Human contact influences the foraging behaviour and parasite community in long-tailed macaques

ALEXANDRA WENZ-MÜCKE[1]*, PAIBOON SITHITHAWORN[2,3], TREVOR N. PETNEY[1] *and* HORST TARASCHEWSKI[1]

[1] *KIT, Zoological Institute, Department of Ecology and Parasitology, Kornblumenstrasse 13, 76131 Karlsruhe, Germany*
[2] *University of Khon Kaen, Faculty of Medicine, Department of Parasitology, 40002 Khon Kaen, Thailand*
[3] *Liver Fluke and Cholangiocarcinoma Research Center, Faculty of Medicine, Khon Kaen University, Khon Kaen 40002, Thailand*

SUMMARY

Human–wildlife interactions have reached unprecedented levels, and humans are influencing the earth's ecosystems more rapidly and extensively than ever before. This situation is cause for serious concern, especially since disease interactions between wildlife and humans have been recognized as major conservation threats. In this study, long-tailed macaques, *Macaca fascicularis,* from 2 forest parks located in north-eastern Thailand were investigated to determine the influence of habitat modification by humans on helminth parasite associations in non-human primates. Macaque populations with contact to anthropogenically modified environments were compared with sylvatic groups in nearby natural environments. In order to test for human–non-human primate transmission of parasites, the local human populations were also examined. Humans were infected with a number of potentially pathogenic parasites, including *Opisthorchis viverrini* and *Strongyloides stercoralis*. However, eggs of these helminths were not detected in macaque feces. Thus, no direct parasite transfer from humans to non-human primates could be confirmed. However, macaque groups with more frequent contact with human-modified habitats, and a higher portion of human-provided food in their diet, had significantly higher prevalences and intensities of *Strongyloides fuelleborni* and of an intestinal fluke (probably *Haplorchis* sp.) than sylvatic groups. Positive correlations were found between the time foraging on the ground and infection with *S. fuelleborni*, and the amount of human-provided food and intestinal fluke infection. Human alteration of habitat and associated modifications in non-human primate behaviour are likely to play a role in determining the occurrence, prevalence and intensity of zoonotic helminth infection of wild non-human primates.

Key words: primates, macaques, parasites, helminth infection, humans, behaviour, habitat alteration.

INTRODUCTION

Land-use change is recognized as one of the most important factors determining modifications to ecosystems, including epidemiological cycles, world-wide (Foley *et al*. 2002; Patz *et al*. 2004). For example, road construction has been linked to increased human contact with wildlife and potentially with novel zoonoses (Wolfe *et al*. 2000), and the formation of culverts has resulted in ponds that are suitable breeding sites for malaria-transmitting anopheline mosquitoes (Marques, 1987; Charlwood and Alecrim, 1989). Parasites are ubiquitous in the lives of animals, and humans and also play a central role in ecosystems, affecting the ecology and evolution of species interactions (Esch and Fernandez, 1993), host population growth and regulation (Hudson *et al*. 1998; Hochachka and Dhondt, 2000), and even biodiversity (Hudson *et al*. 2002).

There is, in general, an established ecological and evolutionary balance such that macroparasites are able to survive and reproduce effectively without killing their hosts, thus maintaining their ability to continue reproducing in the host (Dobson and May, 1986; Lyles and Dobson, 1993). Such stable systems can, however, be disrupted when new pathogens are introduced, potentially altering the natural dynamics and thus becoming problematic for species survival (Laurenson *et al*. 1998; Taraschewski, 2006).

In Asia, more than 70% of primates are classified on the IUCN Red List as Vulnerable, Endangered, or Critically Endangered, meaning that they could disappear forever in the near future (IUCN, 2011). Since the 1970s, the academic community has recognized that many primate populations are severely threatened by human activities (Chapman and Peres, 2001). Beside the commonly cited problems for primate conservation – habitat loss to settlement, logging and agriculture, illegal hunting for bush-meat, traditional medicine, and the live primate trade – disease has been recognized as a serious threat to endangered species (May, 1988; Lyles and Dobson, 1993; Daszak *et al*. 2000; Chapman and

* Corresponding author: KIT, Zoological Institute, Department of Ecology and Parasitology, Kornblumenstrasse 13, 76131 Karlsruhe, Germany. Tel: +49 721 60842701. Fax: +49 721 6087655. E-mail: Alex.wenz@web.de.

Fig. 1. Broad-view map showing the very general location of the study sites Kosum Phi Sai and Pha Na within Thailand.

Peres, 2001; Lafferty, 2003; Smith *et al.* 2006). The dynamics of human–non-human primate inter- actions are changing radically. As a result of human population increase, combined with illegal hunting, and the fragmentation and degradation of formerly natural habitats, non-human primates are often forced to live in an anthropogenically disturbed landscapes comprising, for example, farmland, human settlements, forest fragments or isolated protected areas (Chapman and Peres, 2001; Wolfe *et al.* 2004). Gillespie and Chapman (2006) found that the index of forest patch degradation and the presence of humans strongly influenced the preva- lence of gastrointestinal nematodes in red colobus monkeys. These, and the genetic similarity between humans and non-human primates may facilitate disease transmission between these species (Wolfe *et al.* 1998; Chapman *et al.* 2005). Although there is a growing recognition that the transfer of diseases between humans and non-human primates can be of great significance for conservation biology (Wolfe *et al.* 1998; Woodford *et al.* 2002), only a few studies have focused on helminth parasites (e.g. Legesse and Erko, 2004; Chapman *et al.* 2006; Gillespie *et al.* 2005, 2010; Wenz *et al.* 2010; Standley *et al.* 2011, 2012).

In a previous investigation, we determined the influence of human contact on the presence of gastrointestinal helminths in 2 species of arboreal New World monkey from a tropical rainforest in Peru (Wenz *et al.* 2010). Although no direct transmission of helminths from humans to these primates was detected, tamarins foraging in an area of human- altered habitat showed substantial changes in their parasite communities with potential disease conse- quences.

In the present study, we examined the influence of human contact and habitat modification on the behaviour and on the presence of gastrointestinal helminths in a species of Old World monkey, the long-tailed macaque (*Macaca fascicularis*) in north- east Thailand, where there is a high rate of interaction with human-modified habitats (Fuentes, 2006).

MATERIALS AND METHODS

Study site

The study was conducted in 2 forest parks in north eastern Thailand, Kosumpee Forest Park (16°15′N and 103°04′E) in Kosum Phi Sai, and Don Chao Pu Forest Park (15°67′N and 104°86′E) in Pha Na (Fig. 1).

Kosumpee Forest Park comprises an area of about 0·2 km^2 of mixed deciduous forest and is located next to the Chi River on the northern edge of Kosum Phi Sai. According to the park staff, the Kosumpee macaque population has been isolated from other conspecific populations by extensive agricultural areas and human settlements since at least 1966, when the park was established. Based on information from the park authorities, the park is visited by about 100 000 tourists per year. Designated areas exist in the park for picnicking, sanitation and rubbish collec- tion. Directly in front of the park are a school, a Buddhist temple and villagers' houses. Park staff estimate Kosumpee supports 400–500 long-tailed macaques (*Macaca fascicularis*). However, this num- ber may be greatly underestimated.

Don Chao Pu Forest Park in Pha Na, Amnat Charoen Province comprises an area of about 0·4 km^2 and is also mixed deciduous forest. It is located about 100 m outside the village along a road. This more than 100 years old park is a designated animal sanctuary, but it has seen encroachment. It is visited mostly by people from surrounding areas and is also used for several municipal and religious occasions. For example, at the end of December the park is used by monks for meditation and Buddhist teaching, while local farmers also graze cattle inside the park. However, there are also some minimally impacted areas of forests inside and next to the park. Macaques living in theses areas have access to clear water and natural food sources throughout the year. The park is home to 300–400 long-tailed macaques (*Macaca fascicularis*) with some satellite colonies around the park.

Both study areas represent semi-natural settings with a considerable amount of human–wildlife interaction.

Study animals

The subjects of the study were members of 7 wild groups of long-tailed macaques (*Macaca fascicularis*),

3 groups in the Kosumpee Forest Park and 4 groups in the Don Chao Pu Forest Park. All sampled group members were recognized individually by unique markings such as small injuries in the face, scars, size or pelage colour. The groups investigated differed in their contact with humans and their proximity to human facilities. Additionally, their diet differed with respect to the proportion of human-provided food consumed (see below for details).

In Kosumpee Forest Park, all of the investigated groups had close contact with humans, and consumed daily large amounts of human-provided food. In addition, they had access to the school grounds and the houses in front of the park, where the monkeys would, on occasion, steal food and fight with dogs. In this study, the 3 human contact groups from Kosumpee Forest Park will be referred to as K1, K2 and K3.

In the Don Chao Pu Forest Park in Pha Na four groups were investigated. Two of these groups had contact with humans and consumed human-provided food almost every day; these were designated groups P1 and P2. The two other groups sampled were sylvatic, referred to as P3 and P4. These groups had nearly no contact at all with humans and fed almost exclusively on natural food sources.

Behavioural observations

Behavioural data were collected while following the monkey groups for whole days using 'instantaneous scan sampling' (see Martin and Bateson, 1993). With this method, a group was scanned every 5 min and the maintenance activity of all visible individuals, their height in the forest and their approximate location in the area were recorded during a 2-min period. This is an appropriate scanning period for rather dispersed groups of primates (Martin and Bateson, 1993). The 5-min interval between scans allowed for independent data points for group positions. Definitions of maintenance activities are self-evident: locomotion, feeding, drinking, grooming and resting. We differentiated between human-provided food (FH) and natural food sources (FN). The height classes were grouped into 6 categories: ground, 0–5 m, 6–10 m, 11–15 m, 16–20 m and >20 m above the ground. The behaviour of dependent infants was not recorded.

Collection and examination of fecal samples

Fresh fecal droppings from 135 individual animals comprising 7 groups were collected between July and December 2008 by following the groups during their complete activity period (06:00 h–18:00 h). The study period was restricted to the dry season only. With the beginning of the wet season, both study sites flooded; therefore fecal sample collection would not have been possible during that time. Individual fecal samples were gathered directly after defecation. Locality, date, group, sex, individual and time of defecation were recorded. After collection, the fecal samples were mixed well and about 2 g were immediately preserved in separate 15·0 mL vials containing 10% buffered formalin (solution of 10% formaldehyde and sodium phosphate buffer, pH 7·0). Samples were stored at ambient temperature. Three samples were collected from each of the 135 individuals on non-consecutive days. The finding of a recognizable parasite stage in any one of multiple samples for a given individual resulted in a positive report for that individual.

Similarly, 3 fecal samples were collected from people between the ages of 1 year and 87 years in Kosum Phi Sai (105 individuals) and Pha Na (65 individuals) on different days. These participants lived or worked next to the forest parks.

Preserved fecal samples were processed using a modified formalin-ethyl acetate sedimentation technique (see Ash *et al*. 1994). Samples were filled up with formalin to 1 mL. Samples were examined microscopically for the presence of eggs and larvae of different intestinal helminths. Parasite stages were identified by shape, size or other visible structures. Measurements were made to the nearest $0·1\,\mu$m $\pm$ S.D. Eggs or larvae of each parasite were counted and the number contained in each gram of stool was calculated from the volume of the sample examined ($300\,\mu$L), the total volume of the sample (1 mL) and the weight of the stool specimen. It was measured as an estimate of intensity of infection, and averaged for the 3 samples.

Hookworms were distinguished from *Strongyloides* species using coprocultures based on the agar plate method (Koga *et al*. 1991). Therefore, fresh fecal samples were stored at 4 °C and processed in the laboratory within 1 day. Agar plate cultures were incubated for 5 days at room temperature. The agar plates were examined under a stereomicroscope for the presence of tracks from moving larvae or free-living adults on the 3rd and 5th days. All microscopically positive dishes were further processed by washing the surface of the agar with a 10% formalin solution to collect worms for species identification. Parasites were identified with the help of keys (MAFF, 1979; Ash and Orihel, 1987). Our ability to identify most parasite species from host fecal examination, even with cultured larvae, is limited. Consequently, we present the majority of our findings at the level of family or genus.

Statistical analysis

All statistical tests were carried out using IBM®SPSS® 20 for Windows. In all tests, significance levels were set to $\alpha = 0·05$. To test statistical differences in prevalence of parasites between species, sexes and groups Fisher's exact tests were performed.

Table 1. *Prevalence of parasite infection with 95% confidence intervals in parentheses and mean intensities (eggs g^{-1}, larvae g^{-1}) of parasite infection with standard deviation ($\pm$) for humans sampled in Kosum Phi Sai and Pha Na*

(*N represents the number of sampled individuals.*)

| | Kosum Phi Sai | | Pha Na | |
| | N = 105 | | N = 65 | |
Parasite	Prevalence	Mean intensity	Prevalence	Mean intensity
O. viverrini	26·7% (18·7%–36·3%)	256 ± 392	30·7% (20·2%–43·6%)	79 ± 68
S. stercoralis	31·4% (22·9%–41·3%)	11 ± 7	36·9% (25·6%–49·8%)	39 ± 41
Hookworm	25·7% (17·9%–35·3%)	87 ± 87	36·9% (25·6%–49·8%)	85 ± 77
Taenia sp.	7·6% (3·6%–17·9%)	429 ± 285	6·3% (2·0%–15·8%)	258 ± 127
Trichostrongylus sp.	8·6% (4·2%–16·0%)	28 ± 17	0% (0%–6·9%)	–
Minute intestinal flukes (MIF)	11·4% (6·3%–19·4%)	91 ± 45	10·8% (4·8%–21·5%)	89 ± 48

We then employed binary logistic regression to model the prevalence of each parasite using variables having a *P*-value <0·20 in the univariate analyses. Significant ($P < 0·05$) risk factors in multivariate analysis were expressed as odds ratios (OR) with 95% confidence intervals (CI). Mann–Whitney U-tests were used to investigate for variation in parasite egg output between sexes, age classes and groups of macaques. Correlations of behavioural data with parasite prevalences and intensities were sought using Spearman rank correlation tests.

Ethical considerations

The research protocol was approved by the Ethical Committee of the Medical Department of Khon Kaen University. Human sample collection was conducted in collaboration with the local healthcare centres. Participation was voluntary; all participants were informed about objectives and procedures of this study and sufficient time was given to ask questions. Participants provided informed consent to the study (guardians consented in the cases of minors), associated procedures and use of results and all participants could withdraw from the study at any time without any further obligates. Free, ethically prescribed anthelminthic treatment according to World Health Organization recommendations (World Health Organization, 2006) and health education were provided by the local healthcare centres for those who harboured intestinal parasitic infections.

RESULTS

Human fecal samples

Fecal samples from the inhabitants of the 2 villages studied, Kosum Phi Sai (105 individuals) and Pha Na (65 individuals), contained 7 different types of helminth: *Opisthorchis viverrini, Taenia* sp., *Strongyloides stercoralis, Trichostrongylus* sp.,

hookworm eggs (*Necator americanus/Ancylostoma duodenale*) and small trematode eggs. These belonged to the family Heterophyidae and were possibly *Haplorchis* sp. (referred to here as minute intestinal flukes (MIF)). Prevalences in both villages were similar. For *Opisthorchis, Strongyloides* and hookworms infection rates were moderately high, whereas *Taenia, Trichostrongylus* and MIF infections were less common (Table 1). Mean intensities of infection were generally low and are presented in Table 1.

Macaque fecal samples

One trematode and at least 5 species of nematodes were recovered from macaques. The MIF belonged to the family Heterophyidae and was morphologically identical to that found in human samples. One of the nematodes was identified as *Strongyloides fuelleborni*, another as *Oesophagostomum* sp., while a third nematode was identified as a *Trichuris* sp. The eggs of a nematode taxon from the Order Rhabditida, probably *Globocephalus* sp. were also detected. Furthermore, nematode larvae belonging to the Superfamily Metastrongyloidea were found.

There were no significant differences between the prevalence of any of the parasites relating to host sex (Fisher's exact test, $P > 0·05$) or host age class (infant, juvenile, adult; Fisher's exact test, $P > 0·05$). Further, binary logistic regression revealed no significant association between prevalence of any parasite with either of the variables. Thus the data were pooled for further analyses.

Differences in prevalences among macaque groups

Parasite prevalences between the investigated macaque groups differed markedly from each other (Table 2). *Strongyloides fuelleborni* was absent in the sylvatic group P4 from Pha Na, revealing significant differences with all human-contact groups. In the second sylvatic group, P3, only 1 of the macaques was

Table 2. Prevalence of parasite infection for the different macaque groups

(*N* indicates number of sampled individuals. 95% Confidence intervals including continuity correction are given in parentheses.)

Group	Ind.	*S. fuelleborni* (%)	*Trichuris* sp. (%)	*Globocephalus* sp. (%)	MIF (%)	*Oesophagostomum* sp. (%)	Nematode larvae (%)
K1	20	65 (41–83·7)	70 (45·6–87·1)	5 (0·3–26·9)	20 (6·6–44·2)	0 (0–20)	25 (9·6–49·4)
K2	20	70 (45·6–87·1)	70 (45·6–87·1)	5 (0·3–26·9)	30 (12·8–54·3)	0 (0–20)	30 (12·8–54·3)
K3	20	65 (41–83·7)	75 (50·6–90·4)	25 (9·6–49·4)	20 (6·6–44·2)	0 (0–20)	40 (20–63·6)
P1	21	38·1 (18·9–61·3)	38·1 (18·9–61·3)	14·3 (3·7–37·3)	14·3 (3·7–37·3)	19 (6·3–42·5)	9·5 (3·7–37·3)
P2	20	15 (12·8–54·3)	40 (20–63·6)	5 (0·3–26·9)	20 (6·6–44·2)	10 (1·8–33·1)	10 (4–38·8)
P3	16	6·3 (0·3–29·3)	31·1 (10·7–53·6)	18·8 (4·4–42·2)	0 (0–21·89)	11·1 (1·9–36)	6·3 (4·4–42·2)
P4	18	0 (0–24·1)	33·3 (16·2–64·1)	5·6 (0·3–32·3)	0 (0–24·1)	16·7 (4·9–46·3)	22·2 (8·3–52·6)

infected. All groups from Kosumpee Forest Park had significantly higher prevalences of *S. fuelleborni* infection than the sylvatic groups P3 and P4 from Pha Na (Fisher's exact test, $P < 0.001$). In addition, the prevalence of infection was significantly higher in group K2 than in the human contact P2 group in Pha Na (Fisher's exact test, $P = 0.026$). There were also significant differences in *Strongyloides* infection between the groups from Pha Na. The human contact group P1 showed significantly higher infection rates than either of the non-contact groups P3 and P4 (Fisher's exact test, P1 *vs* P3 $P = 0.05$, P1 *vs* P4 $P = 0.004$) and the second human contact group P2 was significantly more often infected than sylvatic P4 (Fisher's exact test, $P = 0.021$). Multivariate analyses showed 2 significant variables. Macaques originating from Kosumpee Forest Park are at greater risk of being infected with *S. stercoralis* (OR 4·02; CI 1·71–9·42) than macaques from Pha Na. Additionally, human contact raises the risk of being infected with this parasite (OR 17·44; CI 2·15–141·53).

The MIF was totally absent from the sylvatic groups P3 and P4, whereas the parasite was present in all the human contact groups with prevalences ranging from 14·3% ($n = 21$) to 30% ($n = 20$).

Trichuris sp. showed significantly lower prevalences in the sylvatic groups P3 and P4 than in the human contact groups from Kosumpee Forest Park (Fisher's exact tests, $P < 0.05$). In addition, the human contact group P1 from Pha Na had a significantly lower prevalence than K3 (Fisher's exact test, $P = 0.028$). Binary logistic regression revealed that macaques originating from Kosumpee Forest Park are at greater risk of being infected with *Trichuris* sp. (OR 3·94; CI 1·67–9·30) than macaques from Pha Na. Contact with humans, however, showed no significant association.

Oesophagostomum sp. was detected only in the groups from Pha Na. For *Globocephalus* sp. and the nematode larvae, no significant differences in the prevalences of these parasites were detected between any of the macaque groups.

Differences in egg output among macaque groups

Eggs and larvae excreted per gram of macaque stool samples were also examined. In general, the number of parasite stages voided in feces was low for all macaques sampled during the study period (Table 3). The highest mean intensities of infection were observed for *S. fuelleborni* and *Trichuris* sp., with maximum counts of 15 760 eggs per gram (epg) for *S. fuelleborni* and 770 epg *Trichuris* sp.

Inter-group differences in mean intensity were also recorded for *S. fuelleborni* and *Trichuris* sp. For *S. fuelleborni* the mean intensity from Kosum group K3 was significantly higher than from groups P1 and P2 (MWU K3 + P1 = 0·05, K3 + P2 = 0·018). Statistics

Table 3. Mean intensities (eggs g^{-1}, larvae g^{-1}) of parasite infection with standard deviation (±) for the different macaque groups

(*N* indicates the number of individuals sampled, the number of infected individuals appears in parentheses.)

Group	N	S. fuelleborni	Trichuris sp.	Globocephalus sp.	MIF	Oesophagostomum sp.	Nematode larvae
K1	20	673±1728 (13)	44±53 (14)	43·5(1)	2·5±1·5 (4)	0	7·5±12 (5)
K2	20	205·5±242·5 (14)	31±47 (14)	1·5 (1)	3·5±3·5 (6)	0	28±63 (6)
K3	20	561·5±1504·5 (13)	70±83·5 (15)	15±18 (5)	3±1 (4)	0	19±24·5 (8)
P1	21	30·5±48 (8)	9·5±8·5 (8)	5±4·5 (3)	5·0±5·5 (3)	2·5±2 (4)	7·5±10 (3)
P2	20	13±11 (6)	33±39 (8)	1·5 (1)	2·5±1·5 (4)	1·5±0·5 (2)	11±9 (3)
P3	16	2·5 (1)	15·5±15·5 (5)	3±1 (3)	0	8±0·5 (2)	20·5±30 (3)
P4	18	0	10±112 (6)	2·5 (1)	0	1±1 (3)	8±10·5 (4)

for *S. fuelleborni* infection intensity was not useful with either forest group, since only 1 individual was infected in group P3, and no infection occurred in P4.

Mean infection intensity of *Trichuris* sp. was significantly higher in all Kosum groups compared with group P1 (MWU K1 + P1 *P* = 0·008, K2 + P1 *P* = 0·043, K3 + P1 *P* = 0·001). In addition, groups K1 and K3 showed significantly higher intensities than P4 (MWU K1 + P4 *P* = 0·018, K3 + P4 *P* = 0·004) and group K3 showed higher outputs than P3 (MWU *P* = 0·013). K3 showed higher egg outputs than group K2 (MWU *P* = 0·006). All other parasite species had similar mean egg outputs (Table 3).

Correlations of parasitological with behavioural data

The behaviour of human contact groups (K1–3 and P1 + 2) differed significantly from non-contact groups (P3 + 4) by spending more time being fed by humans (MWU *P* = 0·001) and on the ground (MWU *P* = 0·001), while the non-contact groups spent significantly more time 16 m to 20 m in trees above the ground (MWU *P* = 0·001). Time spent moving, resting, feeding, drinking and grooming were significantly different between these groups.

There were positive correlations between the prevalence of *Strongyloides*, *Trichuris* and the nematode larvae and the habit of accepting human-provided food, while the correlation with *Oesophagostomum* was significantly negative. The corresponding correlations of parasite prevalences with the time spent eating natural food sources were also highly significant but in the opposite direction. Similarly, the prevalences of *Strongyloides*, *Trichuris* and the nematode larvae were positively correlated with the time spent on the ground, while for *Oesophagostomum* the correlation was negative and highly significant (Table 4).

There were strong positive correlations between mean intensity of infection of *Strongyloides*, *Trichuris*, MIF and the nematode larvae with the time spent eating human-provided food while the correlation for *Oesophagostomum* was significantly negative (Table 5). The converse was true for time spent eating natural food sources. There were also significant positive correlations between the intensity of infection and the time spent on the ground for *Strongyloides*, *Trichuris* and the MIF while the correlation was again significantly negative for *Oesophagostomum*.

DISCUSSION

In this study, the macaques that foraged near human-inhabited areas and ingested human-provided food differed markedly from the groups that had less overlap with humans and human-modified habitats. Human contact groups displayed significantly higher prevalences of *Strongyloides fuelleborni* and of minute

Table 4. *Spearman rank correlation of parasite prevalence with behavioural data of macaques*

Parasite	% time on ground			% of human-provided food in diet		
	N	r_s	P	N	r_s	P
S. fuelleborni	135	**0·364**	**0·01**	135	**0·362**	**0·01**
Trichuris sp.	135	**0·382**	**0·007**	135	**0·397**	**0·005**
Globocephalus sp.	135	0·291	0·131	135	0·195	0·18
Minute intestinal flukes (MIF)	135	0·078	0·156	135	0·263	0·068
Oesophagostomum sp.	135	− 0·589	**<0·001**	135	− 0·593	**<0·001**
Nematode larvae	135	**0·811**	**<0·001**	135	**0·821**	**<0·001**

Table 5. *Spearman rank correlation of parasite intensity with behavioural data of macaques*

Parasite	% time on ground			% of human-provided food in diet		
	N	r_s	P	N	r_s	P
S. fuelleborni	135	**0·748**	**<0·001**	135	**0·762**	**<0·001**
Trichuris sp.	135	**0·464**	**0·001**	135	**0·448**	**0·001**
Globocephalus sp.	135	0·131	0·371	135	0·164	0·26
Minute intestinal flukes (MIF)	135	**0·545**	**<0·001**	135	**0·568**	**<0·001**
Oesophagostomum sp.	135	− 0·542	**<0·001**	135	− 0·537	**<0·001**
Nematode larvae	135	0·249	0·085	135	0·244	0·091

intestinal flukes (MIF). We conclude that the presence of humans and the associated changes of primate behaviour can lead to substantial changes in the community structure of intestinal helminths in wild non-human primate populations.

Land unaffected by humans is increasingly rare (Lilly *et al*. 2002) and human activities can alter the ecology of wildlife and environmental parameters in ways that increase the probability of an infection. In a previous study, we showed significantly higher infection rates of *Prosthenorchis elegans* (Acanthocephala) in 2 tamarin species living in a human-modified habitat compared with their conspecifics in more natural settings (Wenz *et al*. 2010). Human modification of the habitat may favour the intermediate host of this potentially pathogenic parasite thereby increasing the probability of exposure. With the modification of wildlife habitat humans may also bring about a change in the behaviour and diet of non-human primates (Weyher *et al*. 2006); indeed many such changes are attributed to the availability of anthropogenic nutrition (Else and Lee, 1986; Box, 1991; Weyher *et al*. 2006). In addition, a change in foraging strategy may also alter the relationship that primates have with the parasites already present in their environment as shown in wild-foraging and crop-raiding baboons in Nigeria (Weyher *et al*. 2006).

Our behavioural observations show a positive correlation of *S. fuelleborni* infection with the time macaques spent on the ground. Further, as a consequence of foraging on human-provided food, the daily activity and ranging patterns were different from those of the completely wild foraging macaques

in the same area. Human-contact macaques spent more time on the ground than the wild foraging groups resulting possibly in greater exposure to, for example, the infective L3 larvae of *S. fuelleborni*. Similar observations have been made for baboons. As a consequence of rubbish foraging, daily activity and ranging patterns differed from those of wild foraging baboons (Altman and Muruthi, 1988). Semi-provisioned baboons rested significantly more and their home ranges were significantly smaller than non-provisioned individuals (Altman and Muruthi, 1988).

Strongyloides fuelleborni belongs to a genus of widely distributed nematodes parasitic in the intestine of humans and other mammals. While it is transmitted mainly by non-human primates (Crompton and Savioli, 2006), it is also considered a helminth of human health importance, especially in Central Africa and Papua New Guinea (Viney *et al*. 1991; Grove, 1996). Although infections of humans in Thailand have not been reported, this highlights the potential for zoonotic transmission. Infection is assumed to occur when the infective L3 larvae penetrate the skin (Crompton and Savioli, 2006). Animals often defecate, cough, vomit, bleed or urinate on or near the ground or low-lying substrates dispersing infective stages of parasites (Nunn *et al*. 2000). Thus, it is reasonable to expect that terrestrial primates experience greater infection risk than arboreal primates (Nunn *et al*. 2000).

S. fuelleborni infections can have fatal consequences. The parasite was implicated in the death of several rhesus monkeys (Remfry, 1978). The pathological effects of *S. fuelleborni* range from a

cough, bronchopneumonia and diarrhoea to peritonitis (Flynn, 1973). In a young rhesus male, for example, a small abscess on the caecum, blood-staining of the omentum and peritoneal fluid and ulceration of the mucosal surface of the small and large intestines were detected post-mortem. In that case it was suggested that the peritonitis was caused by perforation of a *Strongyloides*-induced ulcer (Remfry, 1978).

Although *S. fuelleborni* infections occur naturally in non-human primates, high prevalence and intensity of infection may have negative consequences for a population of wild animals. Prevalences of more than 70% in the macaques from Kosumpee Forest Park are therefore a potentially disturbing observation, particularly since the mean intensity of infection was also significantly higher than in sylvatic groups. On the other hand, the number of parasite eggs measured in fecal material is affected by many factors (e.g. bias due to variation in egg production, clumping of eggs and differences in stool consistency; Stuart and Strier, 1995, Engels *et al.* 1996, 1997; Ross *et al.* 1998). This was considered when choosing the sampling regime. Since a roughly linear, positive relationship between numbers of eggs g^{-1} of feces and parasite burden has been observed for a number of parasites (Keymer and Hiorns, 1986; Pritchard *et al.* 1990; Sithithaworn *et al.* 1991), we suggest that intensity of infection by the adults of *S. fuelleborni* was also higher in the groups with human contact compared to the sylvatic ones.

Although *Globocephalus* has a similar life cycle and transmission mode to *S. fuelleborni*, we did not detect any significant differences in prevalence of infection. However, only a few individuals were infected with this parasite and the time macaques spent on the ground seems not to have had an influence on parasite transmission. Mean intensity of infection was also very low, and it is possible that only a few infective stages were present in the environment.

The heterophyid trematode (MIF) was present only in human contact groups. Exact identification of the MIF was not possible based on fecal examination alone. Adult worms would have assisted, but a treatment of wild monkeys with antihelminthics to expel the parasites was not possible. Heterophyid flukes live as adults in the intestine of mammals and birds, and they infect snails and freshwater fish as intermediate hosts. Human infection occurs through consumption of raw or undercooked fish which harbour the metacercariae (Fried *et al.* 2004). *Haplorchis taichui* is the most prevalent trematode in the gastrointestinal tract of humans in northern Thailand (Pungpak *et al.* 1998; Radomyos *et al.* 1998), and in cyprinid fish (Srisawangwong *et al.* 1997; Sukontason *et al.* 1999). Sukontason *et al.* (2005) presented the pathology associated with *H. taichui* in the small intestine for 3 human subjects. Microscopic examination revealed mucosal

ulceration, mucosal and submucosal haemorrhages, fusion and shortening of the villi, chronic inflammation and fibrosis of the submucosa. Significant pathology in the heart, brain and spinal cord of humans may also occur, and is thought to be caused by the atypical carriage of fluke eggs through the circulatory system (Fried *et al.* 2004).

Haplorchis taichui is also known to occur in macaques (Yamashita, 1963), but information on pathogenicity of this parasite in non-human primates is scarce. Given the knowledge of pathological effects of intestinal flukes in humans, it is likely that there is also a health risk for the monkeys.

Infection of macaques with *H. taichui* implies that they come into contact with raw or undercooked fish. Our observations point to the conclusion that human-provided food is an important factor influencing non-human primate infection. Behavioural observations show a positive correlation between this MIF infection and the amount of human-provided food in the diet of the macaques. Since the wild foraging macaques were not infected with this parasite and their preferred foraging height strongly reduced the likelihood of a diet containing fish, it seems likely that infected intermediate hosts of the MIF formed part of the human-provided food.

Consuming human foods can have significant epidemiological costs due to an increased risk of disease transmission for both humans and primates. Some studies have documented the costs to baboons from foraging in human rubbish dumps, including infections with antibiotic-resistant bacteria, probably acquired from humans (Rolland *et al.* 1985; Routman *et al.* 1985) and infections with tuberculosis from eating contaminated meat (Sapolsky and Else, 1987; Keet *et al.* 2000). In contrast, Eley *et al.* (1989) found no increased risk of baboons in Kenya acquiring gut parasites from rubbish dump foraging.

The divergences observed in the parasite community structure of the macaques appear not to be related to a transfer of parasites directly from humans. Sampled humans harboured important parasites with pathological effects (*O. viverrini*, MIF, *N. americanus/A. duodenale*, *Trichostrongylus*, *Taenia* and *S. stercoralis*). Their presence can mainly be traced back to diet and hygienic standards. The only concurrence that was found was the minute intestinal fluke that was present in both humans and monkeys. This parasite may be indirectly transmitted to the monkeys by humans through contaminated food. To determine the reason why macaques were not infected by *O. viverrini* detailed studies will be needed of the items which the macaques are picking out of the food offered to them by humans. On the other hand, a low suitability of macaques as hosts of *Opisthorchis* has been reported (Wykoff, 1964).

In addition, the macaques in the present study harboured several parasite species which exhibit potential for zoonotic transmission. The eggs

identified as *Trichuris* sp. in this study – although resembling the human whipworm *T. trichiura* – were larger. On the other hand, morphological studies on adult parasites identified as *T. trichiura* showed differences between specimens collected from non-human primates and humans using light and scanning electron microscopy (Ooi *et al*. 1993). Additionally, in our study none of the humans had eggs of *T. trichiura* in the feces and both sylvatic and human contact macaques displayed similar prevalences of infection. Therefore, infection with *Trichuris* sp. seems to occur naturally in macaques and not as a result of anthropozoonotic transmission. The same seems to be the case for *Oesophagostomum*. Additionally, we did not detect any evidence of zoonotic transmission or double infection of either species of *Strongyloides*. The villagers were infected only with *S. stercoralis*, whereas the macaques harboured only *S. fuelleborni*.

Results from the present study in Thailand, as with the results of a previous study by us in Peru (Wenz *et al*. 2010), lead to the conclusion that the presence of humans and/or an environment modified by humans can lead to substantial changes in the community structure of intestinal helminths in wild primates. These changes may even have pathogenic consequences. In terms of conservation, it follows that geographical areas designated for the protection of non-human primate populations should be placed far enough from human settlements to prevent foraging on disturbed land and that the food provisioning by humans requires more effective control in areas like forest parks.

REFERENCES

Altman, J. and Muruthi, P. (1988). Differences in daily life between semiprovisioned and wild-foraging baboons. *American Journal of Primatology* **15**, 213–221.

Ash, L. R. and Orihel, T. C. (1987). *Parasites: a Guide to Laboratory Procedures and Identification*. American Society of Clinical Pathologists Press, Chicago, IL, USA.

Ash, L. R., Orihel, T. C. and Savioli, L. (1994). *Bench Aids for the Diagnosis of Intestinal Parasites*. World Health Organization, Geneva, Switzerland.

Box, H. O. (1991). Training for life after release: simian primates as examples. *Symposia of the Zoological Society of London* **62**, 111–123.

Chapman, C. A., Gillespie, T. R. and Goldberg, T. L. (2005). Primates and the ecology of their infectious diseases: how will anthropogenic change affect host-parasite interactions? *Evolutionary Anthropology* **14**, 134–144. doi: 10.1002/evan.20068.

Chapman, C. A. and Peres, C. A. (2001). Primate conservation in the new millennium: the role of scientists. *Evolutionary Anthropology* **10**, 16–33.

Chapman, C. A., Speirs, M. L., Gillespie, T. R., Holland, T. and Austad, K. M. (2006). Life on the edge: the gastrointestinal parasite from forest edge and interior primate groups. *American Journal of Primatology* **68**, 1–12. doi: 10.1002/ajp.20233.

Charlwood, J. D. and Alecrim, W. A. (1989). Capture-recapture studies with the South American malaria vector *Anopheles darlingi, Root. Annals of Tropical Medicine and Parasitology* **83**, 569–576.

Crompton, D. W. T. and Savioli, L. (2006). *Handbook of Helminthiasis for Public Health*. CRC Press, Boca Raton, FL, USA.

Daszak, P., Cunningham, A. A. and Hyatt, A. D. (2000). Emerging infectious diseases of wildlife – threats to biodiversity and human health. *Science* **287**, 443–449.

Dobson, A. P. and May, R. M. (1986). Disease and conservation. In *Conservation Biology: The Science of Scarcity and Diversity* (ed. Soule, M. and Sunderland, M. A.), pp. 345–365. Sinauer Associates Inc., Sunderland, MA, USA.

Eley, R. M., Strum, S. C., Muchemi, G. and Reid, G. D. F. (1989). Nutrition, body condition, activity patterns and parasitism of free-ranging baboons (*Papio anubis*) in Kenya. *American Journal of Primatology* **18**, 209–219. doi: 10.1002/ajp.1350180304.

Else, J. G. and Lee, P. C. (1986). Primate–human conflict. In *Primate Ecology and Conservation*. (ed. Else, J. G. and Lee, P. C.), pp. 213–214. Cambridge University Press, Cambridge, UK.

Engels, D., Sinzinkayo, E. and Gryseels, B. (1996). Day-to-day egg count fluctuation in *Schistosoma mansoni* infection and its operational implications. *American Journal of Tropical Medicine and Hygiene* **54**, 319–324.

Engels, D., Sinzinkayo, E., DeVlas, S. J. and Gryseels, B. (1997). Intraspecimen fecal egg count variation in *Schistosoma mansoni* infection. *American Journal of Tropical Medicine and Hygiene* **57**, 571–577.

Esch, G. W. and Fernandez, J. C. (1993). *A Functional Biology of Parasitism: Ecological and Evolutionary Implications*. Chapman & Hall, London, UK.

Flynn, R. J. (1973). *Parasites of Laboratory Animals*. Iowa State University Press, Ames, IA, USA.

Foley, J. A., DeFries, R., Asner, G. P., Barford, C., Bonan, G., Carpenter, S. R., Chapin, F. S., Coe, M. T., Daily, G. C., Gibbs, H. K., Helkowski, J. H., Holloway, T., Howard, E. A., Kucharik, C. J., Monfreda, C., Patz, J. A., Prentice, I. C., Ramankutty, N. and Snyder, P. K. (2002). Global consequences of land use. *Science* **309**, 570–574. doi: 10.1126/science.111772.

Fried, B., Graczyk, T. K. and Tamang, L. (2004). Food-borne intestinal trematodiases in humans. *Parasitology Research* **93**, 159–170.

Fuentes, A. (2006). Human culture and monkey behaviour; assessing the contexts of potential pathogen transmission between macaques and humans. *American Journal of Primatology* **68**, 880–896. doi: 10.1002/ajp.20295.

Gillespie, T. R. and Chapman, C. A. (2006). Forest fragment attributes predict parasite infection dynamics in primate metapopulations. *Conservation Biology* **20**, 441–448.

Gillespie, T. R., Greiner, E. C. and Chapman, C. A. (2005). Gastrointestinal parasites of the colobus monkeys of Uganda. *Journal of Parasitology* **91**, 569–573.

Gillespie, T. R., Lonsdorf, E. V., Canfield, E. P., Meyer, D. J., Nadler, Y., Raphael, J., Pusey, A. E., Pond, J., Pauley, J., Mlengeya, T. and Travis, D. A. (2010). Demographic and ecological effects on patterns of parasitism in eastern chimpanzees (*Pan troglodytes schweinfurthii*) in Gombe National Park, Tanzania. *American Journal of Physical Anthropology*. **143**, 534–544.

Grove, D. I. (1996). Human strongyloidiasis. *Advances in Parasitology* **38**, 251–309.

Hochachka, W. M. and Dhondt, A. A. (2000). Density-dependent decline of host abundance resulting from a new infectious disease. *Proceedings of the National Academy of Sciences, USA* **97**, 303–530. doi: 10.1073/pnas.080551197.

Hudson, P. J., Dobson, A. P. and Newborn, D. (1998). Prevention of population cycles by parasite removal. *Science* **282**, 2256–2258. doi: 10.1126/science.282.5397.2256.

Hudson, P. J., Rizzoli, A. P., Grenfell, B. T., Heesterbeek, H. and Dobson, A. P. (2002). *The Ecology of Wildlife Diseases*. Oxford University Press, Oxford, UK.

IUCN (2011). *IUCN Red List of Threatened Species*. Version 2011.2 www. iucnredlist.org

Keet, D. F., Krick, N. P. J., Bengis, R. G., Grobler, D. G. and Michel, A. (2000). The rise and fall of tuberculosis in a free-ranging chacma baboon troop in the Kruger National Park. *Onderstepoort Journal of Veterinary Research* **67**, 115–122.

Keymer, A. E. and Hiorns, R. W. (1986). Faecal egg counts and nematode fecundity: *Heligmosomoides polygyrus* and laboratory mice. *Parasitology* **93**, 189–203.

Koga, K., Kasuya, S., Khamboonruang, C., Sukhavat, K., Ieda, M., Takatsuka, N., Kita, K. and Ohtomo, H. (1991). A modified agar plate method for detection of *Strongyloides stercoralis*. *American Journal of Tropical Medicine and Hygiene* **45**, 518–521.

Lafferty, K. D. (2003). Is disease increasing or decreasing, and does it impact or maintain biodiversity? *Journal of Parasitology* **89**, 5101–5105.

Laurenson, K., Sillero-Zubiri, C., Thompson, H., Shiferaw, F., Thirgood, S. and Malcolm, J. (1998). Disease as a threat to endangered species: Ethiopian wolves, domestic dogs and canine pathogens. *Animal Conservation* **1**, 273–280.

Legesse, M. and Erko, B. (2004). Zoonotic intestinal parasites in *Papio anubis* (baboon) and *Cercopithecus aethops* (vervet) from four localities in Ethiopia. *Acta Tropica* **90**, 231–236.

Lilly, A. A., Mehlman, P. T. and Doran, D. (2002). Intestinal parasites in gorillas, chimpanzees, and humans at Mondika Research Site, Dzanga-Ndoki National Park, Central African Republic. *International Journal of Primatology* **23**, 555–573.

Lyles, A. M. and Dobson, A. P. (1993). Infectious disease and intensive management: population dynamics, threatened hosts, and their parasites. *Journal of Zoo and Wildlife Medicine* **24**, 315–326.

MAFF (1979). *Parasitological Laboratory Techniques, Technical Bulletin No. 18*. Ministry of Agriculture, Fisheries and Food Manual of Veterinary, Her Majestey's Stationery Office, London, UK.

Marques, A. C. (1987). Human migration and the spread of malaria in Brazil. *Parasitology Today* **3**, 166–170.

Martin, P. and Bateson, P. (1993). *Measuring Behaviour: An Introductory Guide*, 2nd Edn. Cambridge University Press, Cambridge, UK.

May, R. M. (1988). Conservation and disease. *Conservation Biology* **2**, 28–30.

Nunn, C., Gittleman, J. L. and Antonovics, J. (2000). Promiscuity and the primate immune system. *Science* **290**, 1168–1170. doi: 10.1126/science.290.5494.1168

Ooi, H. K., Tenora, F., Itoh, K. and Kamiya, M. (1993). Comparative study of *Trichuris trichiura* from nonhuman primates and from man, and their differences with *T. suis*. *Journal of Veterinary Medical Science* **55**, 363–366.

Patz, J. A., Daszak, P., Tabor, G. M., Aguirre, A. A., Pearl, M., Epstein, J., Wolfe, N. D., Kilpatrick, M. A., Foufopoulos, J., Molyneux, D. and Bradley, D. J. (2004). Unhealthy landscapes: policy recommendations on land use change and infectious disease emergence. *Environmental Health Perspectives* **112**, 1092–1098. doi:10.1289/ehp.6877.

Pritchard, D. I., Quinnell, R. J., Slater, A. F., McKean, P. G. and Dale, D. D. S. (1990). Epidemiology and immunology of *Necator americanus* infection in a community in Papua New Guinea: humoral responses to excretory-secretory and cuticular collagen antigens. *Parasitology* **100**, 317–326.

Pungpak, S., Radomyos, P., Radomyos, B., Schelp, F. P., Jongsuksuntigul, P. and Bunnag, D. (1998). Treatment of *Opisthorchis viverrini* and intestinal fluke infections with praziquentel. *Southeast Asian Journal of Tropical Medicine and Public Health* **29**, 246–249.

Radomyos, B., Wongsaroj, T., Wilairatana, P., Radomyos, P., Praevanich, R., Meesomboon, V. and Jongsuksuntikul, P. (1998). Opisthorchiasis and intestinal fluke infections in northern Thailand. *Southeast Asian Journal of Tropical Medicine and Public Health* **29**, 123–127.

Remfry, J. (1978). The incidence, pathogenesis and treatment of helminth infections in rhesus monkeys (*Macaca mulatta*). *Laboratory Animals* **12**, 213–218.

Rolland, R. M., Hausfater, G., Marshall, B. and Levy, S. B. (1985). Antibiotic-resistant bacteria in wild primates – increased prevalence in baboons feeding on human refuse. *Applied Environmental Microbiology* **49**, 791–794.

Ross, A. G. P., Li, Y., Sleigh, A. C., Williams, G. M. and McManus, D. P. (1998). Faecal egg aggregation in humans infected with *Schistosoma japonicum* in China. *Acta Tropica* **70**, 205–210. doi: http://dx. doi.org/10.1016/S0001-706X(98)00022-9.

Routman, E., Miller, R. D., Phillips-Conroy, J. and Hartl, D. L. (1985). Antibiotic resistance and population structure in *Escherichia coli* from free-ranging African yellow baboons. *Applied Environmental Microbiology* **50**, 749–754.

Sapolsky, R. M. and Else, J. G. (1987). Bovine tuberculosis in a wild baboon population: epidemiological aspects. *Journal of Medical Primatology* **16**, 229–235.

Sithithaworn, P., Tesana, S., Pipitgool, V., Kaewkes, S., Pairojkul, C., Sripa, B., Pauparoj, A. and Thaiklar, A. (1991). Relationship between faecal egg count and worm burden of *Opisthorchis viverrini* in human autopsy cases. *Parasitology* **103**, 277–281. doi: http://dx.doi.org/10.1017/S0031182000062594.

Smith, K. F., Sax, D. F. and Lafferty, K. D. (2006). Evidence for the role of infectious disease in species extinction and endangerment. *Conservation Biology* **20**, 1349–1357. doi: 10.1111/j.1523-1739.2006.00524.x.

Srisawangwong, T., Sithithaworn, P. and Tesana, S. (1997). Metacercariae isolated from cyprinoid fish in Khon Kaen district by digestion technique. *Southeast Asian Journal of Tropical Medicine and Public Health* **28**, 224–226.

Standley, C. J., Mugisha, L., Verweij, J. J., Adriko, M., Arinaitwe, M., Rowell, C., Atuhaire, A., Betson, M., Hobbs, E., van Tulleken, C. R., Kane, R. A., van Lieshout, L., Ajarova, L., Kabatereine, N. B. and Stothard, J. R. (2011). Confirmed infection with intestinal schistosomiasis in semi-captive wild-born chimpanzees on Ngambe Island, Uganda. *Vector-Borne and Zoonotic Diseases* **11**, 169–176.

Standley, C. J., Mugisha, L., Dobson, A. P. and Stothard, J. R. (2012). Zoonotic schistosomiasis in non-human primates: past, present and future activities at the human-wildlife interface in Africa. *Journal of Helminthology* **86**, 131–140.

Stuart, M. D. and Strier, K. B. (1995). Primates and parasites: a case for a multidisciplinary approach. *International Journal of Primatology* **15**, 577–593. doi: 10.1007/BF02735282.

Sukontason, K., Piangjai, S., Muangyimpong, Y., Sukontason, K., Methanitikorn, R. and Chaithong, U. (1999). Prevalence of trematode metacercariae in cyprinoid fish of Ban Pao district, Chiang Mai province, northern Thailand. *Southeast Asian Journal of Tropical Medicine and Public Health* **30**, 365–370.

Sukontason, K., Upunyo, P., Sukontason, K. L. and Piangjai, S. (2005). Evidence of *Haplorchis taichui* infection as a pathogenic parasite: three case reports. *Scandinavian Journal of Infectious Diseases* **37**, 388–390. doi: 10.1080/00365540510034473

Taraschewski, H. (2006). Hosts and parasites as aliens. *Journal of Helminthology* **80**, 99–128. doi: 10.1079/JOH2006364.

Viney, M., Ashford, R. W. and Barnish, G. (1991). A taxonomic study of *Strongyloides* Grassi, 1879 (Nematoda) with special reference for *Strongyloides fuelleborni* von Linstow, 1905 in man in Papua New Guinea and the description of a new subspecies. *Systematic Parasitology* **18**, 95–109.

Wenz, A., Heymann, E. W., Petney, T. and Taraschewski, H. (2010). The influence of human settlements on the parasite community in two species of Peruvian tamarin. *Parasitology* **137**, 675–684. doi: http://dx.doi.org/10.1017/S0031182009991570.

Weyher, A. H., Ross, C. and Semple, S. (2006). Gastrointestinal parasites in crop raiding and wild foraging *Papio anubis* in Nigeria. *International Journal of Primatology* **27**, 2519–1534. doi: 10.1007/s10764-006-9089-1.

World Health Organization (2006). *Preventive Chemotherapy in Human Helminthiasis: Coordinated Use of Antihelminthic Drugs in Control Interventions: a Manual for Health Professionals and Programme Managers*. World Health Organization, Geneva, Switzerland.

Wolfe, N. D., Eifel, M. N., Gockowski, J., Muchaal, P. K., Nolte, C., Tassey Proser, A., Ndongo Torimiro, J., Weise, S. F. and Burke, D. S. (2000). Deforestation, hunting and the ecology of microbial emergence. *Global Change and Human Health* **1**, 10–25.

Wolfe, N. D., Escalante, A. A., Karesh, W. B., Kilbourne, A., Spielman, A. and Lal, A. A. (1998). Wild primate populations in emerging infectious disease research: the missing link? *Emerging Infectious Diseases* **4**, 149–158.

Wolfe, N. D., Tassey Prosser, A., Carr, J. K., Tamoufe, U., Mpoudi-Ngole, E., Ndongo Torimiro, J., LeBreton, M., McCutchan, F. E., Birx, D. L. and Burke, D. S. (2004). Exposure to nonhuman primates in rural Cameroon. *Emerging Infectious Diseases* **10**, 2094–2099.

Woodford, M. H., Butynski, T. M. and Karesh, W. B. (2002). Habituating the great apes: the disease risks. *Oryx* **36**, 153–160. doi: 10.1017/S0030605302000224.

Wykoff, D. E. (1964). *Studies on* Opisthorchis viverrini *in Thailand*. *Experimental Laboratory Hosts*. Annual Progress Report. http://www.afrims.org/weblib/eapr/1963/APR63p161-164.pdf

Yamashita, J. (1963). Ecological relationship between parasites and primates. *Primates* **4**, 1–96. doi: 10.1007/BF01659699.