	8–30	38	4.2 ± 5	1–15
juveniles (n = 1)	0	–	–	0	–	–	0	–	–	1	1 ± 0	1
total (n = 27)	334	12.8 ± 5.6	3–22	114	4.4 ± 3.1	1–13	448	17.2 ± 7.7	4–33	105	4.5 ± 4.8	1–17
Testudo hermanni												
males (n = 13)	5	2.5 ± 0.7	2–3	1	1 ± 0	1	6	2 ± 1	1–3	12	2.4 ± 2	1–6
females (n = 8)	6	2 ± 1.7	1–4	1	1 ± 0	1	7	2.3 ± 2.3	1–5	6	3 ± 1.4	2–4
juveniles (n = 2)	0	–	–	0	–	–	0	–	–	0	–	–
total (n = 23)	11	2.2 ± 1.3	1–4	2	1 ± 0	1	13	2.2 ± 1.6	1–5	18	2.6 ± 1.8	1–6

were found on 9 tortoises (intensity of infestation 2.9 ± 2.3, range 1–8) and larvae on 7 (intensity of infestation 4.4 ± 4.6, range 1–12). Five tortoises were concurrently parasitized by both immature stages. In the year 2006, 30 of 45 *T. graeca* examined at Greci were infested by *H. aegyptium*. Adult ticks parasitized 28 of these tortoises (intensity of infestation 2.3), ten tortoises carried both sexes of ticks (intensity of infestation 3; mean male/female sex ratio 1.7, range 1–4), and 17 tortoises were parasitized only by male ticks (intensity of infestation 1.6). One tortoise carried one single *H. aegyptium* female. Intensity of infestation was 2.1 ± 1.4, range 1–5, for all adult ticks; 1.7 ± 1.2, range 1–5 for males and 1.1 ± 0.3, range 1–2 for female ticks. Larvae of *H. aegyptium* were found only on four tortoises, each tortoise carrying one single specimen, in two cases together with male infestation.

Haemaphysalis sulcata

In Bulgaria, in the vicinity of Melnik, fourteen and ten of 40 inspected *T. hermanni* carried *H. sulcata* larvae and nymphs, respectively. Both immature stages infesting nine tortoises simultaneously. Larvae and nymphs of *H. sulcata* parasitized also six and one, respectively, of sampled *T. graeca* ($n = 15$). Only single *T. graeca* was infested by one *H. sulcata* nymph (simultaneously with six larvae).

Haemaphysalis inermis

One larva and one nymph of this tick species were collected at Melnik in Bulgaria from a single specimen of *T. hermanni*.

Rhipicephalus sanguineus

Four adults (two males and two females) of this tick species were collected from two *T. marginata* found at the periphery of Kardamili, southernmost Peloponnesus, Greece.

Influence of tortoise species, gender, size, and weight on *H. aegyptium* load

Comparing the tortoise species, *Testudo marginata* were significantly larger in all body measurements than *T. hermanni* (length: $Z = -5.421$, $P << 0.01$; width: $Z = -4.827$, $P << 0.01$; weight: $Z = -5.09$, $P << 0.01$). Males of *T. marginata* have significantly wider carapaces than females, but no differences were found in weight and condition index (Table 4).

Mann–Whitney tests showed significant differences in numbers of adult of both sexes *Hyalomma aegyptium* parasitized on *Testudo marginata* and *T. hermanni* (males ticks: $Z = -5.911$, $P << 0.01$; females ticks: $Z = -6.02$, $P << 0.01$), in both cases *T. marginata* was significantly more infested than *T. hermanni* by both sexes of *Hyalomma aegyptium*.

The number of males, females and nymphs of *Hyalomma aegyptium* did not differ between males and females of *T. marginata* (Table 4). Only the number of males of *Hyalomma aegyptium* was correlated with body measurements (Table 5). Also the results of ANCOVA did not show differences in number of ticks between tortoise sexes (males: $F_{1,23} = 0.377$, $P = 0.545$; females: $F_{1,23} = 0.67$, $P = 0.422$, nymphs: $F_{1,23} = 0.083$, $P = 0.776$).

Table 4 Comparison of body measurements, condition index and numbers of ticks between males and females of *T. marginata*. Results of Mann–Whitney tests. Significant *P*-values after Bonferroni corrections marked by asterisk

	Z	*P*-level
Straight carapace length	–1.976	0.048
Maximum width of carapace	–2.240	0.025*
Weight	–1.449	0.147
Condition index	1.133	0.257
Males	–1.054	0.292
Females	0.738	0.461
Nymphs	–0.369	0.712

Table 5 Spearman's correlation coefficients between *H. aegyptium* and *T. marginata* body measurements including condition index. Significant correlations ($P < 0.05$) marked by asterisk. SCL—straight carapace length, CW—carapace width, W—weight, CI—condition index

	r_s	*P*-level
males & SCL	0.489	0.011*
males & CW	0.473	0.015*
males & W	0.422	0.032*
males & CI	0.043	0.834
females & SCL	0.119	0.562
females & CW	0.066	0.748
females & W	0.073	0.725
females & CI	0.097	0.637
nymphs & SCL	0.050	0.808
nymphs & CW	–0.026	0.898
nymphs & W	0.015	0.943
nymphs & CI	–0.261	0.198

Discussion

Tick species parasitizing tortoises of the genus *Testudo*

Barnard and Durden (2000) have reviewed ten species of ticks (three of the Argasidae, seven of the Ixodidae), parasitizing west Palaearctic tortoises of the genus *Testudo*. Only ixodid ticks were found on tortoises during our study. We confirmed the strong dominance of *H. aegyptium* among adult ticks parasitizing *Testudo*, which is in agreement with previous studies (e.g. Robbins et al. 1998; Leontyeva and Kolonin 2002). Also among the immature stages, *H. aegyptium* larvae and nymphs were the most commonly recorded ticks.

In the Balkans, earlier researchers detected only *H. aegyptium* (Drensky 1955; Černý 1959; Haitlinger 1993) on Bulgarian and Greek tortoises. Moreover, records of *H. syriacum* Koch, 1844 (Buresh and Drensky 1932) actually represent *H. aegyptium*, of which that name is a synonym. Recently, Zlatanova (1991) reported from Bulgarian tortoises not only *H. aegyptium*, but also three further tick species, namely *Haemaphysalis erinacei taurica* Pospelova-Shtrom, 1940 (one female), *Hyalomma anatolicum excavatum* Koch, 1844 (one male), and *Hyalomma marginatum marginatum* Koch, 1844 [referred as *H. plumbeum* (Panzer, 1795) (three females)].

Considerable portion of ticks that we have collected from Bulgarian tortoises were larvae and nymphs of *H. sulcata*. Adults *H. sulcata* are known to parasitize a wide variety of wild and domestic mammals, whereas birds, lizards, snakes, tortoises and rodents represent the chief hosts of immature stages of this tick (Pomerancev 1950; Hoogstraal et al. 1981; Barnard and Durden 2000). In the Mediterranean region adult

H. sulcata feed predominantly on ungulates, whereas immature stages feed on reptiles, mostly on lizards (e.g. Drensky 1955; Černý 1959; Estrada-Peña et al. 2004.)

Four specimens of *R. sanguineus* collected from two *T. marginata* at the southernmost part of Peloponnesus peninsula, Greece, were, apart from *H. aegyptium*, the only other adult ticks we found on tortoises. This originally African but now cosmopolitan tick species parasitizes a variety of carnivores, other mammals, and larger-sized birds. Also humans and domestic ungulates are infrequently infested (Hoogstraal et al. 1981). Our finding is the first record of this species from a reptilian host. *R. sanguineus* occurs in all areas of the Mediterranean region, but hosts other than dogs are only infested when dogs are present to maintain a population of the tick (Estrada-Peña et al. 2004).

Haemaphysalis inermis was represented by only two specimens (one larva and one nymph), collected at Melnik from a single *T. hermanni*. On reptiles, this tick species was reported from the lizard *Lacerta viridis* (Laurenti, 1768) (Řeháček et al. 1961; Lác et al. 1972) and from unspecified lizards used as hosts under laboratory conditions (Pomerancev 1950). Our records then represent the first finding of *H. inermis* on tortoises.

The host specificity of ticks is a variable phenomenon and individual tick species differ in the extent of their host spectrum (Sonenshine 1993). Accidental records from various hosts outside the normal host range are frequent in the literature. Findings on tortoises of *R. sanguineus* and *H. inermis* (this study), *Haemaphysalis erinacei taurica*, *Hyalomma anatolicum excavatum*, *Hyalomma m. marginatum* (Zlatanova 1991) and *Ixodes ricinus* (Linnaeus 1758) (Leontyeva and Kolonin 2002) probably represent also accidental records without biological/epidemiological significance.

Host preferences of *H. aegyptium*

Testudo tortoises represent the chief hosts of *H. aegyptium*, despite the fact that also some other reptiles, birds and mammals can be parasitized by this tick species (e.g. Hoogstraal and Kaiser 1960; Barnard and Durden 2000; Burridge and Simmons 2003). Therefore, distribution of this tick species in northern Africa, southern Europe and western Asia (Kolonin 1983) is limited to areas with occurrence of land tortoises of the genus *Testudo*.

This genus contains traditionally five species (Loveridge and Williams 1957; Ernst and Barbour 1989; Fritz and Cheylan 2001). In Europe, simultaneous occurrence of two *Testudo* species is a common trait in the Balkans, where *T. hermanni* lives either alone, or together with *T. marginata* or *T. graeca*. The occurrence of two tortoise species in a particular area represents an interesting model that enables studies focused on tick host preferences. However, this phenomenon was not even addressed by most authors, which either examined only one tortoise species (e.g. Hoogstraal and Kaiser 1960; Kaiser and Hoogstraal 1963; Petney and Al-Yaman 1985; Robbins et al. 1998) or did not specify the host species (e.g. Drensky 1955; Černý 1959; Haitlinger 1993).

Hailey et al. (1988) reported that *T. graeca* carried more *H. aegyptium* (intensity of infestation 2.1–3.5) than *T. hermanni* (intensity of infestation 0.6–2) at the same collection sites in Greece. They explained this trait by different activity level and different habitat preferences of both *Testudo* species. Zlatanova

(1991) reported *H. aegyptium* both from *T. graeca* and *T. hermanni*, however without details.

According to our results achieved in the Balkan countries, *T. graeca* and *T. marginata* represent there the principal hosts of *H. aegyptium*. *T. hermanni* serves only as an alternative host in the areas within or nearby the range of either *T. graeca* or *T. marginata*. These assumptions correspond with our failure to find *H. aegyptium* ticks on *T. hermanni* at Pelješac Peninsula in Croatia as well as with the absence of records of *H. aegyptium* in Mediterranean areas of Spain, France, Italy and western Balkan, where *T. hermanni* represents the only native tortoise (Iverson 1992).

Matsumoto et al. (2004) reported, without further details, about finding of a single male of *H. aegyptium* on tortoise *T. hermanni* at Corsica, which represents the first record of this species for the Corsica island. Spreading of exotic ticks on imported reptiles is common in recent years (e.g. Burridge 2001; Burridge and Simmons 2003; Pietzsch et al. 2006). Active reproductive colonies of *H. aegyptium* introduced on *T. graeca* from Northern Africa were reported from Alicante, Spain and possibly also from Italy (Brotóns and Estrada-Peña 2004). Then, isolated record from Corsica might represent also only introduced tick specimen and the occurrence of *H. aegyptium* at Corsica needs thorough investigation.

The present study confirms our experience from previous fieldwork, that different *Testudo* species living in the same site have a different attractiveness for *H. aegyptium*. *Testudo marginata* is evidently the preferred host for adult stages and nymphs of *H. aegyptium* at Greek localities, where it lives together with *T. hermanni* (Table 3). Observed differences in numbers of ticks parasitizing *T. marginata* and *T. hermanni* are so important that they cannot be explained simply by the different activity level, habitat preferences or by differences in size of tortoises.

Based on recent phylogenetic studies, *Testudo graeca* and *T. marginata* represent closely related species, distant to *T. hermanni* (Fritz et al. 2005). The co-evolution of *H. aegyptium* with the *Testudo graeca-marginata* clade might explain the host preferences of this tick, as well as its absence in areas with distribution of *T. hermanni* only.

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