Parasitol Res (2014) 113:4587–4599
DOI 10.1007/s00436-014-4149-5

ORIGINAL PAPER

Blood parasites in reptiles imported to Germany

Halla Ursula · Korbel Rüdiger · Mutschmann Frank · Rinder Monika

Received: 8 September 2014 / Accepted: 23 September 2014 / Published online: 18 October 2014
© Springer-Verlag Berlin Heidelberg 2014

Abstract Though international trade is increasing, the significance of imported reptiles as carriers of pathogens with relevance to animal and human health is largely unknown. Reptiles imported to Germany were therefore investigated for blood parasites using light microscopy, and the detected parasites were morphologically characterized. Four hundred ten reptiles belonging to 17 species originating from 11 Asian, South American and African countries were included. Parasites were detected in 117 (29 %) of individual reptiles and in 12 species. Haemococcidea (*Haemogregarina*, *Hepatozoon*, *Schellackia*) were found in 84 % of snakes (*Python regius*, *Corallus caninus*), 20 % of lizards (*Acanthocercus atricollis*, *Agama agama*, *Kinyongia fischeri*, *Gekko gecko*) and 50 % of turtles (*Pelusios castaneus*). Infections with Hematozoea (*Plasmodium*, *Sauroplasma*) were detected in 14 % of lizards (*Acanthocercus atricollis*, *Agama agama*, *Agama mwanzae*, *K. fischeri*, *Furcifer pardalis*, *Xenagama batillifera*, *Acanthosaura capra*, *Physignathus cocincinus*), while those with Kinetoplastea (*Trypanosoma*) were found in 9 % of snakes (*Python regius*, *Corallus caninus*) and 25 % of lizards (*K. fischeri*, *Acanthosaura capra*, *G. gecko*). Nematoda including filarial larvae parasitized in 10 % of lizards (*Agama agama*, *Agama mwanzae*, *K. fischeri*, *Fu. pardalis*, *Physignathus cocincinus*). Light microscopy mostly allowed diagnosis of the parasites' genus, while species identification was not possible because of limited morphological characteristics available for parasitic developmental stages. The investigation revealed a high percentage of imported reptiles being carriers of parasites while possible vectors and pathogenicity are largely unknown so far. The spreading of haemoparasites thus represents an incalculable risk for pet reptiles, native herpetofauna and even human beings.

Keywords Blood parasites · Epidemiology · Reptiles · International wildlife trade · Morphology · Light microscopy

Introduction

Infections with blood parasites are very common in reptiles all over the globe (Ewers 1968; Telford 1993; de Thoisy et al. 2000; Smallridge and Bull 2000; Mihalca et al. 2008; Pereira et al. 2010; Cook et al. 2010). Especially, not only apicomplexan parasites, including Coccidea and Haematozoea as well as Kinetoplastea (Tenter and Schnieder 2006), have been recorded to occur in reptiles, but also filariae have frequently been found in snakes, chelonians and lizards (Hull and Camin 1960; Ayala 1978; Schall and Marghoob 1995; Telford 1995c; Bolette 1998; Mutschmann 2002; Sloboda et al. 2007). Common to these parasites is that they need vectors (e. g., arthropods or leeches) for their development or transmission.

Recently, vector-borne diseases have been recognized as an enormous global risk regarding the health of humans and animals. Spread and establishment of endemic populations of formerly exotic pathogens and their vectors were already documented to occur and have been assumed to be caused not only by climate change, but also by expanding civilization and increasing international trade. It has to be expected that this problem will become even more serious in the future (Harrus and Baneth 2005; Karesh et al. 2005).

H. Ursula (✉) · K. Rüdiger · R. Monika
Klinik Für Vögel, Reptilien, Amphibien Und Zierfische, Zentrum Für Klinische Tiermedizin, Tierärztliche Fakultät, Ludwig-Maximilians Universität München, Sonnenstr. 18, 85764 Oberschleißheim, Germany
e-mail: Ursula.Halla@vogelklinik.vetmed.uni-muenchen.de

M. Frank
Exomed—Institut Für Veterinärmedizinische Betreuung Niederer Wirbeltiere Und Exoten, Erich-Kurz-Str. 7, 10319 Berlin, Germany

Examples are *Dirofilaria repens* causing cutaneous dirofilariosis (Keller et al. 2007; Auer and Susani 2008; Sassnau et al. 2009) and *Dirofilaria immitis*, the heartworm in dogs (Czajka et al. 2014; Kronefeld et al. 2014) expanding in Europe (Sassnau et al. 2014). In Africa, *Trypanosoma brucei rhodesiense*, the agent of human sleeping sickness, is emerging in previously free countries due to cattle (reservoir) movements (Fèvre et al. 2006). *Plasmodium relictum* was brought to Hawaii by released exotic birds, mainly from Asia. Because a potent vector species, *Culex quinquefasciatus*, had been introduced before and had already established stable populations on the islands, this avian malarial parasite was able to infect also native birds leading to their extinction (van Riper et al. 1986). *Ehrlichia ruminantium*, the agent of heartwater in cattle, a disease with great economic impact, was detected in ticks introduced to the USA by imported reptiles (Burridge et al. 2000).

The international trade with animals is certainly one important cause of the spreading of diseases. With regard to live reptiles, the commercial value of the legal trade to the 27 member states of the European Union was quoted about US$9 billion in 2011. Germany was listed as the largest market for import and export of reptiles in the EU with an import value of US$2.6 billion (UN_Comtrade 2013). It has to be assumed that every year, millions of exotic reptiles, directly taken from the either wild, ranched or bred in captivity, are traded globally in legal and illegal ways. Since these imported animals are generally not examined for blood parasites, the amount of parasites imported simultaneously and, as a consequence, the resulting health risks are not known.

The aim of this study was to investigate reptiles imported to Germany for haemoparasites using light microscopy as a fast and feasible method and to discuss the consequences in relation to epidemiological and zoonotic aspects.

Materials and methods

During December 2010 to January 2012, 410 reptiles belonging to 17 species (including two species of snakes, two species of chelonians and 13 lizard species) from 11 countries (Table 1) imported by two German pet trading companies were examined for the presence of blood parasites. The animals were imported directly from the country of origin to Germany where they were kept in boxes, terraria or flexaria until they were transported to an inland or outland salesman.

From each individual, 0.2 ml blood, but never more than 0.8 % of its body weight, was obtained by ventral tail venipuncture (lizards and snakes) or dorsal tail venipuncture (chelonians). This procedure was permitted according to the German animal welfare regulations by the German authorities (reference number 55.2-1-54-2531.3-57-10). Immediately after collection, the untreated blood was microscopically examined on slides for 2 min for motile parasites such as microfilaria

Table 1 Reptile species as well as number and geographic origin of individuals included in this investigation

Scientific name	Common name	Number	Geographic origin (*n* individuals included)	
Acanthocercus atricollis	Southern tree agama	22	Tanzania	Africa
Agama agama	Common agama	25	Cameroon (3)	
			Tanzania (5)	
			Togo (17)	
Agama mwanzae	Mwanza flat-headed agama	25	Tanzania	
Kinyongia fischeri	Fischer's chameleon	25	Tanzania	
Chamaeleo dilepis	Flap-necked chameleon	26	Tanzania	
Furcifer oustaleti	Oustalet's giant chameleon	25	Madagascar	
Furcifer pardalis	Panther chameleon	25	Madagascar	
Pelusios castaneus	West African mud turtle	16	Togo	
Python regius	Ball python	20	Togo	
Xenagama batillifera	Beaver tailed agama	25	Ethiopia	
Chelonoidis carbonaria	Red-footed tortoise	20	Brazil	South America
Corallus caninus	Emerald tree boa	25	Surinam	
Iguana iguana	Green iguana	35	El Salvador	
Acanthosaura capra	Green pricklenape	25	Vietnam	Asia
Gekko gecko	Tokay gecko	25	Vietnam	
Physignathus cocincinus	Green water dragon	36	Vietnam	
Tiliqua gigas	Blue-tongued skink	10	Indonesia (4)	
			New Guinea (6)	
∑: 17 different species		410	11 countries	

or trypanosomes using the ×400 magnification. Thin blood films were prepared, air-dried, transported to the laboratory in dust-free slide storage boxes and stained according to Pappenheim using standard protocols (Binder et al. 2012).

Stained smears were scanned using a Leitz Aristoplan microscope connected with a Leica camera (Type DC 300F, Leica Microsystems AG, Heerbrugg/CH) adhering to a standardized protocol comprising four steps: (1) Entire smears were examined with ×100 magnification, and (2) four lines of meandering pattern in the monolayer area were investigated using the ×400 magnification. (3) If extracellular parasites were found in step 1, they were quantified by adding the number of parasites in two meander lines at the beginning, two in the monolayer region and two at the end of the smear. (4) In each smear, 2,000 erythrocytes were examined with a ×1000 magnification for intracellular pathogens, and if found, they were counted to determine the percentage of the parasitaemia (Godfrey et al. 1987). Parasite identification was performed based on their morphology as seen with light microscopy and cellular tropism according to Reichenow (1919); Telford (2009) and in individual cases, based on original descriptions cited below. Taxonomic classification was based on Tenter and Schnieder (2006). Measurements of length and width of the parasites were made from photographs using the calibrated Image Tool Software (UTHSCSA Image Tool for Windows Version 3.00).

Results and discussion

Blood parasites were detected in 117 (29 %) out of 410 reptiles of 12 species (Table 2). Most of the parasites were grouped to the protozoan families Haemogregarinidae, Hepatozoidae, Lankesterellidae, Plasmodiidae, Haemohormidiidae and

Table 2 Detection rate and infection intensity of parasites found in reptilian species included in this investigation

Host species	Parasites	*n* infected (% infected)	Host cells infected (mean±SD)	*n* extracellular parasites[a] (mean±SD)
Acanthocercus atricollis	*Schellackia*	5 (23)	5.5 %±11.3	–
	Sauroplasma	5 (23)	2.8 %±3.3	–
Acanthosaura capra	*Plasmodium*	1 (4)	3 %	–
	Trypanosoma	3 (12)	–	12±16.8
Agama agama	*Schellackia*	3 (12)	0.2 %±0.2	–
	Plasmodium	1 (4)	0.3 %	–
	Filaria	2 (8)	–	259±320.3
Agama mwanzae	*Plasmodium*	4 (16)	0.1 %±0.1	–
	Filaria	3 (12)	–	49±31
Physignathus cocincinus	*Plasmodium*	2 (6)	0.2 %±0.04	–
	Filaria	1 (3)	–	2
Xenagama batillifera	*Sauroplasma*	5 (20)	40.8 %±53.4	–
Gekko gecko	*Hepatozoon*	1 (4)	0.2 %	–
	Trypanosoma	11 (44)	–	16±34.9
Tiliqua gigas		0	–	–
Furcifer oustaleti		0	–	–
Furcifer pardalis	*Sauroplasma*	7 (28)	2.9 %±2.9	–
	Filaria	6 (24)	–	89±82.5
Kinyongia fischeri	*Schellackia*	10 (40)	0.7 %±1.2	–
	Plasmodium	5 (20)	0.4 %±0.3	–
	Trypanosoma	5 (20)	–	6±6.2
	Filaria	2 (8)	–	539±700
Corallus caninus	*Hepatozoon*	23 (92)	4.5 %±7.6	–
	Trypanosoma	1 (4)	–	2
Python regius	*Hepatozoon*	15 (75)	1.5 %±2.7	–
	Trypanosoma	3 (15)	–	2±1.2
Chelonoidis carbonaria		0	–	–
Pelusios castaneus	*Haemogregarina*	8 (50)	1.2 %±1.2	–
Chamaeleo dilepis	Virus	18 (69)	Not counted	–
Iguana iguana	Virus	30 (86)	Not counted	–

[a] In six lines of meandering pattern (two at the beginning, two in the monolayer region and two at the end of the smear)

Trypanosomatidae. In four host species (*Agama agama* (Linnaeus, 1758), *Agama mwanzae* (Loveridge, 1923), *Kinyongia fischeri* (Reichenow, 1887), *Furcifer pardalis* (Cuvier, 1829)), metazoan parasites diagnosed as Filaroidea were detected. During this investigation, blood parasites were not found in the reptilian species *Chamaeleo dilepis* (Leach, 1819), *Furcifer oustaleti* (Mocquard, 1894), *Iguana iguana* (Linnaeus, 1758), *Tiliqua gigas* (Schneider, 1801) and *Chelonoidis carbonaria* (Spix, 1824).

Intracellular apicomplexan parasites of the haemogregarine complex (Haemogregarinidae [*Haemogregarina*] and Hepatozoidae [*Hepatozoon*]) and of the families Lankesterellidae (*Schellackia*), Plasmodiidae (*Plasmodium*) and Haemohormidiidae (*Sauroplasma*) were detected in 8 individual turtles, 38 snakes and 49 lizards. Extracellular parasites of the Kinetoplastea, family Trypanosomatidae (*Trypanosoma*), were found in 4 snakes and 19 lizards, while filarial larvae (Nematoda) were detected in 14 lizards (Table 3).

Haemogregarines

Parasites of the haemogregarine complex (so-called haemogregarines) were found with high prevalence in two snakes and one chelonian species and with low prevalence in one lizard species (Table 2). On average, the parasitaemia was low to medium ranging between 0.1 and 5.5 % infected erythrocytes. The haemogregarines are identified based on their intraerythrocytic elongate to bent cell shape and a prominent dark nucleus. The families and genera within the haemogregarine complex cannot be distinguished based on their blood stages in the reptiles alone, but they are classified in accordance with their developmental pattern (sporogony) in the invertebrate hosts (Telford 2009). According to Siddall (1995), we designated them as *Haemogregarina* when found in chelonians and as *Hepatozoon* when they occurred in snakes or lizards.

Half (50 %) of the examined individuals of the West African mud turtle *Pelusios castaneus* (Schweigger, 1812) imported from Togo were parasitized by *Haemogregarina* (Table 2). At present, only one haemogregarine species has been described to occur in turtle species of the Pelomedusidae, which is *Haemogregarina pelusiensi* Pienaar 1962 found in *Pelusios sinuatus* (Paperna 1989). This parasite resembles in size and morphology that found in *Pelusios castaneus. Placobdella multistriata*, an African turtle leech, has been shown to be a vector (Paperna 1989). Since the geographic distribution of *Placobdella multistriata* (Oosthuizen 1979) overlaps with that of *Pelusios castaneus* (Klein 2006), the haemogregarines detected in the present study might belong to the species *Ha. pelusiensi*.

In both snake species included in this investigation, the ball python *Python regius* (Shaw, 1802) and the emerald tree boa *Corallus caninus* (Linnaeus, 1758), haemogregarines, designated here as *Hepatozoon*, were found with very high prevalence. In *Python regius*, 15 of 20 individuals (75 %) exported as ranched juvenile animals from Togo were infected. Sizes of the parasites were within the size range of *Hepatozoon ayorgbor* (Sloboda et al. 2007) which had been found in ball pythons from Ghana (Sloboda 2008). The *Hepatozoon* found in this study might therefore also belong to this species. A mean of 1.5 % (maximum 9.3 %) of the erythrocytes of *Python regius* was infected.

In *Corallus caninus*, 23 (92 %) wild-caught individuals imported from Surinam showed *Hepatozoon* infection with a parasitaemia ranging from 0.1 to 26 %. The only description so far of haemogregarines found in emerald tree boas and resembling in morphological details to this *Hepatozoon* species was given by Sloboda (2008), but this author did not use a species name. It has to be noted that for accurate identification of *Hepatozoon* found in *Python regius* and *Corallus caninus*, knowledge of morphology of developmental stages in the vectors or, alternatively, molecular genetic information would be necessary. At present, PCR and sequencing are used to characterize the haemogregarines detected in this study (Strütt et al. in preparation).

Interestingly, in only one wild-caught tokay gecko (*Gekko gecko* (Linnaeus, 1758)) out of 25 (4 %) imported from Vietnam, infection with *Hepatozoon* was found. The parasites found here had the same morphological characteristics and effects on the host cell (hypertrophy, deformation and karyolysis) (Fig. 1) as *Hepatozoon mesnili* described in 1936 already from *G. gecko* originating from the same country, Vietnam (Robin 1936).

Coccidea

Parasites of the order Eimeriida, identified as *Schellackia*, were found in African Agamidae and in Chamaeleonidae: in *Agama agama* imported from Togo and Cameroon (interestingly not in the individuals from Tanzania), in *Acanthocercus atricollis* (Smith, 1849) from Tanzania and in *K. fischeri* from Tanzania. The identification of the genus *Schellackia* (Lankesterellidae) was based on the detection of intraerythrocytic encapsulated sporozoites containing one or two homogeneous refractile bodies on each side of the nucleus, which appears usually as a chromatin band (Fig. 2). Species diagnosis, however, is only possible when developmental stages from the host gut and biological data are also taken into consideration (Bristovetzky and Paperna 1990; Telford 1993). In the chameleon *K. fischeri*, the highest prevalence with ten (40 %) animals infected by *Schellackia* but, at the same time, a low parasitaemia (only 0.1–3.6 %) was detected. In contrast, in the southern tree agama *Acanthocercus atricollis*, only 5 out of 25 individuals were found to be parasitized, but up to 25.7 % of the red blood cells were carrying *Schellackia* (with

Table 3 Morphology of the parasites found in this investigation

Parasite	Host	L × W (mean±SD) (μm)	L × W (range) (μm)	Number	Shape	Nucleus	Colour of cytoplasm (Pappenheim)	Characteristics
Haemogregarina	*Pelusios castaneus*	23.9±5.2×4.8±1.1	10.9-33.4×3.1-6.9	42	Blunt-elongate, bent	Dark-spotted	Violet-pale blue	
Hepatozoon	*Python regius*	13.5±2.6×3.4±0.7	9.6-17.5×2.4-5.2	94	Elongate	Good visible	Violet-blue	
Hepatozoon	*Corallus caninus*	13.8±1.8×3.9±1	8.3-18.7×1.4-6.2	105	Blunt-elongate, bent	Thinner-equal thick, central-polar	Violet-blue	
Hepatozoon	*Gekko gecko*	31.6±2.4×7.8±1.2	27.6-37.3×5.4-9.5	15	Elongate-bent	Spotted, short-long	Bright-violet	Massive morphological changes of host cell
Schellackia	*Acanthocercus atricollis*	11.5±0.9×3.6±0.4	9,2-13,8×2,5-4,3	63	Elongate	Accumulation of chromatin granules	Bright	Two refractile bodies
Schellackia	*Agama agama*	10.3±1.3×3.5±0.5	9.1-13.3×3-4.3	10	Elongate	Accumulation of chromatin granules	Bright	Two refractile bodies
Schellackia	*Kinyongia fischeri*	12±1.2×2.7±0.5	9-14.7×2-4.3	53	Elongate	Accumulation of chromatin granules	Bright	Two refractile bodies
Sauroplasma	*Acanthocercus atricollis*	2.3±0.5×2.1±0.5	1.1-3.4×1.4-3	25	Round or irregular	Not visible-marginal	White-red	
Sauroplasma	*Furcifer pardalis*	2.8±0.8×2.5±0.81	1.4-4.4×1-4.3	43	Round or irregular	Not visible-marginal	White-red	
Sauroplasma	*Xenagama batillifera*	2.9±0.7×2.5±0.7	1.6-5×1.1-4.3	69	Round or irregular	Not visible-marginal	White-red	
Plasmodium	*Agama agama*	11.1±1.1×4.9±1.2	10-13×3.8-7.4	7	Oval to spindle shaped	Not visible	Bluish	Haemozoin granules
Plasmodium	*Agama mwanzae*	8.0±1.5×5.5±2	6.1-11.2×3.4-9.3	9	Oval to spindle shaped	Not visible	Bluish	Haemozoin granules
Plasmodium	*Kinyongia fischeri*	13.9±3×4.9±1.1	7.1-19.2×2.7-7.4	42	Ends acuminate	Not visible	Bluish	Haemozoin granules
Plasmodium	*Acanthosaura capra*	4.4±1×2.5±0.5	3.3-5.9×1.7-3.2	22	Round or irregular	Few marginal granules-not visible	Bluish	Bright centre, no or few haemozoin granules
Plasmodium	*Physignathus cocincinus*	6±1.1×4.3±0.6	4.3-8.7×3.3-5.2	17	Oval or round	Not visible	Bright-violet	Few haemozoin granules
Trypanosoma	*Kinyongia fischeri*	37.7±5.3×7.6±2.5	29-50.9×4.3-14.7	23	Leaf-shaped	Not visible-oval bright area	Bluish	Visible undulating membrane, free flagellum 20.6-27.2 (23.9±2.1)[a]
Trypanosoma	*Python regius*	38.5±9.6×5±1.9	31.4-53.5×2.4-7	5	Leaf-shaped	Oval bright area	Bluish, violet granules	Visible undulating membrane, short free flagellum 21.9-34.7 (27.4±6.6)[a]
Trypanosoma	*Corallus caninus*	41.3±7.2×6.0±1.3	36.2-46.4×5.1-6.9	2	Leaf-shaped	Oval bright area	Violet	Prominent undulating membrane 28.6-29.2 (28.9±0.4)[a]
Trypanosoma	*Acanthosaura capra*	47.8±6.2×4.8±1.2	40-56.9×3.1-6.7	15	Slender, elongate	Not visible	Violet, dark stained	Visible undulating membrane, free flagellum kinetoplast not visible
Trypanosoma	*Gekko gecko*	50.7±3.3×9.3±1.6	42.3-58.7×7-12.9	22	Leaf-shaped to elongate	Prominent bright area	Violet	Visible undulating membrane, free flagellum 24.5-39.8 (33.6±6.1)[a]
Filaria	*Agama agama*	82.9±6.6×5.8±0.8	69.9-92.9×4.5-7.7	40	Anterior end blunt, posterior end tapering			Sheath loose around entire body length
Filaria	*Agama mwanzae*	162.6±11.3×3.6±0.6	140.1-184×2.2-4.4	18	Anterior end blunt, posterior end tapering			Sheath loose around entire body length , posterior end bent over
Filaria	*Kinyongia fischeri*	119.7±8.1×5.1±0.9	102.8-129.9×3.6-7.2	32	Anterior end blunt, posterior end tapering			Sheath loose at posterior end
Filaria	*Furcifer pardalis*	116.6±13.9×5.3±0.91	87.7-145.5×3.6-9.2	104	Anterior end blunt, posterior end tapering			Sheath loose at anterior or posterior end
Filaria	*Physignathus cocincinus*	79.5±14.5×5.5±0.4	59.8-104.6×4.7-6	10	Anterior end blunt and broader, posterior end tapering			Sheath not loose, prominent red area

Table 3 (continued)

Parasite	Host	L × W (mean±SD) (µm)	L × W (range) (µm)	Number	Shape	Nucleus	Colour of cytoplasm (Pappenheim)	Characteristics
Virus	*Chamaeleo dilepis*	1.7±0.7×1.5±0.6	0.8-2.7×0.8-2.5	25	Round		Red	With, or without bluish albuminoid body
Virus	*Iguana iguana*	4.0±0.7×2.4±0.6	2.3-6×0.9-3.7	66	Oval-rectangular		White	No albuminoid body

[a] Distance between kinetoplast and anterior body end: range (mean±SD) (µm)

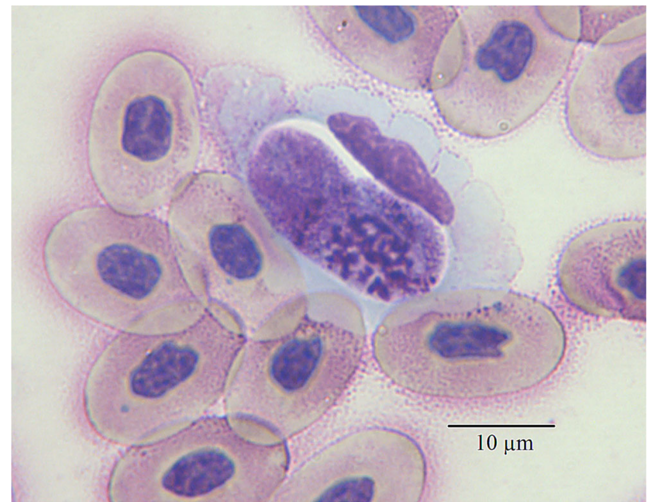

Fig. 1 *Hepatozoon* in tokay gecko (*Gekko gecko*), Pappenheim stain, ×1,000. Note hypertrophy and morphological changes of the host cell (erythrocyte)

very high intraspecific variation in parasitaemia levels, however, as shown in Table 2). It is noteworthy that a species named *Schellackia agamae* (Laveran and Petitt, 1909) was described in *Agama agama* from the Central African Republic (Rogier 1977). The sizes of sporozoites found in the Agamidae in this study were very similar, but the geographical origin of hosts was different. The three infected *Agama agama* in this study came from Togo and Cameroon and *Acanthocercus atricollis* originated from Tanzania. While the parasites detected in *Agama agama* might belong to the species *Sc. agamae*, the very high host specificity described for *Schellackia* sp. (Telford 2009) argues against a classification as *Sc. agamae* of the parasites detected in the other host species (*Acanthocercus atricollis*). Bristovetzky and Paperna (1990) found *Schellackia* in *Agama stellio* from Jerusalem and

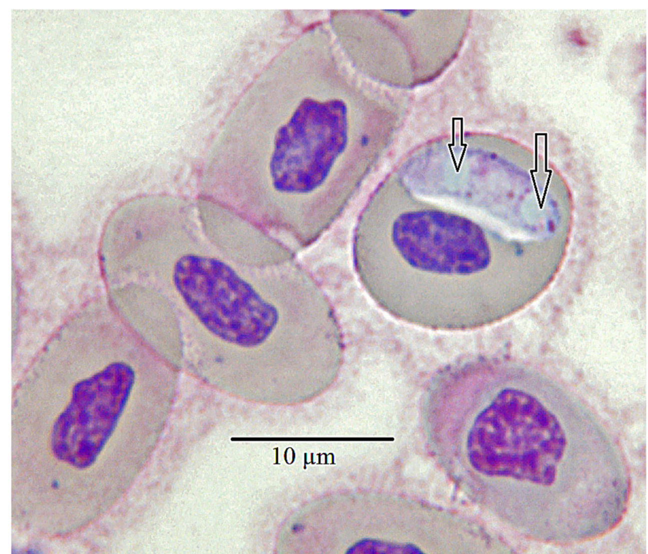

Fig. 2 *Schellackia* in southern tree agama (*Acanthocercus atricollis*), Pappenheim stain, ×1,000. Note the two refractile bodies (➔)

named it *Sc.* cf. *agamae*, because of the incertitude having found *Sc. agamae*.

In the study presented here, parasites of the genus *Schellackia* have been described for the first time in *K. fischeri*.

Plasmodium

The occurrence of intraerythrocytic merozoites, multinucleated meronts and, generally, pigment (haemozoin)-producing gametocytes is a characteristic for *Plasmodium*. Malarial parasites were detected in erythrocytes from African lizards (*Agama agama*, *Agama mwanzae*, *K. fischeri*) and from Asia (*Physignathus cocincinus*, *Acanthosaura capra* Günther, 1861). Interestingly, plasmodia had the lowest parasitaemia of all haemoparasites found in this investigation (0.1–3 %). To our knowledge, this is the first description of *Plasmodium* in *Physignathus cocincinus*. These parasites were morphologically most similar to *Plasmodium volans* Telford, 1995, from a Philippine agamid lizard (Telford 1995b). There are also no records of plasmodiids in *Acanthosaura capra* so far. In order to confirm conspecificity with *Plasmodium* species described already or to designate the parasites detected in this investigation as new species, further knowledge of morphology, biology including life cycle in vertebrate and insect hosts or molecular genomic data would be necessary.

Similarly, at present, records of *Plasmodium* parasites in *Agama mwanzae* from Tanzania/East Africa do not yet exist. The morphological characteristics of these *Plasmodium* spp. being small with irregular shape corresponded most to *Plasmodium robinsoni* Brygoo, 1962, found in a closely related *Agama* species, *Agama agama*, originating from Kenya (East Africa) (Dipeolu and Mutinga 1989). Since the number of parasites and of differing developmental stages found in the present investigation was very small, unequivocal identification of these *Plasmodium* parasites was not possible.

Plasmodium was also detected in a single *Agama agama* imported from Togo/West Africa. These parasites might belong to the species *Plasmodium agamae* Wenyon, 1909, based on morphological similarities and because this parasite was described to occur in common agama from several localities in Africa (Telford 2009). Pigmented plasmodiids, very similar to *Plasmodium acuminatum* Pringle 1960 (Pringle 1960; Telford 1988), were found in a single Fischer's chameleon (Fig. 3). Eponymous and characteristic for this species are the tapering ends of the intraerythrocytic stages which were also detected in the parasites found here. Unfortunately, we could not compare the length and width of the parasites detected here with those of *Plasmodium acuminatum* because no dimensional data are available. *Plasmodium fischeri* (Ball and Pringle 1965) is another *Plasmodium* species described in the same host species. This *Plasmodium*, however, presented much smaller gametocytes (length × width being 8–11×5–8 μm) (Ball and Pringle 1965) compared to the parasites detected in our study.

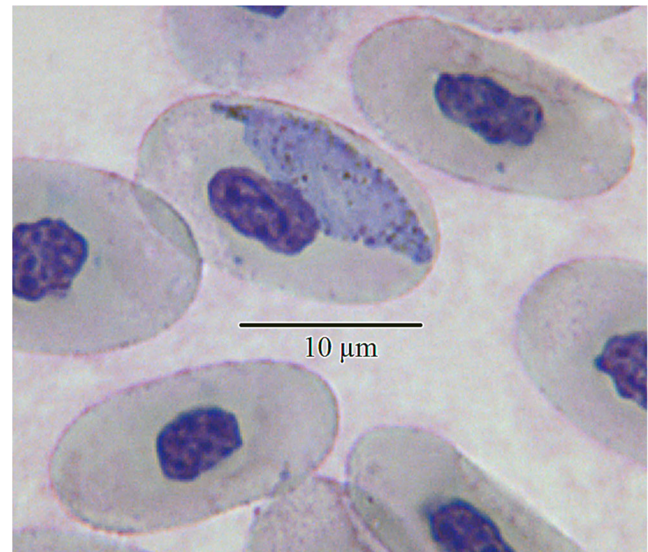

Fig. 3 Plasmodium in Fischer's chameleon (*Kinyongia fischeri*), Pappenheim stain, ×1,000. Note the tapering ends of the parasite

Piroplasms

Piroplasms appear in erythrocytes as small (1.1–5.0×1.0–4.3 μm) inclusions, round to amoeboid in shape (Lainson et al. 1971), containing granules of chromatin associated with a vacuole (Telford 2009). Because of their small size and vacuole-like look, an infection with *Sauroplasma* sp. (occurring in lizards) or *Serpentoplasma* sp. (occurring in snakes) can easily be overlooked or mistaken as artefacts or infections with bacterial organisms or virus particles and, of course, also vice versa (Telford 2009). Piroplasms were found in this study in lizards only and with low to very high degrees of parasitaemia. In *Xenagama batillifera* Vaillant, 1882, imported from Ethiopia, a mean of 40.8 % of the red blood cells was infected by *Sauroplasma* sp. Two animals even showed 99 % parasitaemia (Fig. 4). This was in contrast to previously described cases, where maximum infection rates of erythrocytes of 56 % were found (Du Toit 1937; Pienaar 1962). *Sauroplasma* spp. have not been described previously from the host species *X. batillifera*, and parasite species determination was thus not possible. Whether the *Sauroplasma* detected in the present study in *Fu. pardalis* imported from Madagascar is conspecific with *Sauroplasma* described by Brygoo (1963a) in *Chamaeleo verrucosus* from Madagascar cannot be decided without molecular phylogenetic analyses. Telford Jr (2009) found unspecified *Sauroplasma* in Tanzanian Chamaeleonidae and Skinkidae. *Sauroplasma* parasites were also described in several Asian lizard species, among them *Physignathus cocincinus* Barbour, 1912, and *G. gecko* (Telford 2009). These host species were also included in this

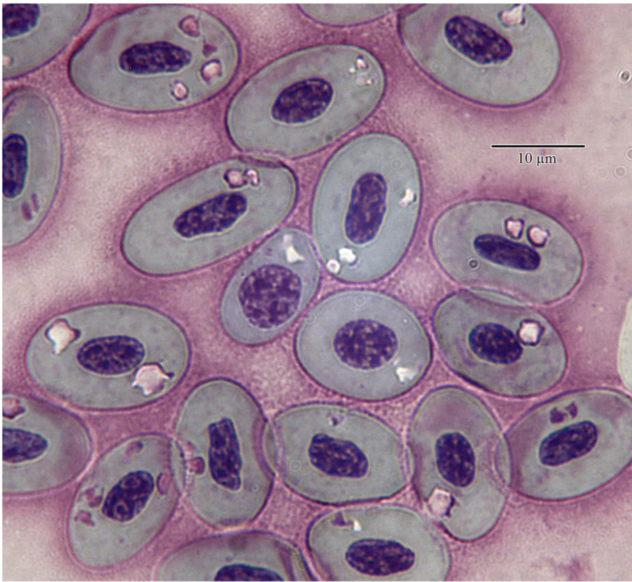

Fig. 4 *Sauroplasma* in beaver tailed agama (*Xenagama batillifera*), Pappenheim stain, ×1,000. Note high parasitaemia

investigation, but intraerythrocytic piroplasmid inclusions have not been detected.

Trypanosoma

Kinetoplastida of the genus *Trypanosoma* was found in lizards from Africa and Asia and in both examined snake species (*Python regius* and *Corallus caninus*) from Surinam and Togo. *Trypanosoma* spp. are motile extracellular leaf-shaped to elongate organisms possessing an undulating membrane and often a free flagellum. In the present investigation, one *G. gecko* showed a very high infection intensity of trypanosomes with 118 parasites found in six meandering lines of a blood smear. Until now, only one species, *Trypanosoma gekkonis* Telford, 1995, has been described in this host species from Thailand (Telford 1995c), and kinetoplastid organisms found here resembled them in morphology and size.

In ball pythons from Ghana, trypanosomes morphologically and genetically very similar to *Trypanosoma varani* from Sudanese monitor lizards (*Varanus niloticus*) were detected (Sato et al. 2009). Based on the morphology and the measurements performed here, the trypanosomes found in *Python regius* from Togo seemed to be nearly identical to the parasites described by Sato et al. (2009) and were tentatively determined as *Tr.* cf. *varani*.

With regard to the trypanosomatids detected in *Corallus caninus* from Surinam, descriptions of morphologically similar parasites of snakes of the family Boidae could not be found. *Trypanosoma alamantae* and *Trypanosoma constrictor* occurring in South American Boidae (Pessôa and Fleury 1969) were much larger, while *Trypanosoma hogei*, described in the snake species *Rhachidelus brazili* (family Colubridae) (Pessôa 1968), was morphologically very similar to the trypanosomes detected in *Corallus caninus*. With 36.5–46.5-µm length × 7–9-µm width, *Tr. hogei* was slightly broader than trypanosomes found in the emerald tree boa examined here. The trypanosomes detected in *Corallus caninus*, however, did resemble *Tr. hogei* not only in size but also in the occurrence of pointed ends, the position of the kinetoplast (26.5–31.5-µm distance to the anterior end of the parasites) and the prominent undulating membrane (Fig. 5). Without further investigations, it remains uncertain, whether the same *Trypanosoma* species is infecting two different families of snakes.

A very slender and elongate *Trypanosoma* form, which could not be attributed to a species, was found in green pricklenape agamas from Vietnam (Fig. 6). In addition, trypanosomes were found in the Fischer's chameleons which were much larger in size than *Trypanosoma therizienii*, a species described in Madagascan chameleons (Brygoo 1963b). We therefore also failed to identify the trypanosome species infecting *K. fischeri*.

Filarioidea

Filarial larvae (microfilariae), extracellular blood parasites, were found in huge numbers in the blood of 14 individuals of five lizard species (Tables 2 and 3). In one *K. fischeri*, more than 1,000 larvae were counted in six meandering lines of the blood smear. Microfilariae were found mainly in reptiles imported from Africa and with high parasitaemia (mean 49–539 larvae in six meandering lines of the blood smear, Table 2). Remarkably, only common agamas imported from Cameroon, but not from Togo and Tanzania, were infected with microfilariae. As microfilariae are growing during their development in the blood (Rasheed 1965), accurate identification based solely on their size is not possible. As shown in

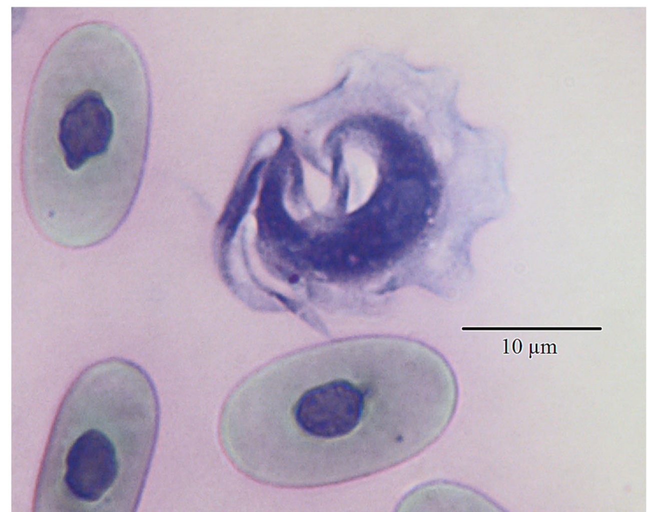

Fig. 5 *Trypanosoma* in emerald tree boa (*Corallus caninus*), Pappenheim stain, ×1,000. Note the prominent undulating membrane

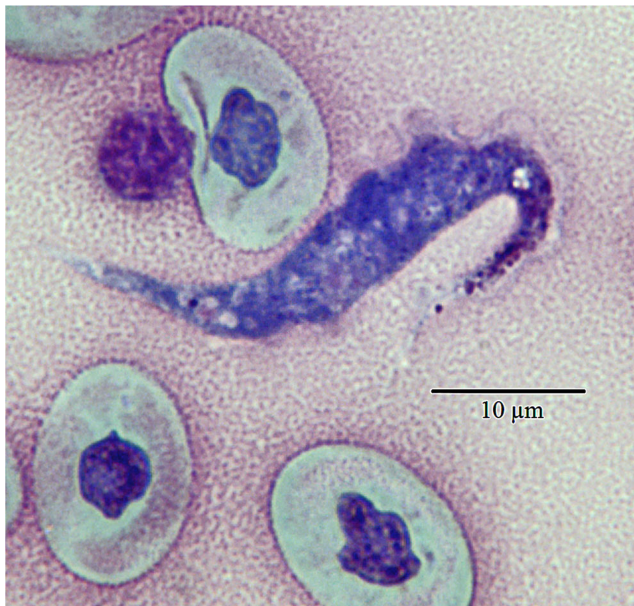

Fig. 6 *Trypanosoma* in green pricklenape (*Acanthosaura capra*), Pappenheim stain, ×1,000

Table 3, a high variation in length has also been detected in our study. Important diagnostic features are therefore the shape of the body and the existence and characteristics of a sheath.

The morphology of microfilariae found in *Agama agama* and in the chameleon species *K. fischeri* and *Fu. pardalis* examined here was similar to *Foleyella* sp. described by Bartlett (1986). Microfilariae of this genus have been morphologically characterized by the presence of a sheath, which is generally loose along the entire length of the body and which varies in shape from oval to elongate, by bluntly rounded anterior and posterior body ends and acuminated tail regions (Bartlett 1986). In addition, species of *Foleyella* seem to have a certain degree of host specificity occurring only in the lizard families Agamidae and Chamaeleonidae (Bartlett 1986). It has even been assumed that all filarial species in chameleons exclusively belong to the genus *Foleyella* (Irizarry-Rovira et al. 2002). Based on the morphology of adult worms Obiamiwe et al. (1995) and Bolette (1998) identified filarial worms parasitizing *K. fischeri* and *Agama agama* as *Foleyella candezei* Fraipont, 1982. Irizarry-Rovira et al. (2002) found *Foleyella furcata* (Linstow, 1899) in *Fu. pardalis*, with the diagnosis also mainly based on morphological characteristics of adult worms. While the microfilariae that we found in *K. fischeri* and *Fu. pardalis* were very similar in appearance (size, sheath only loose at the posterior end), microfilariae from *Agama agama* were much smaller, and their sheaths were loose along the whole length and much longer than the larval body at the posterior end. Thus, it is assumed that the latter microfilariae belong to a different *Foleyella* species. Nevertheless, identification remains vague, until adult worms can be described and compared morphologically.

In *Agama mwanzae* from Tanzania, very long and thin filarial larvae, holding the tail in a hook-like position (Fig. 7), were found. These microfilariae were morphologically similar to *Brygoofilaria* (=*Thamugadia*) *agamae* (Sulahian and Schacher 1968) in *Agama stellio* from Lebanon (Sulahian and Schacher 1968). *Thamugadia* spp. infect lizards (Agamidae, Geckonidae, Lacertidae) in the north of Africa and the Mediterranean region (Bain et al. 1993) but have not yet described to occur in East Africa.

Interestingly, only one reptilian species originating from Asia, *Physignathus cocincinus*, imported from Vietnam, was found being host of filarial larvae (Fig. 8). Morphologically, the microfilariae were similar to *Oswaldofilaria samfordensis* Manzanell, 1982, found in *Physignathus lesueurii* from Australia (Manzanell 1982). They differed, however, in the shape of the sheath. In the microfilariae detected in this study, the sheaths were not longer than the microfilariae in contrast to *O. samfordensis*. A remarkable prominent red area was visible after staining according to Pappenheim. The identity of this structure is not clear to us. Because of the localization within the larvae, it might be the "Innenkörper" described to produce chitinase (Wu et al. 2008). Species identification was not possible because adult worms were not available for analysis.

Viruses

In 69 % of *Chamaeleo dilepis* from Tanzania, red round inclusions associated with an albuminoid body were found in the erythrocytes. These particles were identified as Iridovirus using electron microscopy (Behnke, Heckers unpublished). In addition, in *Ig. iguana* imported from El Salvador, white rectangular vacuole-like intraerythrocytic inclusions were found in 30 out of 35 (86 %) of the animals. They morphologically resembled to lizard erythrocytic virus, which is an Iridovirus (Campbell 2012). Further investigations, however, are necessary to clarify the identity of these inclusion bodies.

Risks of emerging infectious diseases

Haemogregarines require vectors for development. While at least some of the individual haemogregarine species seem to

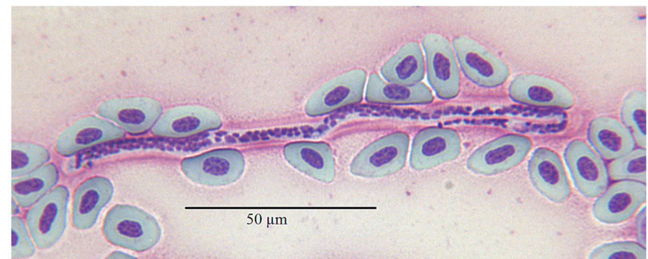

Fig. 7 Microfilaria in mwanza flat-headed agama (*Agama mwanzae*), Pappenheim stain, ×400. Note hook-like tail position

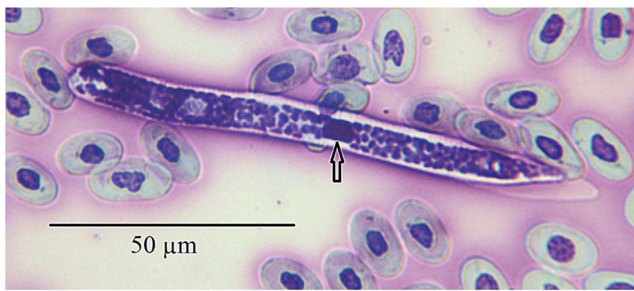

Fig. 8 Microfilaria in green water dragon (*Physignathus cocincinus*), Pappenheim stain, ×400. Note the prominent red area (➔)

be rather specific for their vectors, a huge range of possible arthropod vectors has been identified for these parasites as a group. Lice, fleas, bugs, ticks, mites, mosquitoes, sandflies or tsetse flies may serve as definitive hosts for *Hepatozoon* (Smith 1996). Leeches were found to transmit *Haemogregarina* to aquatic animals (Paperna 1989). The vertebrate host range for haemogregarines is also very large. Mammals, birds, reptiles and amphibians from all over the globe were described to be infected with *Hepatozoon* (Smith 1996). In Europe, haemogregarines were found in lizards from i.e. Portugal, Poland and Scandinavia (Svahn 1974; Majlathova et al. 2010; Roca and Galdon 2010). With regard to individual *Hepatozoon* species, vertebrate host specificity is more restricted but seems not always limited to a single species. In experimental conditions, *Hepatozoon* and *Haemogregarina* from one reptile species were able to infect other species (Siddall and Desser 2001; Sloboda et al. 2007). Sloboda et al. (2007), for example, succeeded in transmitting *He. ayorgbor* from *Python regius* to *Boa constrictor*. Whether the imported *Hepatozoon* and *Haemogregarina* found in the present investigation would be able to establish populations and threaten health of the native fauna or pet reptiles remains unclear because parasite species identity as well as vertebrate and invertebrate (final) host range is unknown. It cannot be excluded that such hosts already occur and that a risk for emerging in wildlife or pet reptile populations exists.

With regard to the haemococcidia of lizards found in this study, it is known that *Schellackia* develops in the gut of the vertebrate host by merogony, gamogony and sporogony. Sporozoites, formed in the oocysts, however, are not excreted with faeces as known in e.g. species of the family Eimeriidae but invade circulating blood cells. Bloodsucking arthropods ingest the parasites during blood meal. It has been documented that the arthropod hosts only serve as mechanical vectors while further development or multiplication of the haemococcidia occurs in the vertebrate host (Reichenow 1919). With regard to the mechanical vector, low host specificity has to be expected. Thus, reptiles can be infected not only by a variety of hematophagous arthropods like ticks, mites and mosquitoes and sandflies depending on local conditions, but also by predation of infected reptilian hosts or, experimentally proven, also vertebrate host tissues (Reichenow 1919; Klein et al. 1988; Bristovetzky and Paperna 1990). Concerning reptiles, haemococcidia have a very high host specificity probably limited to single host species (Telford 2009). The risk of *Schellackia* transmission is regarded as high for conspecific reptilians, especially stock partners, because potential arthropod vectors are nearly ubiquitous and are also common in private pet reptilian husbandry. The risk for humans or for the wildlife herpetofauna, however, is considered to be very low.

The geographic distribution of malarial parasites in reptiles has so far been documented to be very limited and probably depending on the occurrence and biology of the vectors (Ball and Pringle 1965; Schall and Marghoob 1995). The identity of vectors for reptilian plasmodiids is largely unknown so far, and it has been assumed that the host specificity is rather high (Schall 1990). With regard to lizard *Plasmodium* parasites, a vector has been described only for two *Plasmodium* species. Phlebotomine sandflies of the species *Lutzomyia vexator* and *Lutzomyia stewarti* were detected as definitive hosts for *Plasmodium mexicanum* (Ayala and Lee 1970), and the mosquito species *Culex erraticus* has been shown to be a vector for *Plasmodium floridense* (Klein et al. 1987). Reptile malaria was found on all continents (except Europe so far) and in varying climate conditions (Schall 1990). Phlebotominae occurs in regions where the mean temperature per year is above 10 °C (Maier et al. 2001). Thus, it cannot be excluded that vectors for saurian malarial parasites may emerge during climate change. Recently, Phlebotominae was detected in Hessen (Germany), which represents the most northern area of their distribution (Melaun et al. 2014). At present, because of their host specificity and because of probably limited occurrence of vectors, the infection risk for stock partners in housings close to nature and for wildlife in Europe is considered very low. Nevertheless, as shown for birds in Hawaii, malarial parasites have the potential of eradicating naïve species, when, in addition to potent vectors, the parasites are introduced in formerly free areas and find susceptible host populations (van Riper III et al. 1986).

With regard to piroplasms, scientific publications on the genera *Sauroplasma* and *Serpentoplasma* occurring in reptiles are very scarce so far. Infection has been described once for Europe. In Sweden and Denmark, lizards were found to be infected with *Sauroplasma boreale* (Svahn 1976). The natural vectors are not known, but it was assumed that ticks serve as definitive hosts, like in other piroplasmid taxa (*Babesia*, *Theileria*) (Svahn 1976). In local surveys of lizard populations, prevalence of less than 0.1–6 % was found, and it was assumed that they are generally low (Telford 2009). Because of vacuole-like appearance of the parasites in the erythrocytes, infections might sometimes be overlooked. Since their life cycles, vectors, ways of transmission and pathogenicity are essentially unknown, we cannot assess the risk for endemic

herpetofauna or for humans arising from imported piroplasms. Some piroplasmid species occurring in mammals, *Babesia divergens* in cattle and *Babesia microti* in rodents transmitted by *Ixodes ricinus*, are known zoonotic pathogens and were shown to cause diseases, mostly in immunosuppressed patients (Uilenberg 1995; Granström 1997).

Trypanosoma species were found to have no strict host specificity. Trypanosomes from a snake species of the family Viperidae could be successfully transmitted to a snake species of the Colubridae (Viola et al. 2008). Another *Trypanosoma* sp. was transferred between species within the family Chamaeleonidae, but not to other lizard families (Brygoo 1963a; Telford 1995a). In general, experimental infection of unnatural hosts (host switches) resulted in high mortality (Brygoo 1963b). Host switching was shown to occur during evolution of reptilian trypanosomes (Jakes et al. 2001; Viola et al. 2008) and is still taking place at present (Sato et al. 2009). Consequently, when potential vectors like leeches, sandflies or tsetse flies (Telford 1995a) are available, imported trypansomes might lead to a thread for new reptilian hosts. It has also been shown that reptiles may serve as possible reservoirs for *Trypanosoma brucei*, the agent of the African sleeping sickness (Woo and Soltys 1969; Njagu et al. 1999).

Reptilian filariae are transmitted by sandflies and mosquitoes where they develop from the first to the third larvae which are infective for new vertebrate hosts. In general, vector specificity is regarded as rather low (Bain and Babayan 2003), but partial development of filariae connected with disease has been documented in accidentally infected unnatural hosts. Several cases of subcutaneous dirofilariosis in humans, caused by adult worms of *D. repens*, a parasite known from carnivore mammals, have been described (Auer and Susani 2008). It is known that the geographic distribution of culicid mosquitoes (vectors) is much wider than that of the filarial worm that they transmit and that low ambient temperatures limit filarial larval development and thus spread of the nematodes. In Europe, for example, some culicid mosquitoes, able to transmit *D. immitis* and *D. repens*, are known to be widely distributed over the continent (Kronefeld et al. 2014), but only in the last decades, an increasing number of autochthonous cases of canine and human cutaneous dirofilariosis caused by *D. repens* have been documented in Germany, Austria and the Netherlands (Sassnau and Genchi 2013), indicating spread of *D. repens* from endemic areas in the Mediterranean area into the Northern regions of Europe. This spread was attributed to climate change and rising temperatures, and it has been shown that in the last years, temperatures necessary for filarial development can also be achieved in northern regions (Genchi et al. 2011; Sassnau et al. 2014). Endemic occurrence of the canine heartworm *D. immitis* in Germany, however, is at present still subject of ongoing debate. Recent detection of its DNA in German mosquitoes argues for a spread of this parasite into regions of Northern Europe as well (Sassnau et al. 2014).

Some filariae in reptiles have a very wide geographic distribution. The occurrence of *Fo. candezei* was found to range from West and East Africa to the Middle East (Obiamiwe and Iredu 1982). Interestingly, in our study, common agamas from Cameroon, but not from Togo, were found infected with microfilaria. The majority of the *Agama agama* from Togo was wild-caught, but the exact localization of capture was not documented. Environmental conditions (climate, occurrence of vectors) which might explain this difference are therefore not known to us.

In conclusion, our investigation demonstrates that reptiles imported from their country of origin to Germany or other industrial countries often are carriers of haemoparasites. We assume a risk of spreading especially for parasites with low host specificities for both reptilian hosts and vectors. Previous studies revealed that imported reptiles are frequently infested with ticks and mites (Burridge 2001; Kenny et al. 2004; Pietzsch et al. 2006). These ectoparasites might find ideal conditions at the new destination (in the terraria or even in nature) and establish new populations, and they might also contribute to the expansion of haemoparasites. Since a risk for introduction of agents of new diseases to domestic and companion animals, wildlife and humans by reptiles exists, we strongly recommend quarantine combined with examination for pathogens, control of potential vectors and, if necessary, treatment against ectoparasites.

References

Auer H, Susani M (2008) Der erste autochthone Fall einer subkutanen Dirofilariose in Österreich. Wien Klin Wochenschr 120(4):104–106. doi:10.1007/s00508-008-1031-4

Ayala SC (1978) Checklist, host index, and annotated bibliography of *Plasmodium* from reptiles. J Eukaryot Microbiol 25(1):87–100. doi:10.1111/j.1550-7408.1978.tb03874.x

Ayala SC, Lee D (1970) Saurian malaria: development of sporozoites in two species of Phlebotomine sandflies. Science 167(3919):891–892

Bain O, Babayan S (2003) Behaviour of filariae: morphological and anatomical signatures of their life style within the arthropod and vertebrate hosts. Filaria J 2(1):16. doi:10.1186/1475-2883-2-16

Bain O, Wanji S, Petit G, Paperna I, Finkelman S (1993) Filaires Splendidofilariinae de lézards: nouvelles espèces, redescription, cycle chez phlébotome. Syst Parasitol 26(2):97–115. doi:10.1007/bf00009217

Ball GH, Pringle G (1965) *Plasmodium fischeri* n. sp. from *Chamaeleo fischeri*. J Eukaryot Microbiol 12(4):479–482. doi:10.1111/j.1550-7408.1965.tb03246.x

Bartlett CM (1986) The reptilian filarioid genus *Foleyella* Seurat, 1917 (Onchocercidae: Dirofilariinae) and its relationship to other dirofilariine genera. Syst Parasitol 9(1):43–56. doi:10.1007/bf00009896

Binder T, Diem H, Fuchs R, Gutensohn K, Nebe T (2012) Pappenheim stain: description of a hematological standard stain—history, chemistry, procedure, artifacts and problem solutions. Laboratoriumsmedizin, vol 36. doi:10.1515/labmed-2012-0027

Bolette DP (1998) *Foleyella candezei* (Onchocercidae: Dirofilariinae) from a Fischer's chameleon, *Bradypodion fischeri* (Sauria: Chamaeleonidae) with a comment on the synonymy of *F. candezei*. J Parasitol 84(5):1034–1035

Bristovetzky M, Paperna I (1990) Life cycle and transmission of *Schellackia* cf. *agamae*, a parasite of the starred lizard *Agama stellio*. Int J Parasitol 20(7):883–892. doi:10.1016/0020-7519(90)90026-j

Brygoo ER (1963a) Contribution à la connaissance de la parasitologie des caméléons malgaches. Masson, Paris

Brygoo ER (1963b) Hématozoaires de reptiles malgaches. I. *Trypanosoma therezieni* n. sp. parasite des caméléons de Madagascar. Infestation naturelle et expérimentale. Arch Inst Pasteur Madagascar 31:133–141

Burridge MJ (2001) Ticks (Acari: Ixodidae) spread by the international trade in reptiles and their potential roles in dissemination of diseases. Bull Entomol Res 91(1):3–23

Burridge MJ, Simmons LA, Allan SA (2000) Introduction of potential heartwater vectors and other exotic ticks into Florida on imported reptiles. J Parasitol 86(4):700–704

Campbell TW (2012) Hematology of reptiles. In: Thrall MA, Weiser G, Allison R, Campbell TW (eds) Veterinary hematology and clinical chemistry, 2nd edn. Wiley, New York, pp 277–297

Cook CA, Smit NJ, Davies AJ (2010) Hemoproteids (Apicomplexa: Haemoproteidae) from South African tortoises (Cryptodira: Testudinidae). J Parasitol 96(6):1168–1172. doi:10.1645/ge-2527.1

Czajka C, Becker N, Jöst H, Poppert S, Schmidt-Chanasit J, Krüger A, Tannich E (2014) Stable transmission of *Dirofilaria repens* nematodes, northern Germany. Emerg Infect Dis 20(2):329

de Thoisy B, Michel JC, Vogel I, Vie JC (2000) A survey of hemoparasite infections in free-ranging mammals and reptiles in French Guiana. J Parasitol 86(5):1035–1040

Dipeolu OO, Mutinga MJ (1989) Saurian malaria in Kenya: redescription of some *Plasmodium* species of lizards in Africa and a key for the identification of all reported species. Int J Trop Insect Sci 10(04):531–544. doi:10.1017/S1742758400021585

Du Toit P (1937) A new piroplasm (*Sauroplasma thomasi* ng, n. sp.) of a lizard (*Zonurus giganteus*, Smith). Onderstepoort J Vet Sci Anim Ind 9:289–299

Ewers WH (1968) Blood parasites of some New Guinea reptiles and amphibia. J Parasitol 54(1):172–174

Fèvre EM, Bronsvoort BMDC, Hamilton KA, Cleaveland S (2006) Animal movements and the spread of infectious diseases. Trends Microbiol 14(3):125–131

Genchi C, Kramer LH, Rivasi F (2011) Dirofilarial infections in Europe. Vector Borne Zoonotic Dis 11(10):1307–1317. doi:10.1089/vbz.2010.0247

Godfrey RD, Fedynich AM, Pence DB (1987) Quantification of hematozoa in blood smears. J Wildl Dis 23(4):558–565

Granström M (1997) Tick-borne zoonoses in Europe. Clin Microbiol Infect 3(2):156–169. doi:10.1111/j.1469-0691.1997.tb00592.x

Harrus S, Baneth G (2005) Drivers for the emergence and re-emergence of vector-borne protozoal and bacterial diseases. Int J Parasitol 35(11–12):1309–1318

Hull RW, Camin JH (1960) Haemogregarines in snakes: the incidence and identity of the erythrocytic stages. J Parasitol 46(4):515–523

Irizarry-Rovira AR, Wolf A, Bolek M, Christian JA, DeNicola DB (2002) Blood smear from a wild-caught panther chameleon (*Furcifer pardalis*). Vet Clin Pathol 31(3):129–132

Jakes KA, O'Donoghue PJ, Adlard RD (2001) Phylogenetic relationships of *Trypanosoma chelodina* and *Trypanosoma binneyi* from Australian tortoises and platypuses inferred from small subunit rRNA analyses. Parasitol 123(Pt 5):483–487

Karesh WB, Cook RA, Bennett EL, Newcomb J (2005) Wildlife trade and global disease emergence. Emerg Infect Dis 11(7):1000–1002. doi:10.3201/eid1107.050194

Keller L, Hartmann K, Wess G (2007) Fallbericht und Literaturübersicht kutaner Dirofilariose. Tierärztl Prax Kleintiere 35(1):31–34

Kenny MJ, Shaw SE, Hillyard PD, Forbes AB (2004) Ectoparasite and haemoparasite risks associated with imported exotic reptiles. Vet Rec 154(14):434–435

Klein N (2006) Verhalten, Haltung und Nachzucht von *Pelusios castaneus*. Radiata 15(3):12–18

Klein TA, Young DG, Telford SR Jr, Kimsey R (1987) Experimental transmission of *Plasmodium mexicanum* by bites of infected *Lutzomyia vexator* (Diptera: Psychodidae). J Am Mosq Control Assoc 3(2):154–164

Klein TA, Young DG, Greiner EC, Telford SR Jr, Butler JF (1988) Development and experimental transmission of *Schellackia golvani* and *Schellackia occidentalis* by ingestion of infected blood-feeding arthropods. Int J Parasitol 18(2):259–267. doi:10.1016/0020-7519(88)90066-5

Kronefeld M, Kampen H, Sassnau R, Werner D (2014) Molecular detection of *Dirofilaria immitis*, *Dirofilaria repens* and *Setaria tundra* in mosquitoes from Germany. Parasitol Vectors 7(1):30

Lainson R, Landau I, Shaw JJ (1971) On a new family of non-pigmented parasites in the blood of reptiles: Garniidae fam. nov., (Coccidiida: Haemosporidiidea). Some species of the new genus *Garnia*. Int J Parasitol 1(3–4):241–250. doi:10.1016/0020-7519(71)90027-0

Maier WA, Grunewald J, Habedank B, Hartelt K, Kampen H, Kimmig P, Naucke T, Oehme R, Vollmer A, Schöler A (2001) Possible effects of climatic changes on the distribution of arthropode(vector)-borne infectious diseases and human parasites in Germany. Umweltbundesamt Forschungsbericht 200 61 218/11.

Majlathova V, Majlath I, Haklova B, Hromada M, Ekner A, Antczak M, Tryjanowski P (2010) Blood parasites in two co-existing species of lizards (*Zootoca vivipara* and *Lacerta agilis*). Parasitol Res 107(5):1121–1127. doi:10.1007/s00436-010-1981-0

Manzanell R (1982) *Oswaldofilaria* spp. (Filarioidea, Nematoda) in Australian agamid lizards with a description of a new species and a redescription of *O. chlamydosauri* (Breinl). Ann Parasitol Hum Comp 57(2):127–143

Melaun C, Krüger A, Werblow A, Klimpel S (2014) New record of the suspected leishmaniasis vector *Phlebotomus* (*Transphlebotomus*) *mascittii* Grassi, 1908 (Diptera: Psychodidae: Phlebotominae)—the northernmost phlebotomine sandfly occurrence in the Palearctic region. Parasitol Res 113(6):2295–2301. doi:10.1007/s00436-014-3884-y

Mihalca AD, Racka K, Gherman C, Ionescu DT (2008) Prevalence and intensity of blood apicomplexan infections in reptiles from Romania. Parasitol Res 102(5):1081–1083. doi:10.1007/s00436-008-0912-9

Mutschmann F (2002) Parasiten des Blutes bei Reptilien. Prakt Tierarzt 82(2)

Njagu Z, Mihok S, Kokwaro E, Verloo D (1999) Isolation of *Trypanosoma brucei* from the monitor lizard (*Varanus niloticus*) in an endemic focus of Rhodesian sleeping sickness in Kenya. Acta Trop 72(2):137–148

Obiamiwe BA, Iredu VI (1982) Periodicity of the microfilariae of *Foleyella agamae* (Rodhain, 1906) infecting the rainbow lizard, *Agama agama*. Folia Parasitol 29(4):315–320

Obiamiwe BA, Ohagi IE, Ugbomoiko US (1995) Morphology and cytology study of *Foleyella agamae* complex (Nematoda: Filarinae) infecting the agamid lizard *Agama agama* in Nigeria. Parasitol Res 81(6):527–530

Oosthuizen JH (1979) Redescription of *Placobdella multistriata* (Johansson, 1909) (Hirudinea: Glossiphoniidae). Koedoe 22:61–79

Paperna I (1989) Developmental cycle of chelonian haemogregarines in leeches with extra-intestinal multiple sporozoite oocysts and a note on the blood stages in the chelonian hosts. Dis Aquat Org 7(2):149–153

Pereira FB, Souza Lima S, Bain O (2010) *Oswaldofilaria chabaudi* n. sp. (Nematoda: Onchocercidae) from a South American tropidurid lizard (Squamata: Iguania) with an update on Oswaldofilariinae. Parasite 17(4):307–318

Pessôa SB (1968) *Trypanosoma hogei* n. sp., parasite of the false cobra (*Rhachidelus brazili* Boul.). Hospital (Rio J) 73(4):1257–1262

Pessôa SB, Fleury GC (1969) Duas novas species de tripanosomas parasitas de serpentes do Brasil. Rev Brasil Biol 29:81–86

Pienaar UD (1962) Haematology of some South African reptiles. Witwatersrand Univ. Pr, Johannesburg

Pietzsch M, Quest R, Hillyard PD, Medlock JM, Leach S (2006) Importation of exotic ticks into the United Kingdom via the international trade in reptiles. Exp Appl Acarol 38(1):59–65. doi:10.1007/s10493-005-5318-0

Pringle G (1960) Two new malaria parasites from East African vertebrates. Trans R Soc Trop Med Hyg 54(5):411–414. doi:10.1016/0035-9203(60)90085-7

Rasheed S (1965) Some parasitic nematodes from the Cameroons (W. Africa). J Helminthol 39(1):67–100

Reichenow E (1919) Der Entwicklungsgang der Haemococcidien *Karyolysus* u. *Schellackia* nov. gen. Sitz Ber Ges Naturf Freunde, Berlin, pp 440–447

Robin LA (1936) Cycle évolutif d'un Hepatozoon de *Gecko verticillatus*. Ann Inst Pasteur 56:376–394

Roca V, Galdon MA (2010) Haemogregarine blood parasites in the lizards *Podarcis bocagei* (Seoane) and *P. carbonelli* (Perez-Mellado) (Sauria: Lacertidae) from NW Portugal. Syst Parasitol 75(1):75–79. doi:10.1007/s11230-009-9206-6

Rogier E (1977) Description et cycle biologique de *Schellackia agamae* (Laveran et Petit, 1909) Lankesterellidae parasite d'agames de République Centre Africaine. Proc Natl Acad Sci U S A 13:9–13

Sassnau R, Genchi C (2013) Qualitative risk assessment for the endemisation of *Dirofilaria repens* in the state of Brandenburg (Germany) based on temperature-dependent vector competence. Parasitol Res 112:2647–2652

Sassnau R, Dyachenko V, Pantchev N, Stockel F, Dittmar K, Daugschies A (2009) *Dirofilaria repens*-Befall in einem Schlittenhunde rudel im Land Brandenburg. Diagnose und Therapie der kaninen kutanen Dirofilariose. Tierärztl Prax 37(2):95

Sassnau R, Czajka C, Kronefeld M, Werner D, Genchi C, Tannich E, Kampen H (2014) *Dirofilaria repens* and *Dirofilaria immitis* DNA findings in mosquitoes in Germany: temperature data allow autochthonous extrinsic development. Parasitol Res 113(8):3057–3061. doi:10.1007/s00436-014-3970-1

Sato H, Takano A, Kawabata H, Une Y, Watanabe H, Mukhtar MM (2009) *Trypanosoma* cf. *varani* in an imported ball python (*Python reginus*) from Ghana. J Parasitol 95(4):1029–1033. doi:10.1645/ge-1816.1

Schall JJ (1990) The ecology of lizard malaria. Parasitol Today 6(8):264–269. doi:10.1016/0169-4758(90)90188-a

Schall JJ, Marghoob AB (1995) Prevalence of a malarial parasite over time and space: *Plasmodium mexicanum* in its vertebrate host, the western fence lizard *Sceloporus occidentalis*. J Anim Ecol 64(2):177–185

Siddall ME (1995) Phylogeny of adeleid blood parasites with a partial systematic revision of the Haemogregarine complex. J Eukaryot Microbiol 42(2):116–125. doi:10.1111/j.1550-7408.1995.tb01551.x

Siddall ME, Desser SS (2001) Transmission of *Haemogregarina balli* from painted turtles to snapping turtles through the leech *Placobdella ornata*. J Parasitol 87(5):1217–1218

Sloboda M (2008) Harmless parasites with lifelong garantee—hemogregarine infections in boid and pythonid snakes. PhD Thesis, University of Veterinary and Pharmaceutical Sciences Brno, Brno

Sloboda M, Kamler M, Bulantova J, Votypka J, Modry D (2007) A new species of *Hepatozoon* (Apicomplexa: Adeleorina) from *Python regius* (Serpentes: Pythonidae) and its experimental transmission by a mosquito vector. J Parasitol 93(5):1189–1198

Smallridge CJ, Bull CM (2000) Prevalence and intensity of the blood parasite *Hemolivia mariae* in a field population of the skink *Tiliqua rugosa*. Parasitol Res 86(8):655–660

Smith TG (1996) The genus *Hepatozoon* (Apicomplexa: Adeleina). J Parasitol 82(4):565–585

Sulahian A, Schacher JF (1968) Brygoofilaria agamae gen. et sp. n. (Nematoda: Filarioidea) from the lizard Agama stellio in Lebanon. J Parasitol 54:831–833

Svahn K (1974) Incidence of blood parasites of the genus *Karyolysus* (Coccidia) in Scandinavian lizards. Oikos 25(1):43–53

Svahn K (1976) A new piroplasm *Sauroplasma boreale* sp. n. (Haemosporidia, Theliriidae) from the sand lizard *Lacerta agilis*. Norw J Zool 24:1–16

Telford SR Jr (1988) Studies on African saurian malarias: five *Plasmodium* species from chamaeleons of the Uzungwe Mountains, Tanzania. Int J Parasitol 18(2):197–219

Telford SR Jr (1993) A species of *Schellackia* (Apicomplexa: Lankesterellidae) parasitising east and southeast Asian lizards. Syst Parasitol 25(2):109–117. doi:10.1007/bf00009980

Telford S Jr (1995a) The kinetoplastid hemoflagellates of reptiles. In: Kreier J (ed) Parasitic Protozoa, vol 10, 2nd edn. Academic Press, San Diego

Telford SR Jr (1995b) *Plasmodium* spp. (Apicomplexa: Plasmodiidae) of the flying lizard Draco volans (Agamidae). Syst Parasitol 31(1):53–60

Telford SR Jr (1995c) A review of trypanosomes of gekkonid lizards, including the description of five new species. Syst Parasitol 31(1):37–52. doi:10.1007/bf00009301

Telford SR Jr (2009) Hemoparasites of the Reptilia: color atlas and text. CRC Press, Taylor & Francis Group, Boca Raton, FL

Tenter A, Schnieder T (2006) Erreger von Parasitosen: Taxonomie, Systematik und allgemeine Merkmale. In: Schnieder T (ed) Veterinärmedizinische Parasitologie, 6th ed. pp 26–72

Uilenberg G (1995) International collaborative research: significance of tick-borne hemoparasitic diseases to world animal health. Vet Parasitol 57(1–3):19–41

UN Comtrade, United Nations Commodity Trade Statistics Database, Department of Economic and Social Affairs/Statistics Division (2013) http://comtrade.un.org/db/. Accessed 11 December 2013

van Riper IIIC, Riper SGV, Goff ML, Laird M (1986) The epizootiology and ecological significance of malaria in Hawaiian land birds. Ecol Monogr 56(4):327–344. doi:10.2307/1942550

Viola LB, Campaner M, Takata CSA et al (2008) Phylogeny of snake trypanosomes inferred by SSU rDNA sequences, their possible transmission by phlebotomines, and taxonomic appraisal by molecular, cross-infection and morphological analysis. Parasitol 138(5):595–605

Woo P, Soltys MA (1969) The experimental infection of reptiles with *Trypanosoma brucei*. Ann Trop Med Parasitol 63(1):35–38

Wu Y, Preston G, Bianco AE (2008) Chitinase is stored and secreted from the inner body of microfilariae and has a role in exsheathment in the parasitic nematode *Brugia malayi*. Mol Biochem Parasitol 161(1):55–62. doi:10.1016/j.molbiopara.2008.06.007