University of Veterinary Medicine Budapest,
Department of Parasitology and Zoology

Diversity of gastro-intestinal parasites of wild Guinea baboons in Senegal

by

Alexia Coles

Supervisors:

Dr. Alexandra Juhasz, DVM, PhD, Senior lecturer,
Department of Parasitology, UVMB, Hungary

Prof. Dr. Julia Fischer, Head of Cognitive Ethology Laboratory,
German Primate Centre, Germany

Dr. David Modry, Department of Parasitology,
Veterinary and Pharmaceutical University, Brno, Czech Republic

Budapest
2020

Contents

Abbreviations 1

Abstract 1

Part I. Introduction 3

1. Impacts of human encroachment 3

2. Research models 4

Part 2. Literature review 5

1. Guinea Baboons 5

1.1 - External systematics of the genus *Papio* 5

1.2 - Internal systematics of the genus *Papio* 5

1.3 - Distribution, habitat & conservation status 6

1.4 - Eidonomy 7

1.5 - Reproduction 8

1.6 - Ecology: nutrition & daily activities 9

1.7 - Particularities of Guinea baboons' social system & related behaviours 9

2. Gastro-intestinal parasites found in Guinea Baboons 12

2.1 - Protozoa 13

2.1.1 - Flagellates: description & species of interest 13

2.1.2 - Amoebas: description, evolution cycle, SOI 14

- *Entamoeba histolytica* 15

2.1.3 - Coccidia: description & SOI 16

2.1.4 - Ciliates: description & species of interest 17

- *Balantioides coli* 17

2.2 - Helminths 18

2.2.1 - Trematodes: description & life cycle 18

2.2.2 - Cestodes: description & life cycle 19

2.2.3 - Nematodes: description, life cycle & species of interest 20

- *Trichuris* spp. 20
- *Strongyloides* spp.*: S. stercoralis* and *S. fülleborni* 21
- Spriruroidea*: Streptopharagus*, *Physaloptera* & *Protospira* species 24
- Strongyles*: Necator americanus, Oesophagostomum* & *Trichostrongylus* 26

3. Non-invasive methods to diagnose intestinal parasites of Baboons 29

3.1 - Basic sampling rules 29

3.2 - Preservation, fixation 29

3.3 - Direct smear 30

3.4 - Flotation methods 30

3.5 - Sedimentation method 32

3.6 - Baermann's larval extraction method 32

3.7 - Copro-cultures 33

3.8 - Colorations 34

3.9 - Molecular diagnostic methods 34
Part 3. Goals of this study 35
Part 4. Study site & subjects 36
1. Research area 36
1.1 - Niokolo Koba National Park, Senegal 36
1.2 - Simenti: the research site & its climate 37
2. Study group & subjects 41
2.1 - Demography 41
2.2 - Tracking 41
Part 5. Field work: materials & methods 43
1. Faecal samples 43
1.1 - Sample collection in the field 43
1.2 - Storage, ID & shipping 44
2. Coprological study 44
2.1 - In the field: Flotation method 44
2.2 - In the lab: Flotation, Concentrated sedimentation & Natural sedimentation methods 45
3. Molecular study 46
3.1 - DNA isolation 46
3.2 - Checking DNA concentration of isolated samples 47
3.3 - Amplifying DNA 47
3.4 - Gel electrophoresis, purification & sequencing 48
3.5 - Phylogenetic analysis 48
Part 6. Results 49
1. Coprological work results 49
1.1 - Identified parasites 49
2.2 - Prevalence, semi-quantitative evaluation & richness 51
2.3 - *Balantioides*-like ciliates 52
3. Molecular work results 53
3.1 - Spectrophotometry evaluation of DNA isolation 53
3.2 - Electrophoresis analysis of DNA amplification 53
3.3 - Sequencing results 54
3.4 - Phylogenetic analysis 55
Part 7. Discussion 56
1. About the methods: comments, errors & improvements 56
Sensitivity of methods: 56
Optimisation of conservation: 56
To better identify parasites: 57
To optimise the molecular analysis: 57
Assessment of severity of parasite burden in individuals: 57

Comparing seasonal variations: 58
Looking for correlations between parasites and hosts: 58
2. Comparing results with previous studies & interpretations 58
2.1 - *Entamoeba* spp. 58
2.2 - Trichuris eggs 58
2.3 - Spiruroidea 59
2.4 - Strongylida 59
2.5 - *Enterobius* sp. 60
2.6 - *Strongyloides* spp. 60
2.7 - *Balantidium-like* ciliates 61
2.8 - Absent parasites 61
3. Extrinsic factors which could influence parasite infection 62
3.1 - Spatial parameters 62
3.2 - Climactic parameters 62
3.3 - Fauna & flora of the environment 63
4. Intrinsic factors which could be linked to parasite infection 63
4.1 - Immunity 63
4.2 - Age 64
4.3 - Reproductive physiology 64
5. Zoonotic importance 65
5.1 - *Zoonotic agents of the Simenti baboons* 65
5.2 - Transmission risks 65
5.3 - Phylogeny: Hypothesis on Strongyloides evolution 66
5.4 - Prevention & control measures 66
5.5 - Use of the zoonotic potential in favour of development 67
6. Behaviours 68
Part 8. Conclusion 70
Part 9. Bibliography 73
Acknowledgements 83
Annexes 85

Abbreviations

%: per cent
°C: degrees Celsius
µm: micrometers
Ab: antibody(ies)
Ag: antigen(s)
BZA: benzimidiazoles
CRP: Centre de Recherche de Primatologie (Primatology research center)
DNA: deoxyribonucleic acid
DPZ: Deutsches Primatenzentrum
ELISA: enzyme-linked immunosorbent assay
EPG: eggs per gram of feces
FEC: faecal egg count
GI: gastro-intestinal
IF: immuno-fluorescence
IH: intermediate host(s)
LOQ: limit of quantification
mL: milliliter(s)
MLST: multilocus sequence typing
mm: micrometers
n: number of samples
PCR: polymerase chain reaction
pers. obs.: personal observation
PNNK: Park National du Niokolo Koba
RBC: red blood cells
SOI: species of interest
sp(p): species(plurial)
SSA: Sub-Saharan Africa
STH: soil-transmitted helminths

Abstract

Parasitological studies have been lead on numerous baboon groups but little is known concerning Guinea baboons, probably due to remote geographical locations and rapid decline of wild population caused by anthropogenic pressure.

The primary aim of the study was to investigate the presence, composition and prevalence of gastro-intestinal parasites harboured in a wild population of Guinea baboons and whether the findings converge with the previous studies done using samples from unidentified individuals, or with other baboon populations in Africa.

159 faecal samples were collected from 95 known individuals from 3 main habituated parties of Guinea baboons (*Papio papio*) living in the Niokolo Kobe National Park of Senegal. From these, 75 were partially or totally analysed.

Coproscopic examination was conducted under the microscope in order to analyse the presence of gastro-intestinal parasites' cysts, larvae or eggs. From 5 types of helminth eggs found, only three were identified to genus: *Strongyloides* sp., *Trichuris* sp. and *Enterobius* sp., whereas the two other types could not be further identified only based on morphological observation under the microscope: spirurid eggs and strongylid-type eggs. Additionally 2 types of protozoa were found: *Entamoeba* sp. and curious *Balantioides/Buxtonella*-like ciliates.
Molecular methods performed on 54 samples allowed identification of *Strongyloides stercoralis, Strongyloides fülleborni, Necator americanus* and *Oesophagostomum* sp. DNA was isolated and analysed with Sanger sequencing to compare phylogenetically the Strongyloides sequences with those currently found in the GenBank.
Although some differences have been observed, the overall results support the diversity reported by previous studies. Taking into consideration the optimisation of methods discussed, these results - along with the rest of the samples yet to be analysed - can serve as a base for longitudinal parasitology studies on this particular population.

These parasites are transmitted either directly via faecal-oral/cutaneous route (soil-transmitted) or trans-placental/mammary route or indirectly via intermediate hosts. By sharing habitat and resources with humans, livestock and other wild species, Guinea baboons harbour parasites which have high zoonotic potential as well as inter-species transmission risks.

On top of shedding light on multi-host-parasite interactions, understanding the role of parasites in the host's behavioural ecology, exploring baboon socialness and solving public health issues, parasitology must be taken into account when addressing wildlife conservation issues.

Part I. Introduction

Baboons are a captivating animals, especially regarding their adaptability in many fields.

1. Impacts of human encroachment

Baboons are the most widespread non-human primates (NHP) around the planet, inhabiting a variety of environments, from remote deserts to village surroundings. In a world where human population growth and consumption are never ending, human encroachment on the surrounding nature is continuously modifying the human-wildlife interface. Environmental modification, habitat fragmentation or destruction, poaching, international trade of animals, illegal commerce of animal products (such as meat) and tourism (even ecotourism) keep contributing to increased anthropogenic pressure on wildlife populations, including Guinea baboons who have seen their population rapidly decline [1].

Due to the close phylogenetic relatedness humans have with NHP, increased levels of contact between humans and animals multiply inevitably the potential of sharing diseases. Importance of this issue is already supported by the large number and wide range of pathogens currently shared with NHP [2,3]. Viruses include the wide spread Enteroviruses A and B (causing diarrheal diseases), the yellow fever virus, Herpes papio 2 viruses (causing flu-like symptoms but also potential paralysis and fatal meningoencephalitis in humans), simian retroviruses, SV40 polyomavirus, monkeypoxvirus, encephalomyocarditis virus (which is fatal in baboons but not humans) and many more [4]. Bacteria such as *Mycobacterium leprae*, *Shigella*, *E.coli*, *Campylobacter* sp., *Salmonella* sp. are also shared by both humans and NHP. Treponematose caused by *Treponema pallidium* is observed in many NHP in West Africa including baboons in Senegalc[5].
Shared parasites include *Plasmodium* sp. (causing malaria), *Trypanosoma cruzi* (causing Chagas disease), Giardia, *Cryptosporidium* sp., *Entamoeba histolytica* (causing severe dysentery), etc… A large variety of gastro-intestinal (GI) are shared, with special attention brought on soil-transmitted helminths (STH) which affect around 1.5 billion people worldwide [6], especially in areas with less sanitation and poor access to potable water [7].
It is estimated that over 70% of emerging pathogens of zoonotic importance have a wildlife origin [8]. But transmission can also occur in the other direction, from humans to NHP. In the 80s, a tuberculosis outbreak resulted in high death rates in a troop of olive baboons in Kenya. The outbreak was due to Mycobacterium tuberculosis originating from beef meat from an open bin in a tourist lodge [9]. Similarly epidemics of polio, respiratory diseases and scabis ariginating from humans have affected NHP [10,11]. Human coronaviruses have also been reported in baboons in Saudi Arabia.

Human proximity and related activities may also alter parasite community of wildlife. For example, parasites present in macaques living in human-modified environments were not found in more isolated groups [12]. Additionally, human encroachment pushing animals away and modifying their

geographical movements can lead to areas concentrated in wildlife, where parasitic cross-species transmission risk is increased. Certain species may then act as transport hosts or reservoirs for sympatric animals, sometimes threatened or endangered. For example baboons can transport pathogens to Chimpanzees in Senegal [13]. Threats on wildlife conservation are therefore obvious. In order to study primate health for conservation aspects, it is important to study wild and more geographically isolated populations such as the Guinea baboons in Senegal.

2. Research models

Baboons are established as good research models. They are used for experimental biomedical studies including heart & lung diseases, bone diseases, diabetes, stem cell therapy, organ transplantation, genetics, HIV vaccination, hepatitis C, respiratory syncytial virus, neonatal research, etc.. [4].

Baboons are also used in order to study early human adaptations because they evolved in African savannas alongside ancestral hominins, and to study social evolution. Most research is conducted in captivity but field studies are also conducted on wild baboons. However, due to their geographic isolation, wild Guinea baboons are subjects to less studies than other baboon species.
Recently, more research projects are oriented towards understanding relationships between social behaviours, health and fitness. Indeed, pathogen infections can cause pathological lesions like diarrhoea, anaemia, pain, respiratory issues and in some cases, death. Although infections often do not lead to clinical symptoms and do not seem to be a major contributing factor to mortality of baboons, they may have an effect on overall health and therefore interactions with peers. On top of that, pathogens may impose important selective pressures on wild NHP populations. For example female olive baboons copulate less with males showing clinical infection with *Treponema pallidum* [14]. Long term studies have also suggested that fitness of female baboons is enhanced by sociality [15]. For these studies, Guinea baboons are ideal subjects due to the complexity of their social system.

Additionally, scientific research has begun to explore ecological relationships NHP are involved in. On top of immunological mechanisms which NHP posses, behavioural strategies seem likely to be used to avoid pathogen exposure [16]. As primatologists and ethologists try to understand time-and-energy budgeting, daily activities and movement, social interactions, as well as peculiar behaviours, through longitudinal studies, parasitology must be taken into account. For example yellow baboons seem to avoid STH eggs by changing sleeping sites [16]and hamadryas baboons digest leaves and berries which are toxic to trematodes [17].

Finally, in-field research on feeding habits of primates, disease ecology and so on, is also helping shed light on parasite biodiversity and investigations of life cycles yet unknown.

Part 2. Literature review

1. Guinea Baboons

1.1 - External systematics of the genus *Papio*

Following the classification of Wilson and Reeder (2005), baboons (*Papio* spp.) belong to the phylum *Chordata*, class Mammalia and order Primates (characteristics of taxa are described by Mivart, 1873) [18]. Baboons are further classified (**Error! Reference source not found.**) in the sub-order Haplorhini due to the presence of a postorbital plate, in contrast to the postorbital bar found in strepsirrhines (Groves, 2001). They are part of the infra-order Simia (from the Latin "monkey-shaped") and the parv-order Catarrhini, referred to as "Old world monkeys and apes, including humans". Within this clade, baboons belong to the family Cercopithecoidea, to the subfamily Cercopithecinae based on their cheek pouches and ischial callosities, and to the Tribe Papionini.

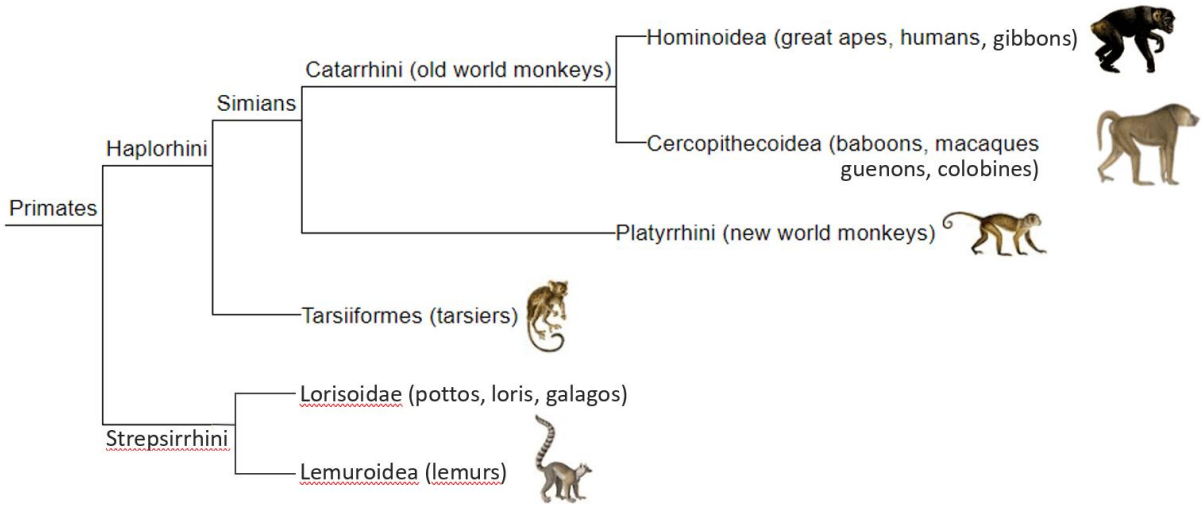


Figure 1 - "Cladogram" of living primates. *Modified from source: Wikipedia.*

1.2 - Internal systematics of the genus *Papio*

Generally, six distinct morphotypes or taxa are recognised within the genus. Between adjoining populations hybridisation may occur and long-lasting hybrid zones can be found [19].
Depending on the applied species concept these six taxa are ranked as subspecies (Biological Species Concept, Mayr 1942) or as species (Phylogenetic Species Concept, Cracraft 1983): *P. (h.) papio* (Guinea baboon), *P. (h.) anubis* (olive baboon), *P. (h.) cynocephalus* (yellow baboon), *P. (h.) kindae* (Kinda baboon), *P.(h.) ursinus* (chacma baboon), *P. (h.) hamadryas* (Hamadryas baboon)[20].

1.3 - Distribution, habitat & conservation status

The geographic distribution of baboon species is quite extensive across Sub-Saharan Africa (SSA) and the South-west of the Arabian Peninsula (**Error! Reference source not found.**). Guinea baboons, *Papio papio* (Desmarest, 1820), only occupy a small portion of this distribution and are endemic to West Africa with populations located in parts of Gambia, Senegal, Guinea-Bissau, Guinea, western Mali, southern Mauritania and potentially north-western Sierra Leone [21].
Scientific studies on wild Guinea baboons are less numerous than for other baboon species and are - to our knowledge- limited to those in Guinea-Bissau [6,7] and Senegal [5,13,23–32]. Most of the Senegalese population is concentrated in the Parc national du Niokolo Koba (PNNK). Outside the park, populations in Boundou and Casamance seem to be declining [21].

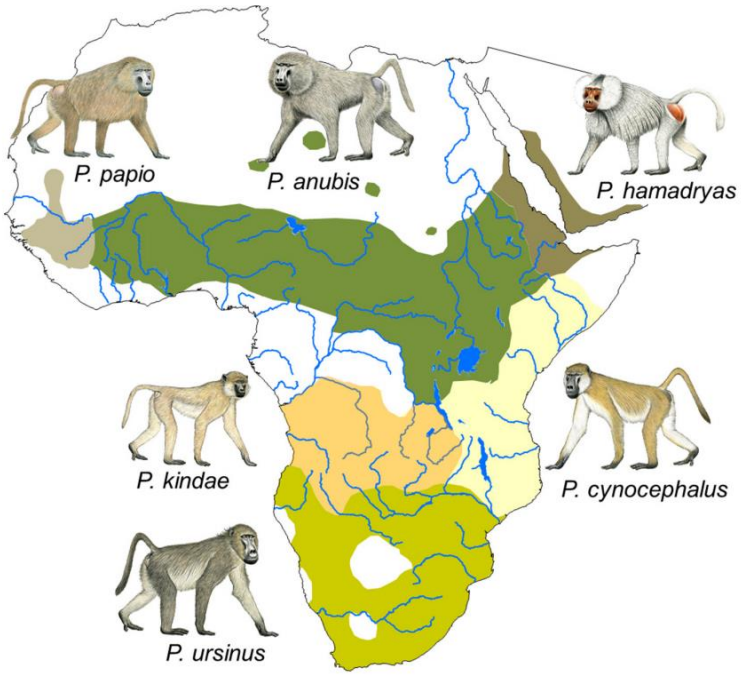


Figure 2 – The male phenotypes and geographic distribution of *Papio* species.
Originating from Fischer et al. (2017) [33] *with permission. Drawings by Stephen Nash*.

Like the other baboons, Guinea baboons are found in a wide range of habitats including semidesert, savannah woodland, scrubland, gallery forest and mangroves [34–36]. The vegetation in their habitat varies greatly with seasonality and rainfall [37]. Guinea baboons show great ecological flexibility, and their occurrence is mostly limited by the availability of water, food & sleeping sites. In other baboons, these factors have been shown to affect home range size and use, as well as population densities [38–41].
To date, the ecological role of Guinea baboons in their habitats appear to be mainly in soil aeration (by digging of corms, roots, and tubers), in dispersal of seeds, as prey for their larger predators and as predators of smaller vertebrates and invertebrates which they sometimes feed on [29].

Guinea baboons have more recently become recognised as Near Threatened. They are listed under Appendix II of CITES, as well as Class B under the African Convention, and included in the Red List

of threatened species of Guinea-Bissau. In other countries, the species is protected only in some areas: in the Niokolo-Koba National Park (Senegal), the Boucle du Baoulé National Park (Mali), and in Outamba-Kilimi National Park (Sierra Leone), the "Lac Gabou et le Reseau Hydrographique du Plateau du Tagant" (Mauritania) [21].

Although they are common within the large protected areas where they occur, populations have rapidly declined outside these protected areas in the last 40 years due to environmental and anthropologic factors. These include (i) heavy hunting for local or urban (luxury) bushmeat consumption, for skin trade in folk medicine, for crop protection, by military against a salary [42] but also (ii) capturing for local trade of juveniles as pets, for international export as biomedical research subjects and (iii) destroying their habitat for human encroachment and agricultural practices, as well as (iv) severe climate changes inducing extreme weather conditions.

1.4 - Eidonomy

Baboons are among the largest Old world monkeys with body masses ranging from 14 to 41kg.
They express a very obvious sexual dimorphism with males up to 50% heavier than females, and in some species, have coat variations.

Also called "Red baboon", Guinea baboon males weigh on average 20kg whiles females weigh on average 12kg. Sex-differentiation can also be seen in their prominent ischial callosities (patches of keratinised skin ground-dwelling baboons sit on) which are continuous in males and separate in females. Females also show perineal tumescence of their anogenital sex skin, which varies in volume & colour due to cyclic ovarian- or gestation hormonal fluctuation.

They have a dense reddish-brown coat, with some geographic variation: baboons in the western part of the range seem lighter and redder whereas those in the eastern part appear darker and more brown. As opposed to some other baboon species, they display the same colour fur on back, arms, legs, abdomen and head [43]. Like in most other baboon species, infants have a characteristic black natal pelage, which gradually changes into the adult colour within the first three to six months.
Guinea baboon males also have a shoulder mantle similarly to *P. hamadryas* but less pronounced [44].

Guinea baboons have a medium size head with a blackish red face, rounded ears, close-set eyes under a brow ridge and lighter-coloured eyelids which they flash to communicate. They have a long prominent hairless muzzle (shaped like that of dogs) ending in nares set close together (the Latin origin of the Catarrhini parv-order is "narrow turned down nostrils"). Guinea baboons have cheek pouches that are extensions of their cheeks which can extend below each mandible ramus to quickly store food for later eating. Their powerful jaws contain 32 teeth (dental formula: (4I +2C + 4 PM + 6M) x2) and especially in males, upper canines are relatively long and sharp.

They have a moderately long arched tail like in hamadryas baboons, long arms and legs and 5-fingered/toed grasping hands/feet.

Figure 3 – Comparaison of morphology of adult male, adult female and juvenile baboons *(Photos: A. Coles)*

1.5 - Reproduction

The lifespan of Guinea baboons is still unknown but can be assumed similar to that of wild yellow baboons averaging 20 years old [45].

Like other baboon species, they do not show a pronounced seasonality in their reproduction. Females have menstrual cycles all year round and the intermenstrual interval is reported between 26-34 days [46]. The sex skin of adult females swells and reddens according to these cycles; turgescence is associated with ovulation, allowing other members to be informed of their status.
Interbirth interval lasts about 13 months and gestation lasts around 6 months (179-186 days) [46]. Like in humans, the single haemochorial placenta usually carries a single embryo.

Baboon neonates are born with their eyes open and are carried in a ventral-ventral position by the mother for several weeks before riding dorsally. Infants start exploring their environments at 2 months old, are weaned at around 6 months old and juveniles reach sexual maturity at around 4 years old, which is younger than other baboon species [46]. They disperse to other groups as adolescent or young adults.

Figure 4 - Evolution of neonates into infants with visible change of skin and coat colour *(Photos: A. Coles)*

1.6 - Ecology: nutrition & daily activities

Guinea baboons are diurnal, primarily terrestrial, quadruple primates. They do nevertheless climb trees for eating fruit, playing and sleeping. The size of their home range (HR) is estimated to 20.0-42.0km² and their daily travel distances (DTD) are 0.5-13.0km [34]. Shifting of HR area, modification of HR size and variation of DTD show a behavioural flexibility. It has been suggested that the behavioural flexibility of baboon species is a response to the quantity and quality of food and water resources available according to seasonality[7, 14,16,17] and predation risk [1].

Like other baboon species, Guinea baboons are omnivorous but the bulk of their diet is fruit [25]. They range most of the day in order to feed on a variety of plants: dry or fleshy fruit, tubers, bulbs, rhizomes, flowers, grasses, leaves, twigs, bark, seeds, tree gum. Their diet also comprises invertebrates and small vertebrates like birds, reptiles, hares, infant antelopes, smaller monkeys… [29]. The will opportunistically feed on human food left as "traps" for tourists to observe wildlife or left-overs from tourist/military/scientific camps (*pers. obs.*) and may raid a variety of crops [1] when in close contact with agricultural land.

Regarding other baboon species, the ranging behaviour is also dependant on distribution of sleeping sites [48]. They prefer to sleep in closed-canopy trees but occasionally steep cliffs or caves (Marais 1939), as these provide protection from predators such as wild felids, hyenas, dogs, chimpanzees, crocodiles and raptors [29,34,39–41]. A large Guinea baboon group may spend the night split between a couple trees. Each small unit situate themselves on a branch, far from the trunk (especially females and juveniles).

The use of caves has been reported in *P. ursinus* as water sources, thermoregulation [49], in *P. ursinus* and *P. anubis* and to escape predators [50,51] and in *P. anubis* [52] , *P. papio* and *P. ursinus* [26] for suggested salt-licking purposes.

Figure 5 - Daily activities include traveling, feeding in trees, occasional meat eating *(Photos: A.Coles)*

1.7 - Particularities of Guinea baboons' social system & related behaviours

The social organisation of baboons shows some diversity. Hamadryas baboons live in a multi-level society, based on one-male units with female-biased dispersal [22]. Olive yellow, chacma and Kinda baboons live in uni-level societies, where related females are the core of the groups and males disperse

to other groups after reaching maturity. The social organization of Guinea baboons remained unclear until recently.

Dunbar & Nathan (1972) observed a multi-male social organisation with sub-structuring into groups of which the composition changed throughout the day. Sharman (1982) reported that as opposed to other baboon species, adult females did not seem constraint to mate with one and only male. He also noted male-male grooming, which is uncommon in other baboon species. Studies on Guinea baboons at Simenti, in the Niokolo-Koba National Park revealed that the baboons live in a social system superficially similar to the multi-level system of hamadryas baboons. Their social organization is based on units, with one adult male and several females and their offspring [27,29,33]. The study population at Simenti of more than 40 baboons is organised into three levels:

- *Units* are composed of one primary male and one or more females (OMU) with immature baboons (infants, yearlings, juveniles). Sometimes one or more secondary males (MMU) may be present, who have social access but no sexual access to the present females.
- *Parties* consist of several core units showing social interaction and bonding, which join to forage, sleep and travel most of the time together.
- *Gangs* are formed by 2 or more parties showing more social interactions among their members than members of other parties.

Therefore "the system can be understood as having OMUs at the level of the mating system, and OMUs as well as MMUs at the level of the social organisation" [29]. This organization facilitates a certain fission-fusion patterns dynamic. At rich source large groups can forage together, but if resources are scattered, these large groups can easily fission into smaller units and exploit such resources without strong direct competition.

The complex multilevel social organisation relies on strong affiliative ties between individuals including related and familiar males. Intense, elaborate and reciprocated behaviours are described. On top of facial expression and vocalisation, physical contact is frequent between Guinea baboons. Surprisingly "intimate" greetings such are hind-quarter presentation, genital fondling and mounting are seen between males of all age categories, requiring tolerance and cooperation. Male-male, female-female but also female-male and male-female grooming sessions are very frequently observed, as well contact-sitting amongst affiliative individuals and playing among the younger individuals. Threats via physical aggression and fights are rare [15] notably as, in contrast to other baboon taxa, Guinea baboons do not show coercion towards females, who have more spatial and social freedom. Females may freely transfer between units, parties, gangs [15], and may also copulate with more than one male [25]. Parental care is also observed including breast-feeding, grooming and carrying.

A new field of studies is opened regarding social connectedness between baboon individuals: the research of parameters correlating with strong-links and weak-links. Such particular and numerous physical interactions observed give rise to questions notably regarding transmission of pathogens, such as parasites.

Figure 6 - a: Grooming session within units. **b**: Individual grooming and resting of a unit in a tree. **c**: Grooming in pairs and morning sunbathing. **d**: Different units around a water point. **d**: Several parties come together in open space.

2. Gastro-intestinal parasites found in Guinea Baboons

As Gillespie (2006) stated, "parasites play a central role in ecosystems, affecting the ecology and evolution of specific interactions, host population growth and regulation and community biodiversity". They also play a role in the host's social interactions with conspecifics and groupmates as they affect reproduction, nutrition and overall condition (fitness). Understanding life cycles, transmission routes, pathogenicity and host specificity is essential to understanding the impact of a parasite on its host [53]. To date, only few surveys have documented parasites in free-living Guinea baboons. Following an extensive literature review, available information regarding gastrointestinal parasites found in Guinea baboons is presented.

Baboons harvest a wide range of epizootics but the same parasites are often found in their faecal samples [54–63]. This correlates with the only three previous studies on gastro-intestinal parasite diversity and prevalence in Guinea baboons, all in South East Senegal (Figure 11 map). These studies were conducted at Lion Valley and Stella's Valley, Mount Assirik in the Niokolo Kobe Park (McGrew et al.,1989) [24], (Ebbert et al. in 2000-2012) [23] and in Fongoli Park (Howell et al. in 2005) [13]. Reported parasites and associated prevalence are summarized in Table 1 – **Diversity and prevalence of intestinal parasites of Guinea baboon from previous studies. *Values have been rounded to the nearest unit. U.I. = unidentified***Table 1.

Table 1 – Diversity and prevalence of intestinal parasites of Guinea baboon from previous studies.
Values have been rounded to the nearest unit. U.I. = unidentified. N = number of samples.

	Lion Valley, 1976-79 *N = 39*	***Lion Valley, 2000*** *N = 48*	***Stella's Valley, 2000*** *N = 52*	***Fongoli Park, 2005*** *N = 17*
Parasites	**Prevalence (%) per location**			
Nematodes				
Trichuris	28	85	92	35
Ascaris	0	4	2	0
Enterobius	0	42	37	
Physaloptera	31	0	0	0
Protospirura	0	70	62	
Streptopharagus	23	6	2	
Other U.I. spirurid sp.		4		
Strongyloides	26	2	4	41 (*S. fülleborni*)
Strongyle eggs		4	6	
Necator sp.	38			29
U.I. hookworm				53
U.I. nematode larvae		12 (maybe *S. fülleborni*)	10	0
U.I. *egg*		2	4	
Trematodes				
Schistosoma	23	0	0	26
Watsonius	0	54	35	
U.I. fluke eggs	0	0	4	65
U.I. *Stringeodea* sp.	44			
Protozoa				
Balantidium coli	72	90	60	12
Entamoeba coli	87			71
Entamoeba histolytica/dispar				47
Chilomastix mesnili				29

Troglodytella abrassarti		6
Troglocorys cava		6
Iodamoeba buetschlii	38	0

Based on the previous studies, the main categories of gastro-intestinal parasites susceptible of being found in wild Guinea baboons in Senegal are detailed here-under.

2.1 - Protozoa

Protozoa are protists (eukaryotic unicellular living beings) which are often mobile and with non-cellulosic walls. They are of small size: cysts are a dozen of micrometres and vegetative forms are up to 250µm. They have a well-defined nucleus and variable locomotory organs.

Protozoa may live freely in environment or may be facultative- or obligatory parasites.
Reproduction among adult forms (trophozoites) can be sexual or asexual.
Asexual reproduction may be via simple bipartition (mitosis), internal budding (endogeny) or schizogony (merogony). Sexual reproduction may be via gametogonia (an egg is produced when male microgamete and female macrogamete fuse) or via conjugation (2 trophozoite individuals fuse into one). Trophozoites are generally shed into faeces/excretions as resistant forms which can survive in the environment: either as cysts (within a thick shell) or as spores (formed inside the trophozoite). They are then taken up orally by the new host and produce more trophozoites which can be responsible for clinical signs of infestation.

In general, the monkey has a spectrum of enteric protozoa that is essentially a replica of the pattern of protozoa seen in man. All of the evidence points to the probability of interchange where contact between man and monkey is established. Morphologic evidence alone might be suspect, but over the years the utilization of the monkey for experimental studies has permitted the conclusion that cross infection occurs.

Although recent classification is based on DNA sequencing, it remains easier to classify Protozoa based on morphological features.

2.1.1 - Flagellates: description & species of interest

These protists present one or more flagella enveloped in a sheath which inserts on the cell membrane. Flagella are locomotory organs formed by a central axoneme (2 microtubules), surrounded by 9 pairs of microtubules.

They are classified into enteric flagellates or hemoflagellates depending on tropism-type.
In NHP, only enteric flagellates have been reported. These are transmitted by faeco-oral routes and many are non-pathogenic. Infestations are often asymptomatic but can cause diarrhoea and vomiting in NHP.

Based on morphological characters such as number of flagella, cystic form, etc.. 4 orders of flagellates are found: Kinetoplastida (not present in vertebrates), Retortamonadida (including *Chilomastix sp.*), Trichomonadida (not reported in baboons), Diplommonadida (including *Giardia sp.*).
SOI:

- *Giardia intestinalis* (syn: *G. lamblia, G. duodenalis*) are found as cystic form in host's faeces and environment, or as trophozoite infectious form in host's intestines. Cystic form is very resistant to dry, humid and cold conditions [64].

- *Chilomastix mesnili* (syn: *C.suis, C.hominis*) have a cosmopolitan distribution in NHP but also humans and swine. Trophozoite and cystic forms are shed into faeces. Via faeco-oral transmission, cysts progress into the host's large intestine and release trophozoites which reproduce and thrive in host's caecum and colon.

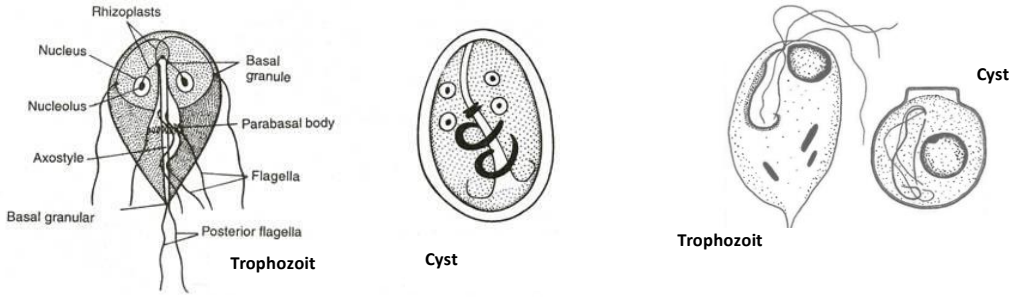


Figure 7 - *Left*: Both forms *Giardia intestinalis (Taylor et al., 2016); Right*: Forms of *Chilomastix* sp. *(Bussieras & Chermette, 1992)*

2.1.2 - Amoebas: description, evolution cycle, SOI

These protists are characterised by pseudopod-type locomotory organs and generally asexual reproduction. Most species are free-living and non-pathogenic. The following pathogenic species present enteric tropism, infest the caecum and colon, and may cause severe enteric diseases in NHP and humans. Based on the nucleus' aspect (endosome and perisomes), 3 genera are differentiated: *Entamoeba, Endolimax* and *Iodamoeba*.

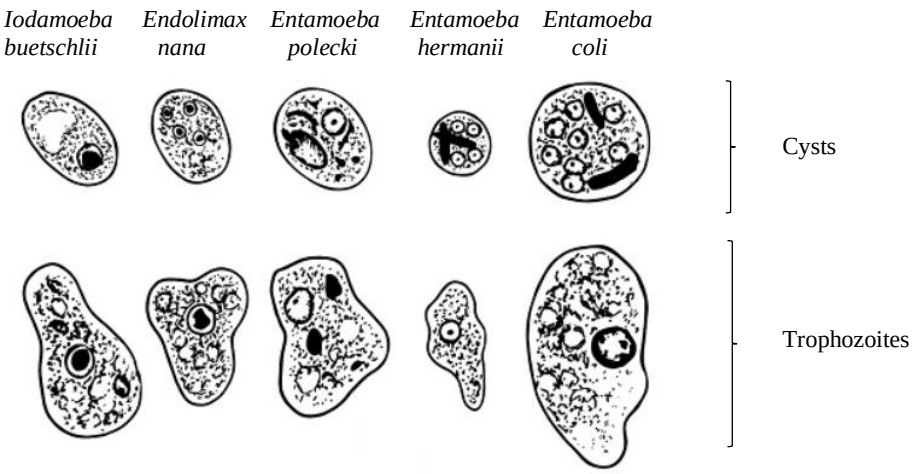


Figure 8 - Pathogenic Amoeba species (*Modified from source: www.CDC.gov/)*

- *Entamoeba histolytica*

From all the *Entamoeba* species, it is the only pathogenic one. *Entamoeba histolytica* has a worldwide distribution and is commonly reported in many New world monkeys, Old world monkeys and apes. Several studies have recorded relatively high prevalence in various African NHP [2,13,65–68] but infections are rarely pathogenic. *E. histolytica* has been reported in Guinea baboons by Howells et al. (2010), but also in olive and chacma baboons [62,69–71].

LIFE CYCLE

The lifecycle of *E. histolytica* is direct, but insects such as flies and cockroaches may serve as mechanical carriers of the amoebic cysts [72].

Transmission between hosts occurs via various forms of contact [73], and evolution cycle is complex, passing through various stages. Trophozoites of *E. histolytica minuta* are commensally in intestinal lumen of host. They evolve into cysts which mature in the intestines or outside the body. They reproduce by binary fission so that each cyst divides into 4 trophozoites [74]. These may transform into pathogenic *E. histolytica histolytica*, the second vegetative form. Pathogenicity depends on host species, nutritional status, environmental factors and bacterial gut flora.

MORPHOLOGY

E. histolytica histolytica trophozoites measures 20-40µm in diameter and contain ingested red blood cells (RBC). They are slightly larger than the *E. histolytica minuta* trophozoites and than the related non-pathogenic *E. dispar* which don't ingest RBC. Cysts formed in the large intestine are uninucleate or binucleate. Mature cysts measure 10-20µm in diameter, contain 4 nuclei and rod-like chromatin bodies [75].

CLINICAL SIGNS

Generally living in the intestinal lumen, *E. histolytica* is non-pathogenic but in heavy infections, the mucosa is invaded, causing amoebic dysentery. Symptoms include severe enteritis, haemorrhagic or catarrhal diarrhoea, dehydration, apathy, anorexia, lethargy [76].

EPIDEMIOLOGY

E. histolytica ranks third in worldwide causes of human morbidity by parasitic infections [56] causing dysentery and colitis [78]. Humans are thought to be the primary reservoir of this pathogen but it can also infect NHP and sporadically dogs, cats and swine.

DIAGNOSIS

Cysts may be seen in large numbers in faecal samples under the microscope in direct or flotation faecal examination. Staining [79] with trichrome, Giemsa, Lugol and iron haematoxylin is very useful to reveal the cysts. *E. histolytica* and *E. dispar* are morphologically similar and cannot be differentiated by standard microscopic examination, so molecular work is required for accuracy.

TREATMENT

Treatment consists of Metronidazole administration, possibly with diiodohydroxyquin. tetracyclin, chloroquin, chloramphenicol and paramomycin are also possible treatments.

2.1.3 - Coccidia: description & SOI

Coccidia are Sporozoans; obligate pathogens without any locomotory organs but with an apical complex situated at their anterior end, allowing penetration into host cell. Coccidia have the ability to produces spores and as opposed to other Sporozoans, are never transmitted by hematophagous arthropods. They generally present enteric tropism but gastro-intestinal infection with most of the frequently reported Coccidia in NHP do not cause lesions or diseases. From the Eimeriida order, 2 families with homoxenous cycles contain species pathogenic to NHP including baboons:

- *Cryptosporidium* spp.: Mature sporulated infectious oocysts are shed in the faeces and remain resistant in dry and wet conditions (up to months long). They are ingested orally and release sporozoites which develop via schizogony on epithelial cell surfaces of ileus and are responsible for cryptosporidiosis, a zoonosis of major importance due to its worldwide distribution, low host-specificity and high morbidity rate in humans [64]. Symptoms are seen in younger monkeys and include gastroenteritis, profuse diarrhoea, weight loss, dehydration [80]. In baboons, only *C. hominis* has been reported [59].

- *Cyclospora* spp: Similar evolution cycle but develop inside the epithelial cells of guts. Present in many NHP including apes and humans, causing similar digestive symptoms to *Cryptosporidium spp*., but often seem specie-specific [81]. In baboons, only *Cyclospora papionis* has been reported [82] and seems common [83].

- *Isospora* spp : biggest diversity of hosts but generally specie-specific. They develop inside the epithelial cells of host's guts but their presence hasn't been corelated to any clinical signs. *Isospora papillonis* has been reported *P. ursinus*.

2.1.4 - Ciliates: description & species of interest

Ciliates are protists characterised by numerous short hair-like organelles called cilia, located all arranged in rows (kineties) around the cell, and used to swim, crawl, attach, feed and feel. The cell contains 2 nuclei: a smaller diploid micronucleus assuring reproduction and a larger polyploid macronucleus assuring the vegetative state of the cell. Food vacuoles and contractile vacuoles are formed when food and water is passed through the cell.
Ciliated protists are found wherever there is water and although the majority are free-living or symbiotic, some species may be parasitic. In all baboons, other NHP and humans, only *Balantioides coli* is known to cause disease [84].

Troglodytella abrassarti and *T. cava* have also been reported in Guinea baboons [13] and apes, but do not seem pathogenic.

- *Balantioides coli*

Balantioides coli is found worldwide as a commensal protozoa of caecum and colon of many animals including swine (primary reservoir), dogs, ruminants, horses, rats and many primates [76]. In baboons, high prevalence is often recorded [65].
B. coli is the only pathogenic ciliate and the largest protozoa that parasitizes humans, however the pathological risk for wild NHP has not been demonstrated [13].

LIFE CYCLE

Balantidium coli is found in two developmental stages: cysts and trophozoites.
Infection occurs through faeco-oral transmission route as cysts are ingested, often via contaminated water. In healthy hosts, they are destroyed by acids in the stomach. But if not, once they reach the small intestine, they produce trophozoites which then colonise the large intestine. This requires an adaptation period to adjust to the host's flora, which they feed on. Sometimes trophozoites enter the mucosa and multiply via binary fission, whiles others undergo encystation in the lumen as bowel content is dehydrated. Mature infectious cysts are shed in the faeces and released into the environment. Evolution cycle is shown in Annex 1.
Pathologic infection occurs only when the balance between *B. coli* and host's flora is broken, due to factors such as malnutrition, compromised immune systems, etc.. [85].

MORPHOLOGY

Balantidium coli trophozoites are large ciliated ovoid cells measuring 30–150 µm long and 25–120 µm wide [86]. Their surface is covered with cilia and are able to move around. They contain very small micronucleus and a kidney shaped macronucleus, along with vacuoles.
Cysts are spherical or ovoid and measure 40-60µm in diameter. A tough multilayered shell protects them against stomach acid of host, when ingested. Unlike trophozoites, cysts cannot reproduce and do not have any cilia available for moving.

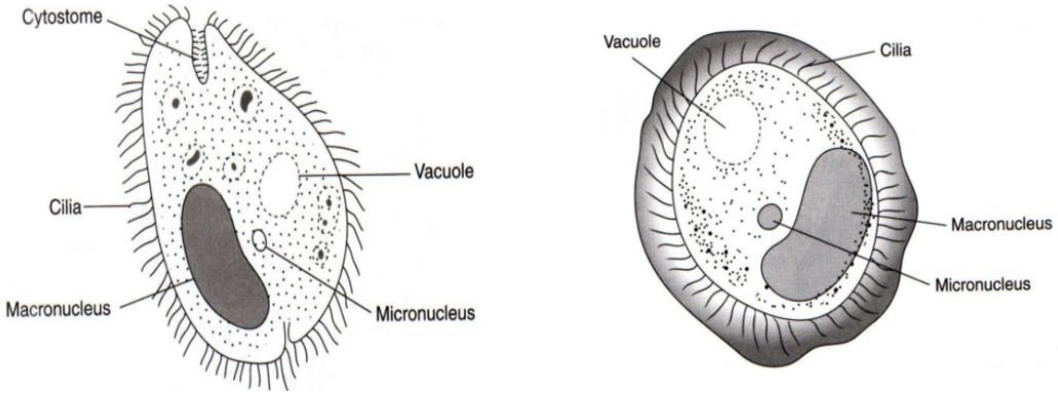


Figure 9 - Balantidium coli stages. *Left* : trophozoite form. *Right* : cyst form.
Extracted from: "Essentials of Human Parasitology"

CLINICAL SIGNS

Generally asymptomatic but some individuals develop balantidiosis = dysentery with explosive diarrhoea and weight loss associated with bloody faeces, tenesmus, fever, pain. Life-threatening ulcerative enterocolitis and colon perforation has also been reported [76,87].
Clinical symptoms of Balantidiasis include diarrhea, dysentery, and colitis [88].

TREATMENT

Balantidiosis can be effectively treated with metronidazole, tetracycline or diiodohydrowyquin in NHP as well as humans. Prevention includes proper hygiene practices and cooking contaminated food.

2.2. Helminths

There are 228 species of helminth parasites of primates including 166 of nematodes, 24 of trematodes, 33 of cestodes and 5 of acanthocephales [89]. In baboons, the following are found:

2.2.1 - Trematodes: description & life cycle

Trematoda, also called flatworms, are flat, non-segmented and big helminths (reaching up to 10cm long), covered in a tegument. They all have 1-3 suckers: a buccal sucker, often a ventral or dorsal sucker, and sometimes a genital sucker. All except some Schistosoma are hermaphrodites (Bussiéras & Chermette, 1995), without sexual dimorphism. Based on these morphological characters, around 9000 species are classified into 5 groups : Distoma, Amphistoma, Holostoma, Monostoma, Schistosoma.

Most trematodes have complex life cycles. Following (auto-) fertilisation in terminal host, eggs are shed into faeces / secretions. Hatching of swimming miracidia (triangular embryos) takes place in the environment. To survive, miracidia have a few hours to find and penetrate a gastropod mollusc[10,67] – a snail – as intermediate hosts (IH), in which they will transform into sporocysts, enter a germinative state and become redia (larvae). These larvae can mature into one or multiple cercaria which leave the IH into the wet environment. Cercaria will either enter terminal host (TH) through skin penetration, or fixate onto plants and transform into metacercaria which will be eaten by a herbivore TH, or penetrate a second IH and transform into metacercaria which will be eaten by an omnivore/carnivore TH. Once in TH, sexual maturation takes place, leading to an adult form of the trematode.

Many species of trematodes are found in NHP but only several genera have been recorded in baboons, including Guinea baboons [13,24,62,90]. Species of interest are therefore: *Watsonius* spp., *Fasciola hepatica*, *Schistosoma* spp. (blood flukes). These infestation be associated with gastro-intestinal symptoms such as (bloody) diarrhoea, severe enteritis, and death, or systemic symptoms including pyrexia, haematuria, ascites.
Diagnosis is based on observation of characteristic eggs in host's faeces or adult worms in host's organs/blood vessels/body cavities during necropsy (in the case of Schistosomiasis, female and male worm are usually found attached in constant copulation).
No treatment is reported for Trematoda infections except for schistosomiasis which, due to its public health significance, can be treated with Praziquantel.

2.2.2 - Cestodes: description & life cycle

Also called Tapeworms, these platyhelminths have very long, flat and segmented bodies. The anterior end of a tapeworm has a scolex with hooks or suckers to anchor into the host. Cestodes are osmotrophic: they have no digestive system but their body is covered in a tegument with microvilli which allows high ability to absorb nutrients from host. Tapeworms are hermaphrodite. Body segments, called proglottids, are independent reproductive units (containing both male and female genital tracts), ranging from immature (near the scolex), to mature and gravid (posterior end): containing numerous eggs.. Species are determined based on egg morphology or on adult worm characters such as scolex features, location of uterus opening and uterus shape.

The complex lifecycle of Cestoda requires 1 or 2 intermediate hosts. Once completed, adults reside in the small intestines of the vertebrate TH such as ruminants, domestic carnivores and NHP. The only identified Cestoda in baboons are *Hymenolepis nana* in *P. anubis* [62] and *Bertiella* spp. in *Papio papio* [13] and *P. ursinus* [91], which rarely affects humans. Although these parasites may infect baboons in large numbers, enteric lesions and clinical signs such as diarrhoea and abdominal pain, are unfrequently associated. Diagnosis is based upon identification of eggs or proglottids in TH's faeces, or the presence of adult worms at necropsy. Niclosamide, bunamidine hydrochloride and praziquantel can be used to treat the infection.

2.2.3 - Nematodes: description, life cycle & species of interest

Nematodes are non-segmented round worms present in abundance, which may live freely or infect many animal and plant species [92]. The genus and/or species can be determined based on the clear sexual dimorphism: females are larger and males present a copulatory apparatus.
Nematodes have a thick flexible cuticle covering epidermis and longitudinal muscles and present a pseudocoel mostly filled with an intestine and oviducts or testes. They generally have papillae, setae and amphids as main sense organs. Setae detect motion (mechanoreceptors), while amphids detect chemicals (chemoreceptors) [93]. Nematodes reported in primates and in particular baboons, are all within the Secernentea class. These worms have plasmids; unicellular glands which likely function as chemoreceptors. Females may produce pheromones to attract males.

The Nematoda phylum consists of 6 orders: Rhabditida, Sprirurida, Strongylida, Ascaridida, Oxyurida and Enoplida, including species of interest (SOI) in case of baboons: *Trichuria* spp., *Capillaria* sp. *Oxyuria* sp., *Strongyloides* spp, *Ascarides* sp., strongyles, sprirurids. These SOI contain some of the most frequent human pathogens including STH, infecting over one billion people worldwide, mostly in tropical & subtropical regions such as SSA [94].

STH are nematodes which shed eggs in the host's faeces and are transmitted via the soil in which eggs and/or larvae can survive long periods and variable conditions [95]. Once the soil particles ingested or in contact with the skin, these parasites continue their lifecycle causing infections.
Whipworms (*Trichuris trichuria)*, Hookworms *(Necator americanus, Ancylostoma duodenale)* and roundworms *(Ascaris lumbricoides)* are considered of global importance, but other less studied endemic parasites have been proven to be as important *(Oesophagostomum spp.).* The species of interest are detailed here-under.

- *Trichuris* spp.

  Known as whipworms due to their shape, these STH are the third most common nematodes infecting animals & humans. In primates, *Trichuris trichuria* has been found in *H. sapiens* [96], *Pan troglodytes* [97], *M. sylanus* [98] and *P. papio* [99]. Other primates harbour different Trichuris species for example *T. ursinus* found in chacma baboons [100]. Although it has been assumed that those found in wild Guinea baboons in Senegal are *Trichuris trichuria,* evidence has been made that two distinct genotypes infect both humans and baboons [71,101], and that transmission routes between primates still need to be clarified [102].

LIFE CYCLE

Whipworms have a simple direct lifecycle with faeco-oral transmission. *Trichuris* eggs are ingested by the host and hatch in the small intestine. The larvae invade the villi and after 1-3 months they migrate to the caecum and ascending colon where they penetrate the mucosa and develop into adult worms. Anchored by the whip-like anterior part into the mucosa, the posterior part hangs loose to mate if a worm of the opposite sex passes by. 60 to 90 days later, eggs are

released into the faeces and shedding may last years. After being in contact with moist shady soil for 3 weeks, the eggs become infectious. Eggs may also be disperse in the environment by arthropods, wind and water. They are sensitive to sunlight and perish at extreme temperatures. Adult worms can live up to 5 years.

MORPHOLOGY

Eggs are brown, barrel-shaped, with mucoid polar plugs at both ends, usually measuring 50-70µm x 25-35µm. Ebbert et al. also described very similar but darker, smaller, wider eggs measuring 29-37µm x 15-19µm, also in Guinea baboons. Adult whipworms resemble handle & lash of a whip, are white-pink, and measure 20-50mm long [102].

CLINICAL SIGNS

Trichuris sp. infestation often cause asymptomatic chronic infection, but in very high intensity of infestations they can cause abdominal pain and distension, appendicitis, enteritis and bloody diarrhoea due to wounds of the intestinal wall, rectal prolapse, anaemia due to vitamin- and iron loss, weight loss, growth retardation [103].

EPIDEMIOLOGY

Trichuris trichuria have a worldwide distribution estimated at approximately 10% of the population, but occur mostly in tropical climates such as South Asia and Sub-Saharan Africa, concomitantly with two other common STH: roundworm (*Ascaris lumbricoides*) & hookworm (*Necator americanus* and *Ancylostoma duodenale*). Other whipworm species can infect domestic and wild animals including ruminants, pigs, dogs, cats, rodents.

TREATEMENT

Trichuriasis is often diagnosed by the presence of eggs in stool samples. It can be treated in humans with oxantel pamoate–albendazole [104] Mebendazole (which may alter the eggs' size and shape [105]) and Albendazole, and prevented in humans by good personal hygiene and washed food.

- *Strongyloides* spp.*: S. stercoralis* and *S. fülleborni*

Species from this genus belong to the Rhabditida order and Strongyloididae family. They are intestinal parasites with high-host specificity described in many vertebrate species [24,106] including ruminants, rats, pigs domestiques carnivores like dogs, as well as in most captive and wild primates: *Strongyloides stercoralis* is found in dogs and apes, *S. fülleborni* and *S. papillosus* are found in several Apes and Old world monkeys (Catarrhini), *S. simiae* is found only in old world monkeys and *S. cebus* is found in New world monkeys (Platyrrhini). Their distribution is cosmopolitan but prevalence is higher in subtropical and tropical areas [107]. *S. stercoralis* is the major causative agent of strongyloidiasis in humans, although there have been isolated zoonotic cases of *S. fülleborni* subsp. *fülleborni* [108] and subsp. *kellyi* reported [109,110].

LIFE CYCLE

Strongyloides species have a complex life cycle [111] including both a free-living cycle and a parasitic cycle, following a percutaneous transmission (as shown in Annex 2) or via lactogenic (mostly colostral) transmission. Infective filariform lavae in contact with the definitive host will penetrate its skin or oral mucosa and in under 9 days they migrate via lymphatic routes or more frequently haemo-tracheal routes (from bloodstream to the lungs, then coughed up and swallowed) to the small intestine to develop into female adult worms embedded in the submucosa. Via parthenogenesis, during up to 11 weeks, they produce eggs from which rhabditiform larvae hatch and migrate to the lumen. In case of auto-infection (homogonic cyle) [109], larvae will become filariform once again, penetrate the large intestinal mucosa or perianal skin and migrate to different organs. In the other case (heterogonic cycle), the rhabditiform larvae are excreted in the stool and develop into free-living male and female adult worms. Fertilised females produce eggs, rhabditiform larvae hatch and become infectious filariform larvae.

In contrast to *Strongyloides stercoralis*, *Strongyloides fülleborni* do not have the potential to multiply within the host via auto-infection and need a free-living cycle. Rather than larvae, eggs are shed into the faeces, hatch in the environment and develop into either free-living males and females or directly into infective filariform larvae, as shown in

Annex 3.

MORPHOLOGY

Strongyloides eggs are oval, symmetrical, with thin colourless shells, measuring 40-70µm x 20-35µm. In comparison to *Strongylus* eggs, their embryonated larvae is often U-shaped and the long

edges of the egg are parallel to each other. It is impossible to accurately differentiate the species of the egg under the microscope which is why either coprological cultures on Kogar-agar plate to recover adult worms or DNA analysis of the eggs are necessary and more sensitive.

Rhabditiform L1 larvae are 1,8 to 3,8µm long with a short buccal canal, a bulbed oesophagus taking up a third of the body length and a prominent genital primordium. L2 larvae grow longer and therefore have a smaller oesophagus/intestine ratio. Free-living adult males have a spicule and measure up to 0.75mm whereas free-living females may show a row of eggs inside and measure up to 1µm. Filariform L3 larvae measure up to 600µm with a notched tail and an oesophagus taking up half of the body length. In the intestines, female adult worms may measure from 3 to 8mm.

CLINICAL SIGNS

In humans, other apes and animals, acute *S. stercoralis* infections can produce a localised pruritic erythematous dermatitis at the site of skin penetration, advancing at approximately 10cm/h, tracheitis and coughing when larvae migrate through the trachea, and diarrhoea, with or without haemorrhage, constipation, enterocolitis, abdominal pain, peritonitis, anorexia and emaciation when larvae are in the intestinal tract. Clinical symptoms generally occur in young animals.

Chronic *S. stercoralis* infections rarely show such symptoms other than dermatologic lesions [48], although complications may occur, and most people have mild peripheral eosinophilia or elevated IgE levels. In case of compromised immune system, hyperinfection and disseminated strongyloidiasis (occurring in many organs) may develop in the host, with high mortality rates.

In primates, heavy infections with these parasites have been associated with mucosal inflammation, ulceration, iron deficiency anaemia, protein malnutrition, dysentery, weight loss, and death [88].

In humans, most *S. fülleborni* infections cases, even with heavy infestations, do not show clinical signs. However some infants infected with *S. fülleborni* subsp. *kellyi* may develop a highly fatal condition: a protein-losing enteropathy known as “Swollen belly syndrome” due to peritoneal ascites.

TREATMENT

Benzimidazoles (Albendazole, Mebendazole, Fenbendazole) and Levamisole are supposedly effective against intestinal infections [112]. Ivermectin is effective against adult worms. High level of hygiene is required to reduce larval development and multiplication of free-living generations.

- Spriruroidea*: Streptopharagus, Physaloptera* & *Protospira* species

Although these nematodes belonging to the Spirurida order and Spriruroidea superfamily have not frequently been reported in NHP, they have been mentioned in two studies concerning Guinea baboons. McGrew et al. found eggs of *Streptopharagus sp* and *Physaloptera sp.* Ebbert et al. diagnosed eggs of *Streptopharagus sp*., *Protospirura sp.* and a third unidentified genus, which could belong to *Physaloptera* genus when comparing with eggs found in other NHP faeces. However, in both studies, eggs were identified under light microscopy and the data given are extremely similar so the exactitude of the genus is questionable [31].

Streptopharagus genus are gastric nematodes of Old world monkeys & apes. *S. armatus*, *S. pigmentatus* [113] and *S. baylisi* [114] have been reported in baboons: in *Papio hamadryas*, *P. comatus* and *P. cynocephalus* .

Regarding *Physaloptera* genus, only *P.* (*Abbreviata*) *caucasica* had been reported in oesophagus, stomach and small intestine of baboons [115,116].
Protospirura. reports are often incomplete. Only *P. multipapillata* and *P. muricola* have been reported respectively in hamadryas baboons (*P. hamadryas*) [117] and in dog face baboons (*Papio doguera)* [90] in Tanzania and Kenya, as well as in Tanzanian Chimpanzees by Blumenbach.

LIFE CYCLE

Spirurida are a group of nematodes that use an IH (usually an arthropod) and live in the upper digestive system, primarily in the stomach and oesophagus.
Streptopharagus were first described in detail by Machida et al. in 1978, who supposed that eggs were shed in final host's faeces and ingested by coprophagous beetles which served as intermediate hosts for larvae to develop before being eaten by the primates and developing into adults. It was also mentioned that sexual maturation seemed long (adults worms were found 69 days after infection with infective larvae).
The intermediate host of *Protospirura muricola* in West Africa is the Madeira cockroach, *Leucophaea maderae*, in both its adult and nymph stages [89].
Physaloptera's indirect lifecycle is not completely known. Various dung and meal beetles, cockroaches, and crickets have been implicated as IH for some of the species in *Physaloptera* genus. A second IH or paratenic host may be needed [118].

MORPHOLOGY: EGG, WORM

Spirurid eggs are characterized morphologically as small, thick-shelled, rounded oval and colourless in most genera. *Streptopharagus* & *Protospirura* eggs contain a developed larva. Ebbert described the unidentified spirurid egg he found as with a greenish cast and containing an undifferentiated embryo.
Measurements of *Physaloptera* (=*Abbreviata*) egg described by Linstow (1902) and Meyers et al. (1971) were ranging from 29-52 x 46-70 µm and coincided with the unidentified Spirurid in Ebbert's study.

Streptopharagus eggs are described measuring 31-37 x 13-22 µm although reports by Jesse *et al.* (1970) and Myers *et al.* (1971) state larger sizes.
Protospirura eggs measure 40-43 x 53-75µm and have a thick hyaline coat surrounding the outer wall (Brumpt, 1931), which *Physaloptera* eggs don't have.

Spirurid adult worm sizes vary with the species, but with females always longer and larger than males. *Physaloptera* adults are 30-100mm long, *Protospirura* are 18-48 mm and *Streptopharagus* adults are 14-78mm long [113]. All three genera have a thick cuticle with transverse striations. They have a variety of cuticular ornamentations in the cephalic area, a cylindrical buccal vestibula, lateral chords that can be large, and often an eosinophilic fluid in the pseudocoelom [119]. Oesophagus is divided into short anterior muscular and long posterior glandular portion, which is surrounded by nerve-ring in *Protospirura* worms.
Sex can be distinguished by the genital primordium position and females have a vulva present in middle or posterior region.
Streptopharagus adults also have irregularly arranged spines at tail end and a cervical alae. They have a hexagonal mouth with 4 sub-median cephalic papillae whereas *Physaloptera* adult worms have 2 triangular lips.

PATHOGENESIS

Physaloptera infection can result in painful gastritis, esophagitis, enteritis, erosion & ulceration of mucosa where worms attach to [76]. Hyperplastic gastric lesions & gastric perforation have been described in cynomolgus monkeys [120].
Very little is known about the pathogenesis of Streptopharagiosis. The death of a baby chimpanzee due to perforated oesophagus secondary to migration of larvae was reported (Abee et al., 2021).

EPIDEMIOLOGY

Although these spirurids have a limited geographical range, exotic zoo animals also can become infested with parasitic nematodes for which cockroaches serve as possible intermediate hosts. For example *Protospirura bonnei* and *P. muricola* have been found in cockroaches collected in cages of monkeys. Nothing is known about public health significance of these nematodes.

DIAGNOSTIC

Physaloptera eggs are best diagnosed with formol-ether sedimentation method of faecal concentration. However, spirurid eggs are difficult to differentiate from eggs observed in faecal samples and culturing them into adult worms or observing adults attached to mucosa of upper GIT seem more reliable.

TREATMENT

Prevention and control of these infestation are sanitation & removal of possible arthropod IH.
No treatment is yet reported in case of *Streptopharagus*. In case of *Physaloptera* infection of NHP, the most efficient treatments are Thiabendazole, Levamisole hydrochloride, Carbon bisulfide and dichlorvos.

- Strongyles*: Necator americanus, Oesophagostomum & Trichostrongylus*

Hookworms are strongyles which belong to the Strongylida order, and include many genera of worms [121]. From wild Guinea baboon faeces, *Necator americanus, Oesophagostomum bifurcum* and unidentified species of *Trichostrongylus* have been reported.

N. americanus was also reported in other baboons [55] and wild African primates [10,66].

O. bifurcum has been reported in *Papio ursinus* [122], *P. doguera* [90], *P. anubis* [123,124]. Other species have been found such as *O.brumpti* in *P. sphinx, P. comatus, P. cynocephalus, P. hamadryas* as well as *O. zukowskyi* in *P. sphinx* [90].

Only few *Trichostrongylus* species have been identified in baboons, including *T. delicatus* in *P. hamadryas* and *T. falcultatus* in *P. ursinus* [122] and *P. ursinus* [54].

LIFE CYCLE

The life cycle of these hookworm genera is direct. Per day, thousands of eggs are shed into the baboons faeces and under favourable conditions (warmth, moisture, shade), they hatch within 1-2 days into rhabditiform larvae. These mature during 5 to 10 days into infective filariform L3 larvae which can survive 3-4 weeks in the optimal conditions.

In the case of *N. americanus*, they penetrate the new host upon skin contact (typically hands and feet), and via the bloodstream, reach the lungs. They migrate up to the pharynx, are swallowed and reach the small intestines where they mature into adults in 5-9 weeks and stay attached for up to a couple years, shedding up to 10,000 eggs/day [94,125,126]. Their lifecycle is shown in Annex 4.

In the case of *Oesophagostomum* and *Trichostrongylus*, the larvae are ingested with dirt, food or water by the baboons via faeco-oral transmission and reach the large intestine where they grow to adults [38], as shown in Annex 5 and **Error! Reference source not found.**. Baboons can become infected due to environmental contamination by both domestic and wild pigs [127], ruminants [69] and humans.

EGG MORPHOLOGY

Typical strongylid-type egg (STE) have smooth surfaces, elongated oval colourless shells, lateral sides which aren't parallel and are of variable sizes: 40-110 x 60-230µm. When shed into the faeces, they generally contain an embryo in the morula (cluster of 8-16 blastomeric cells) stage of development. *Necator* eggs are shed in an earlier stage of cleavage and contain a unicellular embryo which can't be differentiated from non-embryonated strongyle eggs. *Trichostrongylus* eggs may have one or both pointed ends [128]. Although one cannot differentiate the genus and species from observing the eggs, larval culture (or larval differentiation or worm typing) is possible, allowing the identification of the larvae released [129].

LARVAE & WORM MORPHOLOGY

Necator L1 usually not found in stool unless there is a delay in processing it, in that case must be differentiated from L1 of *S. stercoralis*. L3 filariform larvae are 500-700µm long, with prominent transverse cuticle striations, a round head, an apparent gap between oesophagus and intestine, and with a pointed ensheathed tail.

Oesophagostomum L3 larvae are longer (710-950µm) than hookworm L3, have a cephalic space and a pointed tail in a long thin, tapered sheath creating a gap between the end of the tail and end of sheath [130]. As a unique feature, intestines contains zigzag gut cells.

Adults of these three Strongylida genera of interest look very similar [125,131]. They have a transversely striated cuticle, are greyish/pinkish white when fresh (turning reddish brown later) with a small round buccal capsule containing 2 cutting plates (*N. americanus)* or a corona radiata (*Oesophagostomum).*Their adult size range between 0.2 to 3cm with *Trichostrongylus*, *Necator* and *Oesophagostomum* in order of average sizes. Males are always smaller than females and as a morphologic characteristic of this group, present a prominent bursa in the posterior end. Spicules can be used to differentiate the species; absent in *N. americanus* [125], various shapes in *Trichostrongylus*). Females have a short and pointed posterior end with vulva anterior to the middle of the body and a row eggs in the long uterus may been visible.

CLINICAL SIGNS

Oesophagostomiasis, Ancylostomiasis (disease caused by *N. americanus*) and Trichostrongyliasis are known in primates [12,132].

These intestinal Strongylida infections are generally asymptomatic but attachment to the intestinal walls can cause intestinal symptoms such as abdominal pain (in humans: similar to appendicitis), anorexia, dysentery, diarrhoea and nausea [125,133]. Due to adult hookworms feeding on blood, and especially in heavy infestations weight loss, fatigue, blood in the stool and pica may be seen along with severe iron-deficiency anaemia and protein malnutrition from chronic plasma protein loss [94]. In humans, this chronic iron-deficiency anaemia may lead to growth retardation and intellectual and cognitive deficiencies [126]. One could wonder if baboons also show modified behaviours. Bowel perforation causing purulent peritonitis has also been recorded. In the case of *Oesophagostomum* infections, intestinal & mesenteric nodules have been reported in olive baboons [132], without severe clinical signs [134] nor associated morbidity and mortality [135] which humans show [134,136,137].

Larvae can also cause dermatologic symptoms. Upon epidermis penetration, *N. americanus* larvae may cause pruritus (“ground itch”), cutaneous larval migrans or creeping eruption [125]. As the larvae migrate in the body, dermatitis, cutaneous nodules (*Oesophagostomum)* and sometimes eosinophilic pneumonia and coughing may be seen too.

EPIDEMIOLOGY

These STH infections are found in tropical and subtropical areas with warm, moist climates for larvae to survive in environment. Human cases are mainly reported from Sub-Sahara Africa, from poverty-stricken areas with poor sanitation [138].

N. americanus is present worldwide and described in New world monkeys, Old world monkeys and Apes, but also humans (considered one of the primary intestinal hookworm species), dogs, cats. *Oesophagostomum* and *Trichostrongylus* on the other hand are generally endemic and distributed where livestock is raised (ruminants but also swine) or where non-human primates live – humans are only incidental hosts for this genus. *O. bifurcum* (primarily found in monkeys) is the most common species of bursate nematode infecting humans in Africa, with highest incidence in Togo & Ghana were it cycles naturally in humans but as a different strain than baboons [139].

TREATMENT

Treatment goal for STH infections is to remove adult worms from the gastrointestinal tract using Benzimidazoles (BZA) in order of effectiveness: albendazole, pyrantel pamoate, mebendazole, levamisole [94]. Prevention measures include better sanitation, decreased use of night fertilizer (human faeces) and mass medical strategies.

- *Other species of interest*

Several other species of nematodes have been found in baboon faeces, in lower quantities and lower prevalence than the previous nematodes.

These include Oxyurid worms such as *Oxyuris armata* reported in hamadryas baboons [117], *Enterobius brevicauda* and *Enterobius vermicularis* reported in chacma baboons, and undetermined species found in olive baboons. Pinworm infection is one of the most prevalent diseases of children in the "western world". The female adult worm lives in the large intestine and when she is mature, she releases herself and all her sticky eggs at the level of the anus. Eggs are very resistant and will infect new hosts by oral route as well as airborne route.

Ascaris spp. have been observed previously in ape populations living in close proximity to humans [10,67] however, pathogenicity of this agent in wild apes remains to be determined. Confirmation of this pathogen, even at low prevalence (5.5%), in Fongoli chimpanzees is troubling and suggests risks of zoonotic transmission from overlapping humans and/or livestock. Ascariasis is the most common helminth-associated human disease worldwide. Infection can cause morbidity and death, by compromising nutritional status, affecting cognitive processes, inducing tissue reactions, such as granuloma, and provoking intestinal obstruction or rectal prolapse [88]. Like other STH it is treated with mebendazole, albendazole, pyrantel pamoate or levamisole.

3. Non-invasive methods to diagnose intestinal parasites of Baboons

Non-invasive diagnostic methods may provide information regarding the presence or absence of gastro-intestinal (GI) parasites, the prevalence and richness (community composition) of parasitic infections [56], without disturbing the natural behaviour of the individuals and population. On top of morphologic identification of parasites under the microscope or after copro-culture, molecular work can also be done to give more detailed and/or precise information about the species and prevalence.

3.1 - Basic sampling rules

In order to analyse samples, optimal conditions should be united when sampling [140,141].

- Using standard terminology all along the study will avoid mis-interpretation and maintain the value of the results,
- Avoiding collection of anonymous samples as the individual's data may be worthy, and as unintended duplicates may bias the results,
- Ensuring a correct sample size (n) for results to be significative such as:

  $n = \ln(a)/\ln(1-p)$ with "a": significance level accepted as 0.05 in free-ranging primates and "p": prevalence assumed at 5%
- To avoid contamination and pseudo-parasites: collecting samples straight after defecation, with new disposable spatula, from middle of dropping, wearing glove,
- Putting samples in cool, shaded place as soon as possible, unless direct smear is planned,
- If not testing directly, immerging the samples in the appropriate fixation medium as soon as possible.

It is also recommended to avoid using egg counts to measure infection intensity as number of eggs is generally not linearly proportionate to number of mature parasites in host. A single adult nematode may lay thousands of eggs, whereas hundreds of cestode adults of another species may lay very few eggs. Egg production can also vary over time (intermittent shedding), environmental conditions, faecal conditions, etc... Only qualitative measures can be reported.

3.2 - Preservation, fixation

Thermal preservation may be used by storing samples at 4°C for several days, or at -15°C for over a year [142].

Generally, chemical media are used to fixate parasites in faeces.

- Ethanol: Preserves DNA but destroys/alters morphology of parasites. Easy usage in field.
- Formol: Preserves helminthic eggs and larvae as well as protozoal cysts, oocysts and spores, but not trophozoites. Slightly alters parasite morphology. Usage requires special lab equipment.

- Merthiolate-Iodine-Formaldehyde (MIF): Easy usage in field is possible but also slightly alters parasite morphology and is toxic due to mercury.
- Sodium acetate-Acetic-Formaldehyde: doesn't contain toxic mercury but doesn't allow a precise visualisation of protozoa so trichrome coloration is recommended.

3.3 - Direct smear

A thin layer of faecal material is smeared on a slide with saline solution. Microscopic examination can reveal protozoa (sometimes still moving) and helminths but only when they are present in high concentrations. The observation and identification of parasites can be challenging due to the large quantity of faecal matter and debris present on the slide. Diluting the sample with water according to its consistency, homogenizing the suspension and smearing a couple of drops onto a slide may be useful to observe a larger quantity of faecal matter which is less dense. But more sensitive techniques eliminating the unwanted plant debris and concentrating the parasites are recommended.

3.4 - Flotation methods

In this most frequently used technique, the faeces are mixed with a solution of higher density than those of most parasite eggs and cysts, allowing them to float up toward the meniscus of the suspension whiles unwanted faecal debris sediments. The supernatant then contains the concentrated eggs, cysts and occasionally ingested ectoparasites.

The most efficient and frequently used solutions in veterinary diagnostics are $NaNO_3$, $ZnSO_4$ and $MgSO_4$. The latter two however are unsuitable to isolate many of the nematodes in wild primates. Saturated salt or saturated sugar solution are also used and more accessible but may be less optimal. The solution is chosen according to the parasites of interest and their density, each with advantages and disadvantages as shown in Table 2. There is no perfect method.

A portion of the faecal sample is taken and diluted in the chosen flotation solution. In the case of quantitative studies, the mass of faecal sample and the volume of flotation solution must be known and if using the Mac Master reading technique, dilution of faeces in solution should be 1:15.
The mixture is sieved in order to eliminate large debris and the collected suspension is homogenized using a pipette and utilized in various ways:

- After 10-15 minutes, eggs and cysts have floated up to the meniscus. A couple of drops are taken from the surface, deposited onto a slide and qualitative reading is done under light microscopy, at low and high magnification.
- In the case of quantitative reading, chambers of standardized Mac Master® slides are filled with the homogenized suspension before the 10-15-minute flotation period. The eggs are then counted and the number of eggs per gram of faeces (EPG) is calculated. This method requires a minimum of 50 EPG (1 egg per slide) as detection threshold and can only be microscoped at low magnification.

- To solve this issue, the Mini-Flotac® method was developed by Prof. Cringoli in Italy, allowing a limit of quantification (LOQ) as los as 5 EPG. On a circular plastic device, two 1-milliliter (mL) chambers are filled with the homogenized faecal suspension and after waiting 5 minutes, the top disc of the Mini-Flotac is twisted, smearing the top layer of both chambers onto a new surface. Microscopy can be done at high magnification and zigzag reading allows easy detection and quantification of eggs, oocysts, cysts and parasite larvae [143].

Table 2 – Most frequently used solutions for flotation methods and their characteristics [142].

Solution	Density	Ingredients	Advantages	Disadvantages
Saturated sugar (Sheather's solution)	1,12	1000mL water 680g sugar 10g phenol	Doesn't modify Nematode eggs Indicated for *Cryptosporidium*, *Strongyloidea*, *Ascaris*	Contamination by mould if phenol isn't added
Sodium chloride	1,15-1,20	1000mL water 400g salt (NaCl)	Easy & cheap to prepare Good for coccidian cyst flotation	Crystallization risk Temperature-dependent density Corrosive; modification of helminth eggs
Sodium nitrate	1,22	1000mL water 400g $NaNO_3$	Good for Nematode reading	Crystallization risk Modifies parasite shapes
Modified zinc sulfate	1,18	1000mL water 330g $ZnSO_4$ 150g zinc-acetate	Good for *Giardia*, *Strongyloidea*, *Ascaris* reading No pollution	High larvae stimulation perturbs reading Many debris also float
Magnesium sulfate	1,28	1000mL water 350g $MgSO_4$	Cheap Indicated for *Trichuris*, Strongyloidea, Ascaris Not many debris float	Crystallization risk
Potassium iodo-mercurate	1,44	399mL water 150g HgI_2 111g KI	All eggs float, including Trematodes	Polluting Corrosive; alters eggs' shape & flotation ability
Sodium nitrate and sugar	1,37	1000mL water 540g $NaNO_3$ 360g sugar	All eggs float except heavy Trematodes	None

Flotation techniques are easy, inexpensive and relatively sensitive methods allowing detection, counting, measurement and identification of many parasites but limited regarding species identification of helminth eggs and visualization of some protozoa. The highly osmotic solutions used alter the parasites viability which therefore cannot be isolated for coproculture [142].

3.5 - Sedimentation method

With this qualitative technique, parasites are separated from most faecal matter by sedimentation. Faeces are diluted in a 1:10 ratio with water (or other solution of similar density) so to give a very fluid suspension. The homogenized mixture is filtrated through a sieve, retaining the unwanted particles. Sediment of the resulting faecal solution can obtained by two methods:

- The solution is left to sediment naturally for up to 1 hour. The supernatant is discarded carefully without disturbing the sediment, from which few drops are then taken with an aspiration pipette. If the given result is too dense with debris once on the slide, the natural sedimentation can be repeated one or multiple times. This method is more time-consuming.
- The solution is centrifugated, the supernatant is discarded and some water is added to dilute the concentrated sediment. A few drops of the homogenized concentrated sediment are taken and placed on a slide for microscopy. This method is harder to read but can be used with very small quantities of faeces and allows better finding of Trematode eggs.

Flotation and sedimentation techniques can be combined to screen for all parasites using the Cauchemez method. Using a 15% salt solution, plant based debris float up first. The floating aggregate is discarded after 5 minutes, allowing light parasite eggs and cysts to float up to the surface and heavier parasites to sediment. After 30 minutes, the surface layer is used for microscopy (Annex 7, Annex 8), the rest of the supernatant is discarded and the sediment is also used with or without centrifugation.

3.6 - Baermann's larval extraction method

Baermann's method is used to recover larvae from fresh faecal samples, based on positive hydrotropism and thermotropism of larvae, resulting in efficient diagnosis of *Strongyloides* larvae [144] as well as hookworm larvae.

The material is installed as follows (Figure 10): a simple funnel is mounted on a metal support and connected via a rubber tube to a lower beaker. A filter or gauze is placed on a sieve in the funnel. Two versions are possible:

(1) either the rubber tube is clamped closed, the system is filled with water from up to the filter,
(2) or the tube is not clamped but submerged in warm water in the beaker.

The faecal sample is placed on the filter/gauze and left for several hours (from 2 to 24h depending on the studies), allowing larvae to sediment. Then either:

(1) as the clamp is slightly released, several drops are collected into a Petri dish or watch-glass
(2) or the content of the beaker is centrifugated at low speed, the supernatant is discarded and several drops of the sediment are placed on a watch glass.

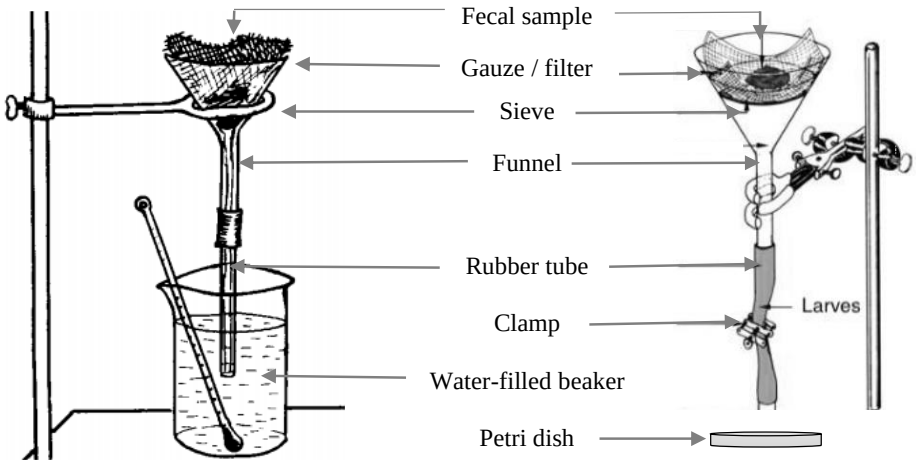


Figure 10 – Assembled apparatus for Baermann's method. *Left*: version (1). *Right*: version (2).

Observation is possible under stereomicroscope on a black surface or under normal light microscopy. Although it requires appropriate material and more time, Baermann's method is very useful to identify nematode genera and species as their identification is often not possible based on egg morphology.

3.7 - Copro-cultures

Several nematodes eggs have extremely similar morphologies, making it impossible to identify genera or species by the human eye. However, their infectious L3 larvae are differentiable. Faecal cultures from fresh samples allow eggs to hatch and larvae to develop. This technique is particularly interesting to diagnose strongyle, trichostrongyloid and rhabditid larvae.

Two methods are suitable for culturing:

- Faecal matter is mixed with vermiculite and water, placed in a cheesecloth and suspended in a closed container filled with water up to the cheesecloth level. Culture is incubated at 20° for 2 weeks. Then, larvae are collected by pipette and transferred onto a microscope slide. Adding iodine-based solution may help identification under the microscope. [56]
- Faecal matter is crushed and smeared in layers in a Petri dish which is placed in a closed incubator containing water-soaked cotton-balls. Incubation last 1-2 weeks at 20-26°C. Then Baermann's method is used to recover the larvae. [64]

Copro-culturing is time consuming and requires an experienced identifier as free-living nematodes from the environment may also be present, but allows precise identification of genus and species of larvae based on morphological traits such as size, oesophagus cell shape, oesophagus:body ratio, tail shape, outer sheath, etc…

3.8 - Colorations

In order to facilitate visualization of small parasites, especially protozoa, under light microscopy, special dyes can be added to the faecal solutions to colour specific component of the parasites.
When examining wild primate faecal samples, the most frequent staining methods used are with acid-fast (modified-) Ziehl-Neelsen [145], Iodine-based (Lugol) solution [64], Iron-haematoxylin, and Trichrome staining.

3.9 - Molecular diagnostic methods

Molecular methods have recently been more popular as the have become more accessible and result in precise diagnostics with great sensitivity and specificity. As they are more expensive, equipment-demanding and labour-demanding than microscopy diagnostics, they should be used in second intention.

From DNA fragments in faecal samples, a thermosensitive DNA polymerase and adequate primers, various polymerase chain reaction (PCR) based techniques can be used to amplify the genetic material present. This combined with multi-locus sequence typing (MLST) can be used to confirm the digestive pathogens present in the host as well as type the parasites .

Detection of parasite antigens (Ag) in the faeces is more and more frequently used for chronically carried pathogens, especially for *Cryptosporidium* and *Giardia* detection, generally using enzyme-linked immunosorbent assay (ELISA) methods.

Detection of circulating antibodies (Ab) in the faeces is also possible during or immediately after acute parasitic infections. Detection is done via several reaction types including humoral reaction induced agglutination (especially used for *Entamoeba histolytica* and *Fasciola* spp*)*, complement fixation resulting in no lysis of red blood cells (RBC) containing anti-RBC Ab, ELISA techniques, indirect immuno-fluorescence (IF) techniques or immuno-electrophoresis techniques.

Part 3. Goals of this study

The goal of this study was to explore gastro-intestinal parasitism in a natural population of Guinea baboons which is already involved in longitudinal studies.

Recovering parasites by non-invasive means can provide data on presence or absence of certain parasites in the population, as well as prevalence and richness of infections at a specific time point. The obtained data can be used to compare individuals within a same population but also to compare wild and captive populations, sympatric species, or different baboon populations elsewhere.

Throughout a study comparing gorillas with different degrees of habituation, Pafco et al. (2017) showed that habituation does not increase the risk of infection of wild gorilla groups. It can be assumed that this is also valid for baboons. With minimal equipment needed, non-invasive sampling of habituated groups can easily be done in the field and is repeatable over time. Therefore this survey can serve as baseline data for future studies.

Collecting samples from identified individuals is necessary to reduce sampling bias and therefore bias of all further analysis. Identification allows the use of the data in particular for behavioural studies. As opposed to other baboons, Guinea baboons groups are very fluid, individuals easily change groups and partners and many social behaviours are observed. Relationships between parasitism dynamics and behavioural patterns may then be studied.

Changes in patterns of faecal parasites may reflect differences of parasitic transmission, host dietary preferences and habitat utilization (Stuart et al., 1998). Describing the intestinal parasitic repertoire of a same group of individuals over time can therefore help understand the role of parasites in primate evolution, ecology and behaviour.

On top of that, anthropogenic effects maybe be observed and zoonotic risks can be assessed.

Part 4. Study site & subjects

The focus of our study was a group of habituated free-ranging Guinea baboons inhabiting the Park National du Niokolo Koba (PNNK) of Senegal. These are the only habituated groups of wild Guinea baboons in Africa.

1. Research area

1.1 - Niokolo Koba National Park, Senegal

Senegal is located on the most western edge of Africa, along the Atlantic ocean. It is bounded by Mauritania, Mali, Guinea and Guinea-Bissau and encircles Gambia. The PNNK is located in South East Senegal (Figure 11Figure 11), at approximately 1,5 hours car journey South from Tambacounda. Its geographic coordinates are 12°30'-13°20'N, 12°20'- 13°35'W. The Gambia river, originating from north Guinea runs through Senegal and Gambia into the ocean, gives rise to the large Niokolo Koba branch which flows through the PNNK. Classified as UNESCO World Heritage Site since 2007, it is one of the most protected areas in West Africa as a great diversity of Sub-Sahara African flora and fauna is concentrated in over 9000 km² (representing 4% of Senegal).

Figure 11 – Location of the NKKP including Simenti (our study site) and both research sites of previous studies on GI parasites in Guinea baboons.

Fauna diversity

Over 80 species of mammals cohabitate in the parc including *Kobus* antelopes, roan antelopes, Lord Derby elands, buffalos, leopards, lions, hyenas, African wild dogs, warthogs, green monkeys, red colobus baboons, chimpanzees, hippopotami, some elephants, etc... 330 species of birds, 36 species of reptile, 60 species of fish and around 20 species of amphibians are also found in the parc.

The dynamics of the complex PNNK ecosystem have been described by several researches [146–150], most detailed in Livre blanc: Le Parc national du Niokolo Koba by Benoit [151]. The ecology of Guinea baboons is currently being described by D. Zinner.

Human presence

Although no permanent human residents other than the military rangers, the staff of a luxury lodge, a depleted hotel and several camping grounds and some scientists, a large tarmac road crosses the north east section of the PNNK and tracks web the rest. Tourists were quite rare although a new luxury lodge was about to open at the time of sample collection. No livestock is legally kept on the PNNK grounds but there is pastoral pressure. Although reducing, poaching is still present. Problematics and countering strategies have been addressed since 30 years [151].

Environmental conditions

The majority of PNNK is Sudanese savannah (north part) & Guinea savanna (south part), composed of deciduous woodland and gallery-forming forest, habitat to 1500 plant species including baobabs, African locust bean trees, African mahogany trees and various palm-trees as well as grass types like Vetiver, Paspalum and bamboo fields. The park is relatively flat, the highest peak being Mont Assirik at around 300m above sea-level. Lower rocky hills are separated by large dry plains and valleys which become lush grasslands and wetland marshes in the rainy seasons before being burned to the ground creating large open spaces [43]. The variety of habitat in our area of study is represented in

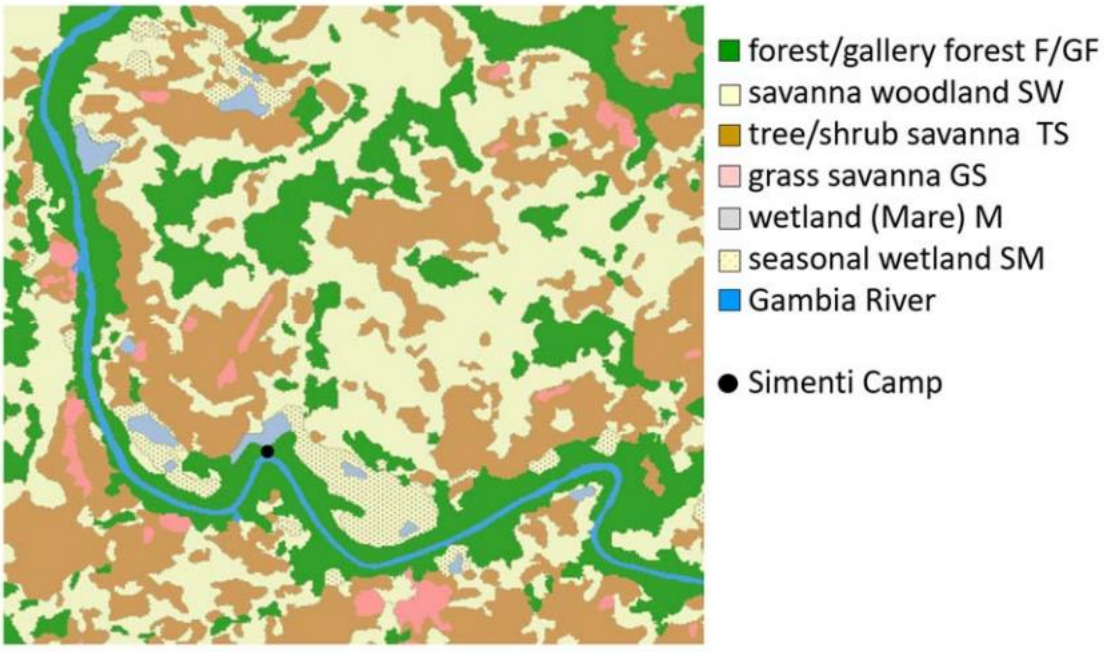


Figure 13.

1.2 - Simenti: the research site & its climate

The Deutsches Primatenzentrum based their field station called “Centre de Recherche de Primatologie (CRP)” at Simenti, located North west of PNNK Figure 12,

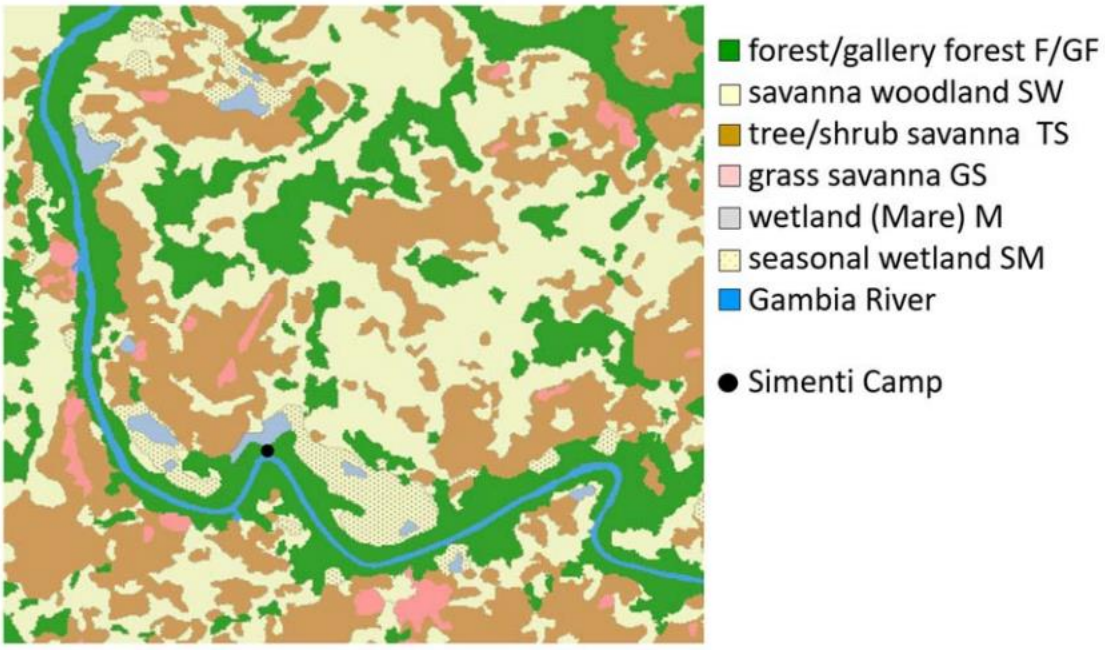


Figure 13). Rainy season from June to October and a dry season for the rest of the year. The average annual rainfall is 8,000-1,100 mm [33].

Alongside three other seasonal wetlands in the area, the Mare de Simenti is the only permanent waterhole in the parc, therefore attracts many animals. Sympatric monkeys are present such as West African green monkeys, patas monkeys, Temminck’s red colobus and Senegal bushbabies. Many many grazing herbivores come to the Mare to drink, as well as warthogs and potential carnivore predators [33]. The river banks are bordered by strips of gallery forest, and all around CRP, the same

diversity of savannah, of habitats as present in the PNNK is observed and represented in

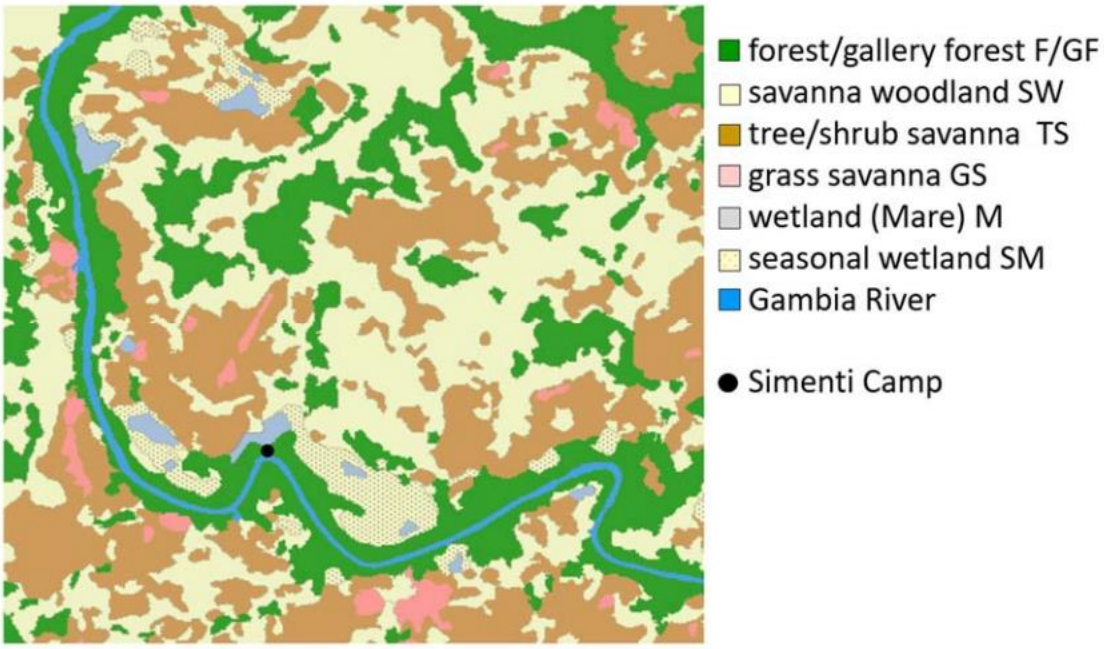


Figure 13. A high proportion of the habitat around CRP is composed by savannah woodland and tree and shrub savannah.

The microclimate varies seasonally as shown in Annex 9. For example in 2019, the year previous to this study, temperatures were as low as 10 degrees Celsius (°C) at night during "winter" and as high as 42°C during the day in dry season. Relative humidity ranged from 10 per cent (%) in dry season to 99% during wet season. Rainy season is from June to October and a dry season for the rest of the year.

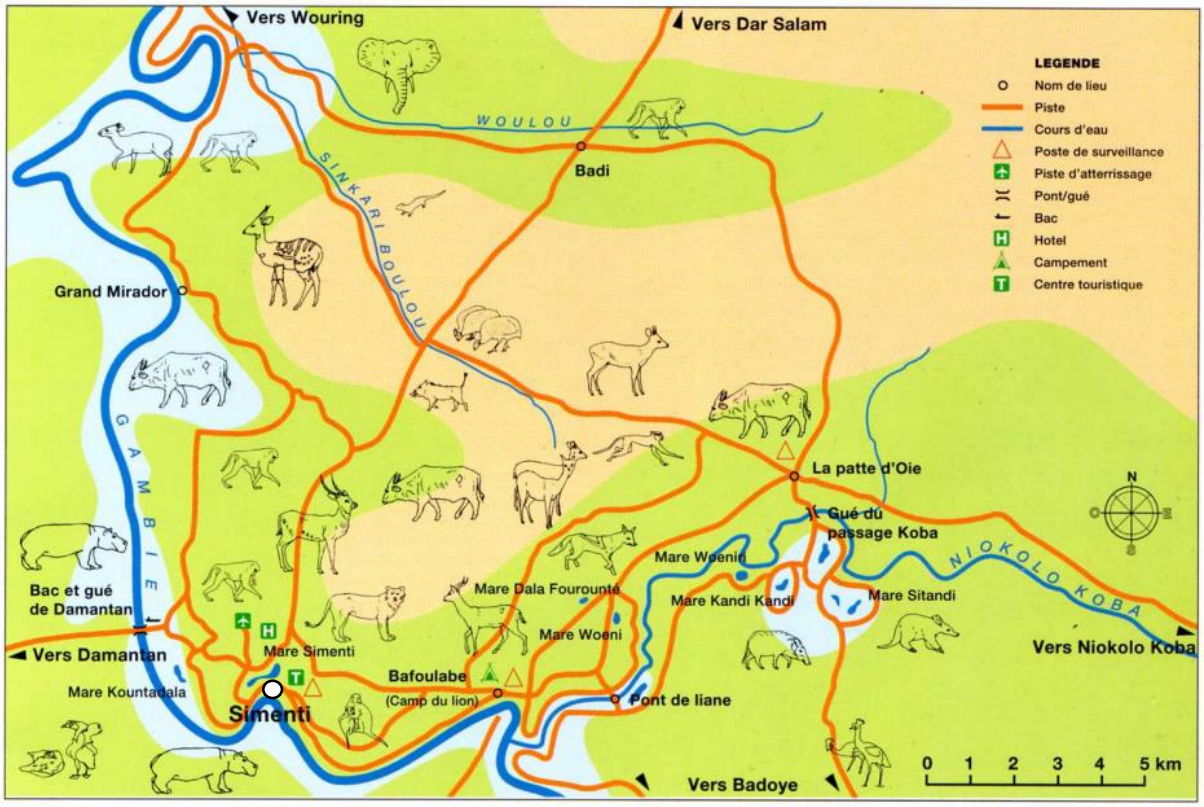


Figure 12 – Map of Simenti area of the PNNK.
Source: Guide à l'usage des visiteurs du complexe écologique du Niokolo-Badiar.

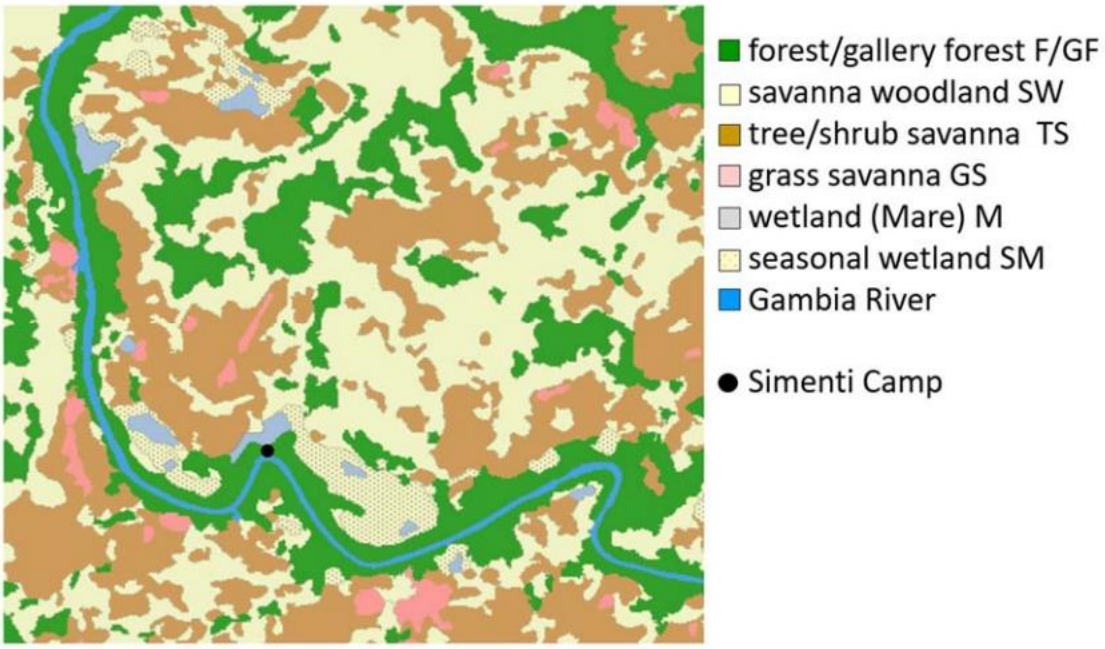


Figure 13 – Distribution of habitat classes around CRP Simenti**.**
*Source***:** *Zinner et al (2021). Comparative ecology of Guinea baboons (Papio papio). subm. to Primate Biology.*

Authorisation

Permission to conduct research in the area was granted by the Direction des Parcs Nationaux and Ministère de l´Environnement et de la Protection de la Nature de la République du Sénégal, as well as by the Conservator of the park. This research complied with the animal welfare regulations. This research was conducted in compliance with the ASP Principles for the Ethical Treatment of Non-Human Primates.

2. Study group & subjects

2.1 - Demography

We surveyed the faecal parasites from Guinean baboons mainly from 3 parties belonging to 2 gangs, which are part of longitudinal behavioural and ecological studies and are therefore identified and habituated. These parties have been followed extensively on average every second day since 2007 by teams from the Deutsches Primatenzentrum (DPZ) now lead by J. Fischer. The parties named 5, 6, 9 numbered 34, 36, 24 individuals respectively at the time of the study and lived around the CRP Simenti camp. Party 6 has divided into 2 new parties: 6L and 6W. Similarly a unit detached from Party 9B creating their own party 9B. Additional parties/gangs range in the study site for which the population density has been estimated at 7.5-10 baboons/km² [34].

All individuals of these parties have been identified as they enter the party, and are recognizable by morphological traits such as size, coat colour, tail shape, pigmented or depigmented skin patches, ischial callosities, ear notches and other particularities.
They are assigned a name with a corresponding 3-letter code and an age category on the basis of morphological and sexual characters. The demography of parties of interest is summarised in Annex 10.

2.2 - Tracking

Tracking collars had been installed on some male baboons, which seemed quite central in the parties of interest. This allows the scientific team accompanied by a local ranger to locate the baboons (generally surrounded by the rest of the troop) rapidly each morning. Baboons become active before dawn, start by urinating and defecating prior to descending from the sleeping sites and either leave quickly or may stay on the grounds for up to a couple of hours resting, grooming, sun-bathing, playing, before travelling and foraging (*pers.obs*). Data from previous years showed that these Guinea baboons have an average home range of 24.8 km² in which they travel over an average of 4km per day. During the time of study [34]. During this study in December, January, February, the daily travel distances were shorter, which corresponds with the seasonal fluctuation observed by Zinner et al. in previous years.

The scientists track the baboon parties daily from 7am to 1pm in order to take data individual behavioural data, as well as collect faecal samples for hormonal and microbiome studies. The team

must be close enough to observe the baboons precisely, whiles keeping a “respectful” distance ensuring that the baboons are not disturbed.

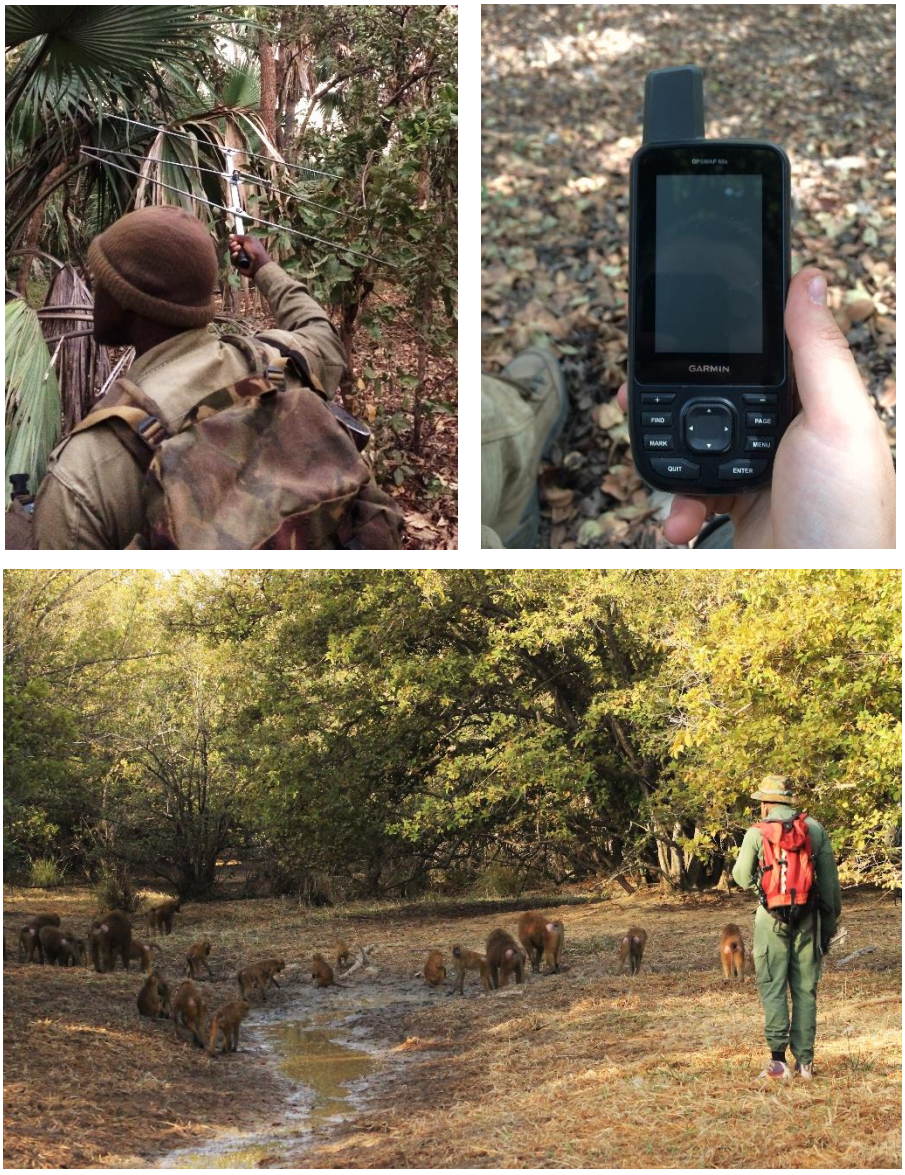


Figure 14 - *Top:* Material used to track baboons include radio telemetry antennas and GPS devices. *Bottom*: Respectful distance is kept from the habituated troop. A ranger is always present. *(Photos: A. Coles)*

Part 5. Field work: materials & methods

1. Faecal samples

1.1 - Sample collection in the field

Between December 21rst, 2019 and February 8th, 2020, 159 samples were collected from 95 individuals belonging mostly to three parties of habituated baboons followed daily. Annex 10 provides information on the demographic composition of the parties.
Faeces were collected only if seen being produced by an identified individual, immediately after defecation and as soon as it was safe enough. Using fresh disposable wooden spatulas, portions of the top, the end, the sides and the middle of the faeces were scooped up (Figure 15), avoiding soil contaminations, so to fill approximately two thirds of a 15mL collection tube, previously weighed. The samples were transported in the shade during 1 to 5 hours back to the camp and then stored in a fridge (temperature range: 4-8°C) for 2 to 26 hours, before being processed.

It was attempted to collect ≥ 1 sample from as many individuals as possible, as well as 3 to 6 samples from the same individual in a 10 day period, to be used as controls. In total, 159 samples were collected from 95 individuals (32 from *Party 5*, 28 from *Party 6L+6W*, 28 from *Party 9+9B* and 7 from other groups). These individuals represent a variety of ages categories with similar sex proportions and represent nearly equally the 3 parties of interest (Table 4 a,b).

Table 3 a - Age and sex of individuals sampled.

Age	Female	Male	Total
Adult	18	23	41
Juvenile	14	12	26
Old	4	5	9
Subadult	10	4	14
Yearling	2	3	5
Total	**48**	**47**	**95**

b - Party and sex of individuals sampled.

Group	*Female*	*Male*	*Total*
Party 5	19	13	**32**
Party 6	14	14	**28**
Party 9	13	15	**28**
Party 10		1	**1**
Party 13	1		**1**
Party 14		2	**2**
Party 9B	1	1	**2**
Other		1	**1**
Total	**48**	**47**	**95**

Figure 15 - Collection of baboon feacal samples *(Photos: A.Coles)*

1.2 - Storage, ID & shipping

Approximately 5g of each sample was taken for processing in the camp. The remaining portion in the collecting tube was weighed. To this tube, 5mL of Ethanol (80%) was added for storage reasons, and the tube was weighed again.
Each shipping tube was labelled with the code of the identified baboon the faeces originated from, the number of sample for that individual if ≥2. the party they belonged to at the time of collection, the date and time of collection as well as the weight of the empty tube (with cap and label), as shown here-under.

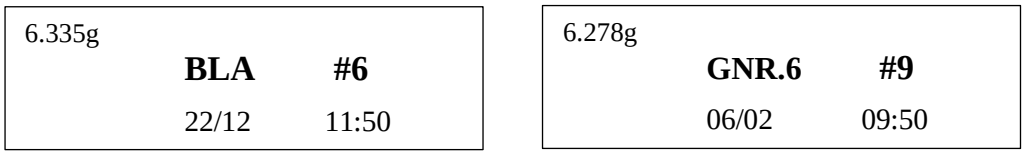


Figure 16 - *Left*: Label identifying the first sample collected from BELA (BLA) in Party 6, the 22/12 at 11:50. *Right:* Example of a label identifying the sixth sample collected from GINGER (GNR) in Party 9, the 06/02 at 9:50.

The sample collection and shipping was agreed to by the local authorities.
Double packaging in screw cap tubes grouped in sealed plastic bags separated by cotton pads, as well as the proper documentation ensured correct transport of patient specimens, following the WHO guidance and FAO requirements for legal export of specimens of animal origin.

2. Coprological study

2.1 - In the field: Flotation method

On site, half of the collected samples were observed under the microscope to have an overview of the method and anticipate the results. A flotation method seemed appropriate.
Modified Sheather's sucrose solution was made by mixing salt, sugar and water (SSW) to a 1:2:3 ratio, kept in the fridge and renewed every second day.
Between 2 and 26 hours after being placed in the fridge, approximately 5g of each sample was extracted, placed into a plastic cup and 15 to 30 mL of SSW solution was added, depending on the quantity and density of the extracted faeces. This was well mixed using a disposable wooden spatula and strained through a sieve, retaining the soil and other unwanted particles but allowing parasite eggs through. This liquid was poured into 12 mL tubes and centrifugated for 5 minutes at XXX rpm (Hettich EBA 200 centrifuge).
Several drops from the meniscus of each tube were placed on a glass slide and observed under a light microscope (SWIFT SW350T, 40x-2000x). Measurements were made to the nearest 0.5 µm using a micrometre integrated into the digital camera (Swiftcam SC1803-CK). Parasite and pseudo-parasites were photographed.

2.2 - In the lab: Flotation, Concentrated sedimentation & Natural sedimentation methods

Due to the Coronavirus epidemic situation, postal services and laboratory work were modified and further analysis was delayed. In September 2020, the samples were sent to the Department of Parasitology of the University of Veterinary Medicine of Brno, Czech Republic. Under the supervision of Prof. Dr. D. Modry and in collaboration with a team of parasitologists, 77 of the samples were totally or partially analysed via different methods. Each of these samples belonged to a different individual baboon. Parasite presence and identification was determined on the basis of egg or cyst shape, size, structure and content [152,153].

Flotation method

Approximately 1g of faecal sample was mortared with water and then sieved in order to collect the filtered liquid into a test tube. After centrifugation at 2000 rpm for 3 minutes, the supernatant was discarded. Saturated salt solution was added to the sediment filling the tube up to 10mL, and gently mixed. 1mL of the mixture was deposited in each chamber of a mini-FLOTAC disc. After 5 minutes the disc was twisted and looked at under the microscope.

Sedimentation methods

Approximately half of each faecal sample was put into a mortar and gently mixed with water using a pillar. The mixture was sieved, retaining big debris, into a test tube. After centrifugation (5 mins, 2000 rpm) the supernatant was discarded and the sediment was weighed (W). The sample was then separated into two, to analyse it both under natural sedimentation as well as concentrated sedimentation methods, as follows:

If $W \leq 0.5g$, 1-2 drops were taken with a pipette for further concentrated sedimentation analysis (placed directly on a glass slide and observed under the microscope) and to the rest, water was added and mixture was left for 40 minutes for natural sedimentation.

If $W > 0.5g$, water was added to each h 3mL and X mL was taken and placed into a new tube in order to have 0.5g of the original sediment, so with $X=1.5/W$, for concentrated sedimentation analysis. To the rest, water was added to the top and left to sit for 40 minutes for natural sedimentation.

In both cases, once the 40 minutes elapsed, the supernatant was discarded, water was added, and let to sit for 10 minutes, and this twice.

Figure 17 - Material needed to process samples for natural and concentrated sedimentation methods *(Photos: A.Coles)*

A semi-quantitative evaluation of the number of parasites within each type found in an individual host was done using a cross-scoring method:

- = negative
1 = only 1 parasite is found in the whole sample analysed
+ = 2-10 parasites are found in the whole sample
++ = 1 parasite is found in every 5-10 field of view
+++ = 1 parasite is found in every field of view
++++ = many parasites are found in every field of view

3. Molecular study

3.1 - DNA isolation

Additionally to the microscopy work, it was decided to search for Strongyloides DNA in our samples using PCR and gene sequencing techniques.
A total of 55 samples were chosen strategically to well represent the population on age and sex categories; originating from 6 old, 19 adults, 10 subadults, 15 juveniles, 5 yearlings, resulting in a total of 30 females and 25 males. Between 0.3g and 0.7g of each sample was taken and transferred to an Eppendorf tube.

The DNA of each faecal sample was extracted using the DNeasy PowerSoil Kit (Qiagen, Hilden, Germany) made especially for isolating genomic DNA from environmental samples using patented Inhibitor Removal Technology® (IRT). Samples were heated overnight in a thermobox at 37°C to evaporate the ethanol they had been stored in. The extraction of DNA was performed according to the instructions of the manufacturer.

The dried faecal samples were added to the PowerBead tubes containing small beads in a buffer, which disperse unwanted particles, partly dissolve humic acids and protect nucleic acids from degradation. To lyse cells, 60 µL of C1 solution (containing SDS and other disruption agents) was added and briefly vortexed. Then all tubes were vortexed horizontally at maximum speed for 30 minutes, before being centrifuged at 10,000 x g for 30 seconds. The supernatant of each sample was transferred to a clean 2 mL collection tube.
To remove contaminating organic and inorganic matter, 250 µL of C2 solution (containing patented IRT and reagents) was added and samples were vortexed for 5 seconds before an 30 minute incubation at 4°C. Tubes were centrifugated again for 1 minute at 10,000 x g.
During that time, clean 2mL tubes were filled up with 200 µL of C3 solution (also containing the IRT and other reagents), briefly vortexed and incubated at 4° for 60 minutes. Avoiding the pellet of debris formed, 600 µL of each supernatant was transferred to respectively to the C3 containing tubes. They were then centrifugated for 1 minute at 10,000 x g.
Avoiding the new pellet, 750 µL of supernatant was transferred into clean 2mL tubes before adding 1200 µL of C4 solution (high-concentration salt solution) into each sample to bind the DNA only and being vortexed for 5 seconds.

To capture all the DNA, the samples were loaded onto an MB Spin Column containing a silica membrane and centrifugated at 10, 000 x g for 1 minute and flow through was discarded. This was repeated until the whole sample had been processed.
To wash the captured DNA, 500 µL of C5 solution (ethanol) was added and samples were centrifuged for 30 seconds at 10000 x g, the flow through was discarded and samples were centrifugated again for 1 minute. The MB Spin Columns containing the captured DNA were placed into clean collection tubes avoiding any splashing.
To elute the DNA from the membrane and salt, 50 mL of C6 solution (sterile elution buffer: 10 mM Tris-HCl, pH 8.5) were added in the centre of the silica filter membrane of the Spin Columns and samples were incubated at 4° for 10 minutes and centrifugated for 30 seconds. Another 50 mL of C6 were added; centrifugation and incubation were repeated. The MB Spin Columns were then discarded. The extracted and eluted DNA samples were labelled and stored in a freezer at -20°C.

3.2 - Checking DNA concentration of isolated samples

The first batch of samples was used to evaluate the isolation method in case modification had to be made. DNA concentrations of 2 µL of each of these isolated samples was measured using spectrophotometry. The C6 solution used to elute the DNA previously was used as a blank.

3.3 - Amplifying DNA

The detection of *S. stercoralis* and *S. fülleborni* was performed by using two primers designed for a partial mitochondrial cytochrome-c oxidase subunit 1 coding region (~217 bp), SPP Cox1 F (5'-TTTGATCCTAGTTCTGGTGGTAATCC-'3) and SPP Cox1 R (5'-GTAGCAGCAGTAAAATAAGCACG-AGA-'3) respectively [154]. The amplification was done using polymerase from the PPP Master Mix (Top Bio, Vestec, Czech Republic). The primers, the polymerase and water buffer were assembled for the total number of wells prepared, according to Table 1. Master Mix alone was used as a positive control and water alone was used as a negative control.

Components of the PCR mixture	Quantity needed per well (µL)	Quantity needed for a strip of 16 wells (µL)
H2O	8	140,8
Top Bio PPP polymerase	12,5	220
SPP Cox 1 F primers	1,25	22
SPP Cox 2 R primers	1,25	22
Isolated DNA sample	2	/

Table 1. Material used to create the optimal conditions to amplify the isolated *S. stercoralis* and *S. fülleborni* DNA. The mixture was made for 16 wells, vortexed and centrifugated. Then 23 µL of this mixture were put with a pipette into each tube of the strip and 2 µL of the isolated DNA were added. The product was then vortexed and centrifugated too.

PCR was performed using the Biometra T-personal Thermocycler (Schoeller), according to the conditions indicated in Table 2.

Step	Time	Temperature
1	2 mins	98°C
2	10 s	98°C
3	10 s	63°C
4	40 s	72°C
5	4 mins	72°C
6	Until cool	8°C

Table 2. Conditions used for PCR. Steps 2, 3, 4 were repeated 44 times for a total of 45 cycles. *Protocol was optimised by Eva Jiroušová*.

3.4 - Gel electrophoresis, purification & sequencing

Electrophoresis was used to separate the PCR products and confirm DNA amplification. For that, a 1,5% agarose gel was prepared by mixing 1.2g of Agarose with 80mL TBE PCR water in a beaker. The beaker was microwaved for 4 minutes and cooled under the tap whiles mixing. 80µL of Midori Green (diluted 1:20 in water) was added to stain the DNA/RNA. The solution was poured into an electrophoresis tray and let to set for 15 minutes.
10 µL of each DNA sample was put into each well of the gel, except the last one in which 5 µL of Marker was put. Electrophoresis was run during 40 minutes. The separated fragments were then visualized and photographed by UV transilluminator (Vilber Lourmat, Quantum).
The products were purified using the Gel/PCR DNA Fragments Extraction Kit (Geneaid).
Bidirectional Sanger sequencing reaction and capillary electrophoresis was carried out by a commercial provider by Macrogen (Korea). The obtained sequences were compared to similar available nucleotide sequences in the GenBank database (www.ncbi.nlm.nih.gov/genbank/) using BLAST (www.blast.ncbi.nlm.nih.gov/Blast.cgi).

3.5 - Phylogenetic analysis

The positive samples for *Strongyloides* of Guinea baboons (*P. papio*) were compared by phylogenetic analysis. The alignment was completed by Geneious 9.1.5 [155]. The final Geneious alignment consisted of the 9 obtained sequences and 27 sequences from the Genbank, selected from reliable and published sources. The length of the sequences was adjusted to 719 bp, the size of the shortest sequence available. Another nematode specie; *Necator americanus* (AJ417719) was chosen as an out-group. Nodal support was assessed to 10^5 replicates. The dataset was tested by multiple approaches (Maximum-likelihood, Neighbour-joining, Bayesian analyses) with the GTR+G+I substitution model (General Time Reversion model + rate of variation across sites (Gamma) + Invariable sites).

Figure 18 - Material needed for DNA isolation and PCR *(Photos: A.Coles)*

Part 6. Results

1. Coprological work results

1.1 - Identified parasites

From the 44 faecal samples observed in the field using a flotation method and the 54 faecal samples examined in the lab using sedimentation (54) and flotation (17) methods, 7 parasite types were found (Figure 19). Two types of protozoa were detected: *Entamoeba* sp., and *Balantioides/Buxtonella*-like ciliates. From the five types of helminth eggs found, only three were identified to genus: *Strongyloides* sp., *Trichuris* sp. and *Enterobius* sp., whereas the two other types could not be further identified only based on morphological observation under the microscope: spirurid eggs and strongylid-type eggs. An unidentified nematode larva was also found. No trematodes nor cestodes were observed. From all samples, only one free-living rotifer larvae was observed, indicating relatively clean collection.

Entemoeba spp. (Figure 19j) were detected as spherical thick-shelled cysts with a 10-20 µm diameter and containing many nuclei (4 to 8).

Balantioides-like cysts (Figure 19d) were also observed as brown spherical structures with a large transparent outer envelope.

Unembryonated *Trichuris* sp. eggs (Figure 19a,b) were identified as brown/yellow barrel-shaped, with transparent bipolar plugs, measuring 50-60µm x 25-30µm.

Strongyloides eggs (Figure 19i) were oval, symmetrical, with thin colourless shells, measuring 50-70µm x 30-40µm and containing U-shaped embryonated larvae.

Oxyurid eggs (Figure 19d) were thick-walled asymmetrical ellipsoids with one side straighter than the other. Although identification of the species requires adult worms, the only genus present in primates is *Enterobius.*

We observed rounded oval thick-shelled eggs containing coiled-up larvae corresponding to spirurid eggs (Figure 19 c,e,g). The smaller ones measured on average 35x17 µm which could be consistent with *Streptopharagus* sp. [23,91] but also *Subulura* sp. [156] (Figure 19 f)& found in the previous reports on primate parasites. The larger eggs measured on average 50x35µm. Those with a thick shell surrounded by a hyaline cuticle, keeping faecal matter away from the ovule membrane, seem consistent with *Protospirura* sp. eggs found previously, whereas those without the hyalin cuticle could be *Physaloptera* sp. [23].

The un-embryonated and blastomeric thin-walled strongylid-type eggs (STE) (Figure 19 k,l,o) could not be differentiated based on their simple morphological characteristics. The elongated oval colourless-shelled eggs had smooth surfaces and were of variable sizes: 40-45 x 55-70µm These could be hookworms like *Necator americanus, Oesphagostomum* sp. or *Trichostrongylus* sp.

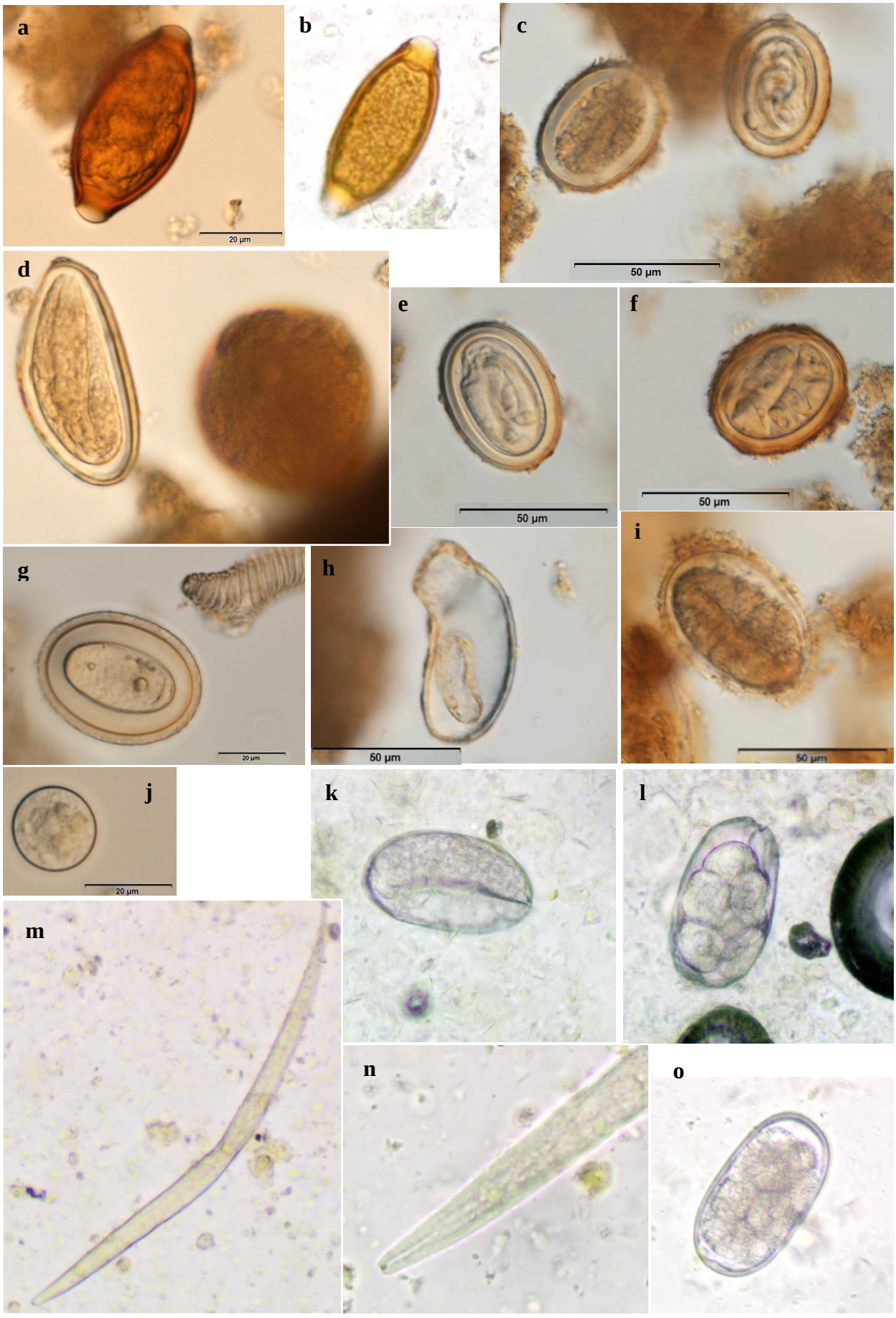


Figure 19 – a, b: *Trichuris* sp. (egg). **c, e, g**: Spirurid nematode (egg). **d**: *Enterobius* sp. (egg) and *Balantioides*-like sp. (cyst). **f**: *Subulura* sp. (egg). **h**: ruptured nematode egg shell. **i**: *Strongyloides* sp. (egg). **j**: *Entamoeba* sp. (cyst). **k, l, o**: Strongylid nematode (egg). **m, n**: nematode (larvae). Observed under light microscopy.

2.2 - Prevalence, semi-quantitative evaluation & richness

As less samples were observed in the field than observed in the lab, only results from the laboratory analysis are used for further calculations.

Prevalence (Table 4Table 4) was measured by calculated the portion of hosts found to be infected by a type of parasite. Each sample belonged to a different individual baboon. The most prevalent parasites were both protozoa; *Balantioides*-like sp. and *Entamoeba* spp. Regarding nematodes; spirurid eggs were the most frequently observed and strongylid-type eggs were also relatively present. Prevalence of *Trichuris* sp., *Strongyloides* spp. and *Enterobius* sp. was fairly low.

Table 4 – Prevalence of parasites found by coprological examination. N = 54 samples.

Parasites	Positive samples (n)	Prevalence (n/N) in %
***Balantioides*-like sp.**	46	85.2
***Entamoeba* spp.**	38	70.4
Spirurid eggs	13	24.1
Strongylid eggs	7	13.0
***Trichuris* sp.**	5	9.3
***Strongyloides* spp.**	4	7.4
***Enterobius* sp.**	1	1.9

The semi-quantitative evaluation showed a variety of intensities of excretion of protozoa cysts observed by sedimentation techniques (Table 5). On the other hand, when *Trichuris* sp. was found, only very few eggs were counted per slide. Most samples containing spirurid eggs also had a low excretion intensity.

Table 5 – Number (n) of samples observed after sedimentation techniques containing parasites according to semi-quantitative categories.

Amount / Parasites	1 in whole slide (n)	+ (n)	++ (n)	+++ (n)	++++ (n)
***Balantioides*-like sp.**		18	12	15	1
***Entamoeba* spp.**		17	12	9	
Spirurid eggs	3	7	1	2	
***Trichuris* sp.**	2	3			

Richness was measured as the number of parasite types (not genus nor taxa) observed in each sample (therefore in each individual) using flotation and sedimentation techniques, as shown in Table 6. Infectious rate was 92.6 %. The majority of the individuals seemed to harbour one, two or three types of parasites. Only one individual was observed as extremely rich in parasites with 6 types found.

Table 6 – Richness of parasites found by coprological examination. N = 54 individuals.

Parasite type(s) present	Number of individuals	Percentage of individuals
0	4	7.4
1	10	18.5
2	18	33.3
3	21	38.9
6	1	1.9

2.3 - *Balantioides*-like ciliates

Whiles observing the faecal samples processed with flotation method, natural and concentrated sedimentation methods, a very high large number of unusual cysts were observed in many samples. These looked very similar to those formed by *Balantioides* sp., also ciliated but larger and encapsulated. Photos were taken to document these ciliates and measurements were made of the outer-shell (average: 59 x 53µm) and inner-shell (average: 47 x 45µm) from 30 of these yet undescribed cysts (Table 7).

Table 7 - Measurements of outer- and inner-shells of 30 cysts formed by *Balantioides*-like ciliates, in "YSS" sample. G: shell was gone, PR: shell was partially ruptured.

	Outer-shell (µm)		Inner-shell (µm)	
Cyst	Length	Width	L	W
1	70	50	42	42
2	60	53	50	50
3	G	G	55	55
4	63	58	50	50
5	68	63	50	50
6	45	40	37	32
7	63	60	50	50
8	62	62	50	50
9	50, PR	45	45	40
10	G	G	58	55
11	60	55	50	46
12	38	37	30	26
13	55	52	40	42
14	G	G	45	45
15	PR	PR	40	40
16	65	55	52	50
17	G	G	58	53
18	G	G	55	53
19	60	53	45	45
20	PR	PR	53	53
21	65	60	45	45
22	65	60	48	47
23	G	G	50	50
24	G	G	48	47
25	G	G	45	37
26	70	60	48	45
27	G	G	45	45
28	G	G	40	40
29	47	47	47	35
30	48	40	35	30
average size	**59.06**	**52.78**	**46.87**	**44.93**

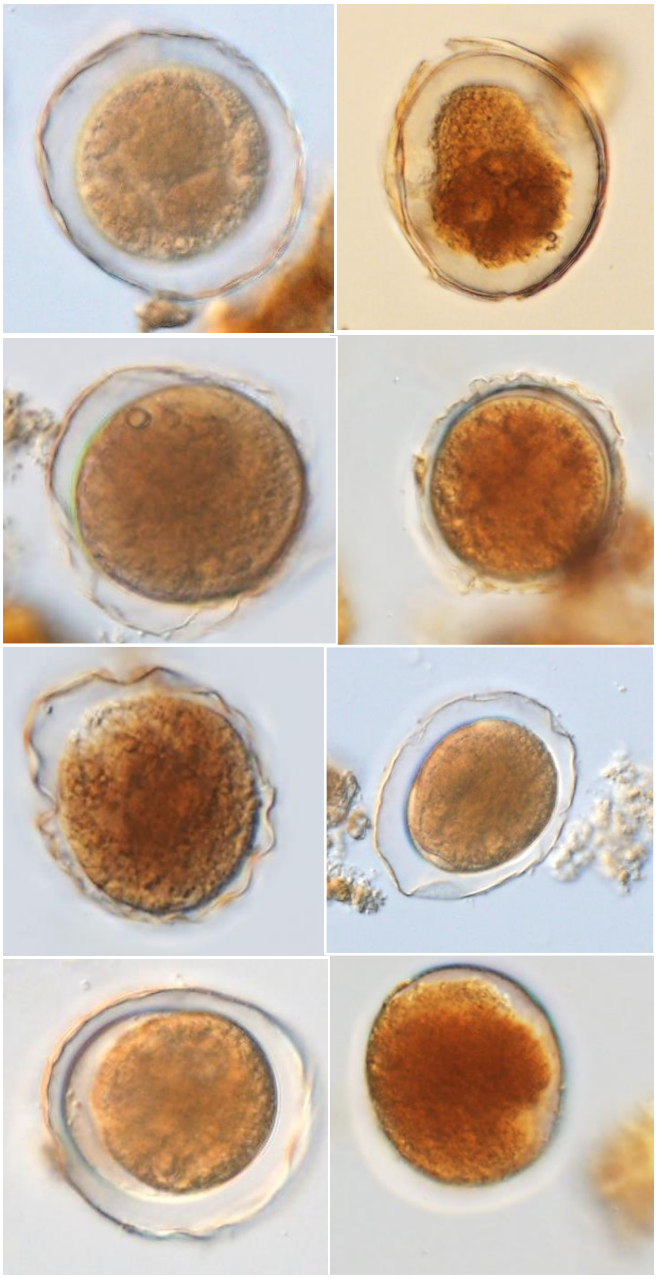
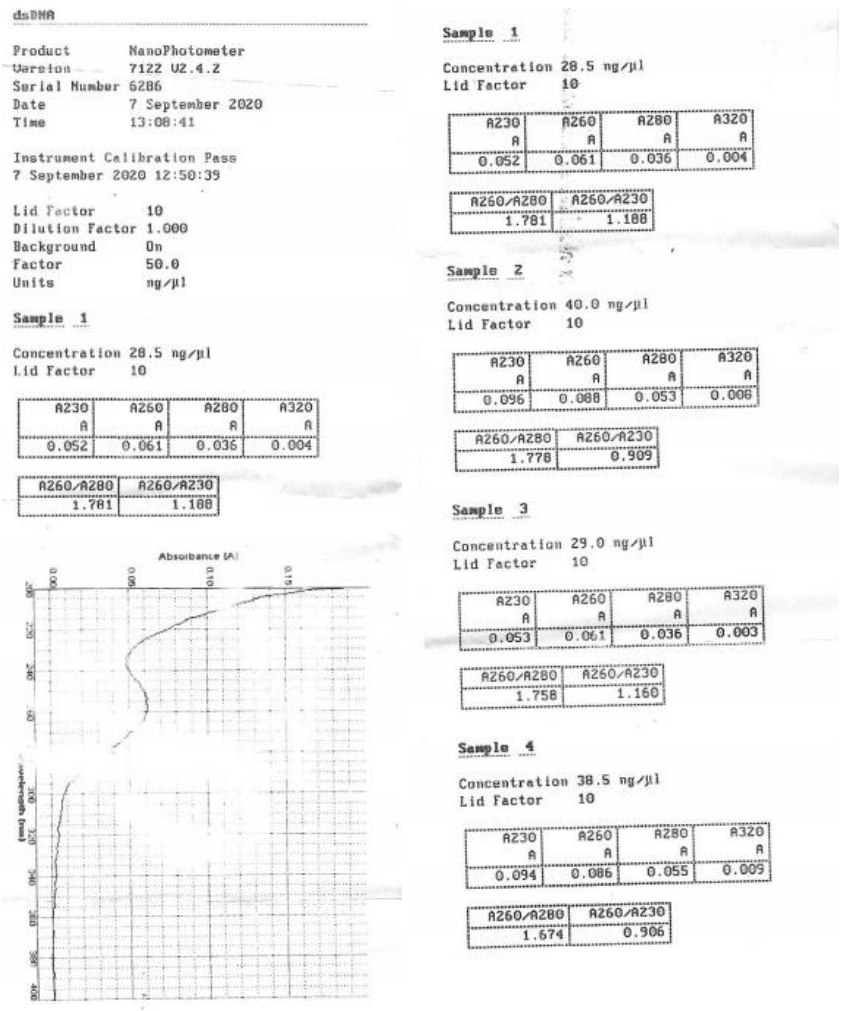


Figure 20 - Examples of *Balantioides*-like ciliates from "LLT, SLY, TQL" samples

3. Molecular work results

3.1 - Spectrophotometry evaluation of DNA isolation

Each of the 24 first samples yielded DNA. DNA concentrations of the samples ranged from 10 to 48 ng/µL, showing a correct isolation method of the first batch. No modification had to be made to the isolation protocol.



dsDNA

Product NanoPhotometer
Version 7122 V2.4.2
Serial Number 6286
Date 7 September 2020
Time 13:08:41

Instrument Calibration Pass
7 September 2020 12:50:39

Lid Factor 10
Dilution Factor 1.000
Background On
Factor 50.0
Units ng/µl

Sample 1

Concentration 28.5 ng/µl
Lid Factor 10

A230 A	A260 A	A280 A	A320 A
0.052	0.061	0.036	0.004

A260/A280	A260/A230
1.781	1.188

Absorbance [A]

Sample 1

Concentration 28.5 ng/µl
Lid Factor 10

A230 A	A260 A	A280 A	A320 A
0.052	0.061	0.036	0.004

A260/A280	A260/A230
1.781	1.188

Sample 2

Concentration 40.0 ng/µl
Lid Factor 10

A230 A	A260 A	A280 A	A320 A
0.096	0.088	0.053	0.006

A260/A280	A260/A230
1.778	0.909

Sample 3

Concentration 29.0 ng/µl
Lid Factor 10

A230 A	A260 A	A280 A	A320 A
0.053	0.061	0.036	0.003

A260/A280	A260/A230
1.758	1.160

Sample 4

Concentration 38.5 ng/µl
Lid Factor 10

A230 A	A260 A	A280 A	A320 A
0.094	0.086	0.055	0.009

A260/A280	A260/A230
1.674	0.906

Figure 21 – Results of spectrophotometry indicating DNA concentration for first samples. A concentration above 10-15 ng/µL is acceptable for further analysis.

3.2 - Electrophoresis analysis of DNA amplification

Photos of the electrophoresis gel under UV-light allowed the observation of PCR results, as shown in Figure 22. Out of the 54 isolated DNA samples having gone through PCR, results were negative for 9 samples (ABU, DAR, DNT, CHP, MCM, MCS, NRI, YGR, UZR). The 45 other positive samples were purified and sent for sequencing.

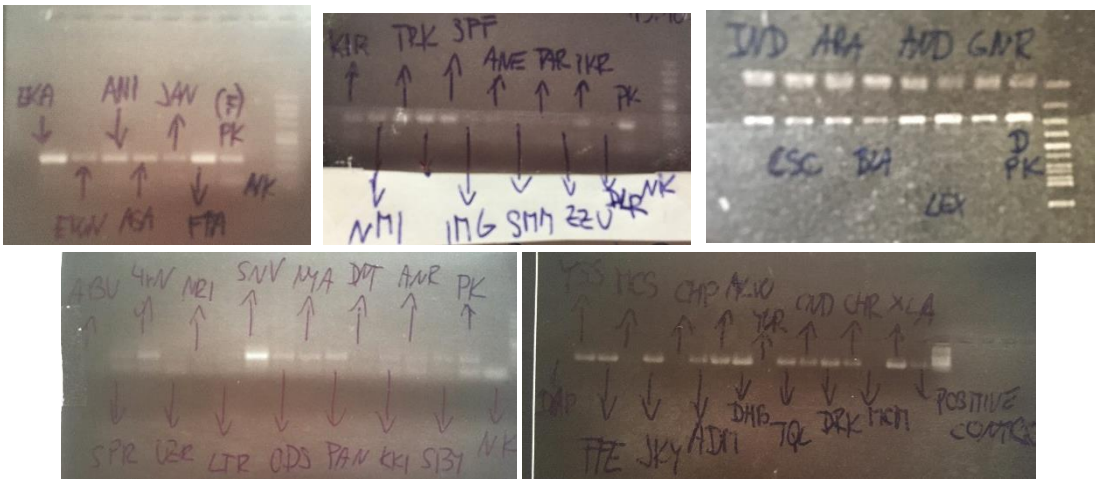


Figure 22 – Photographies of electrophoresis gel. Each lane contains the PCR sample from one individual, marked by it's code. "NK" and "PK" are negative and positive controls.

3.3 - Sequencing results

This molecular work enables the differentiation of closely related species co-circulating within the same host population. To determine Strongyloides and Strongyle species present in the samples, 46 amplicons (both forward and reverse) were sent for Sanger sequencing.

Strongyloides stercoralis was identified in 4 samples (ARA, BLA, CSC, IND) and *Strongyloides fülleborni* was identified in 5 samples (DRK, CHR, SBY, PAN, EWN).

In other samples, Strongyle nematodes (hookworms) were amplified; *Necator americanus* was amplified in 19 samples (LEX, ADM, JKY, EKA, ANI, ASA, FFE, ODS, NYA, ANR, SNV, TAR, KEN, TRK, ANE, KIR, SFF, SMM, IKR) and *Oesophagostomum* sp. were amplified in 4 samples (AND, CND, JAN, SPR).

Multiple peak of sequences were evident in the chromatograms of 13 samples indicating mixed infections occurred, probably by Strongyle nematodes or *Strongyloides sp.* in these 13 individuals (YSS, XLA, ALW, TQL, DMB, HVN, KKI, LTR, IMG, NMI, DLR, ZZU, FTA). Sequencing was unsuccessful for 1 individual sample (GNR).

Table 8 – Prevalence of parasites detected by PCR.
As 1 sample was unsuccessfully sequenced, N = 45.

Sequencing result	n	% (= n/N)
Strongyloides stercoralis	4	8.89
Strongyloides fülleborni	5	11.11
Necator americanus	19	42.22
***Oesophagostomum* sp.**	4	8.89
Mixed infection	13	28.89

3.4 - Phylogenetic analysis

The resulting phylogenetic tree based on a part of Cox1sequence analysis is shown in Figure 10. All of the supposed *Strongyloides* isolates were placed into highly supported clades.

The 5 samples containing *S. fülleborni* sequences created a separate branch in the clade of "African *S. fülleborni"*. None of them were placed in the "Asian *S. fülleborni"* clade containing species found in orangutans, lutungs, proboscis monkeys and macaques, all originating from Asia.

The 4 samples containing almost identical *S. stercoralis* sequences were placed in a branch with samples from humans of different countries (LC179524: Myanmar; KU962139: Laos; KY081226: Thailand). It is highly supported that this *S. stercoralis* shares a common ancestor with strains found in humans and dogs (LC 179497).

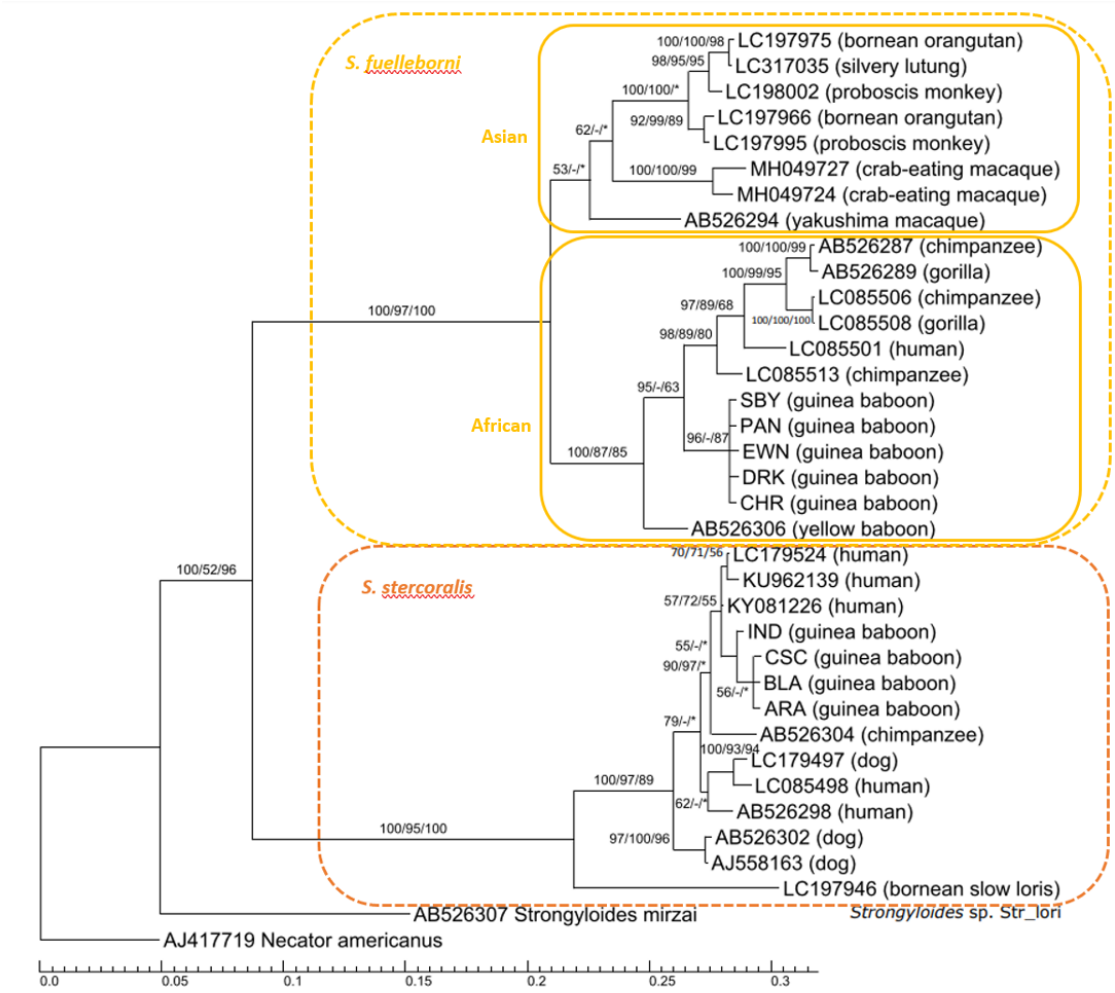


Figure 23 - The phylogenetic tree of *Strongyloides* sp. as inferred from a part of Cox1. The phylogeny was constructed from Geneious alignment combining 3 methods: Maximum-likelihood, Neighbor-joining, Bayesian analyses, and using a GTR+G+I model. Numbers on branches show support from 10^5 replicates, with each of these 3 methods. The scale expresses the expected number of substitutions per site. Samples from this study are marked using the 3-letter code name of the baboon host.

Part 7. Discussion

Although parasites in primates and in the baboon genus have been studied, wild Guinea baboons are relatively rarely included.
This study was initiated to deepen the parasitological knowledge of Guinea baboons in their natural habitat. As they have revealed to be quite intriguing subjects in primatology due to the mystery of their social organisation, it also seemed interesting to see if there may be correlations between parasitism and their social connectedness.
However the series of hosts for the present study is not sufficiently inclusive and the diagnostic methods are not sufficiently optimised to allow detailed deductions relative to parasite-socialness of host associations.
Nevertheless, understanding the parasites' life cycles, transmission routes, host specificity and pathogenicity is essential to understand the impact it has on its host [53], influencing its ecology and fitness. By adding parasitological data about Guinea baboons, this research is useful for further use.

1. About the methods: comments, errors & improvements

Sensitivity of methods:

- The sedimentation method (SM) was more successful than the Sheather's-solution flotation method (FM). For example *Balantioides* sp. was detected in 89% of samples observed with SM but only in 13% of samples observed with FM. Similarly *Entamoeba* spp. were detected in 70% of SM samples but only 7% of FM samples. Other parasite were also more frequently found using SM. However, flotation method gave us positive results for *Trichuris* sp. in 2 individuals (KIT, TQL) which were found negative with sedimentation methods. SM was therefore retained.
- Although concentrated sediment is much quicker to obtain than natural sediment, the density of debris and overall characteristics of baboon faeces made it harder to observe parasites. On the other hand concentrated sediments allow the observation of a larger quantity of faeces and a high chance of containing parasites. Combining both SMs gives more accurate results but focusing on natural SM seemed more efficient in this study.
- As some parasite eggs or oocysts may not be passed continuously throughout the day, collecting samples for a same individual over several days increases sensitivity of diagnostics. This was done for some individuals but these samples have not yet been analysed.

Optimisation of conservation:

- *A posteriori* the tubes should have been shaken vigorously to ensure maximum contact between the faeces and the storage solution [56].
- A portion of each faeces should have been stored in formalin straight away, to ensure conservation of helminth eggs and larvae, allowing better microscope observations.
- A portion of each faeces could have been fixed in poly-vinyl acetate (PVA) in order perform more sensitive tests for protozoa identification.

To better identify parasites:

- A drop of Lugol's iodine could have added to help the identification of protozoan cysts under light microscopy
- Formol-ether concentration technique could have been used to isolate parasite eggs and protozoan cysts [62].
- To recover *Strongyloides* larvae developing shortly after defecation, the Baermann technique could have been used directly in the field.
- To recover adult worms in the field, the faecal debris could have been washed on strainers with aperture sizes of 5 mm, 0.5 mm, and 0.1 mm and the residues could then be transferred to a Petri dish and observed under a stereomicroscope. The adult worms would be collected, fixed in formalin, cleared in glycerol-alcohol solution by evaporation, and observed under light microscope with Nomarski contrast [156].
- To identify *Strongyloides* spp., a Koga agar plate culture [157], could have been realised before direct microscopy and formalin-ethyl acetate microscopy.
- As it has been observed, molecular diagnostic methods are much more sensitive. With more funding and time, this could be used to screen of all the other parasites.

To optimise the molecular analysis:

- Next generation sequencing could avoid the uninterpretable dual peaks on the chromatogram which are generated by Sanger sequencing in case of mixed infection [154].
- As Cox1 is present in multiple helminth species including members of *Strongyloides* genus and various strongyles, simultaneous dominant and submissive chromatograms can also be reduced by using more specific primers, but their design is time-consuming.
- To support of the phylogenetic hypothesis of *S. fülleborni* subspecies, rigorous analysis would be needed based on different part of genes, rather than only Cox1. In helminths several genes are useable: *Cox1* (good for variability studies, and enough sequences are recorded in Genbank), *ITS* (used in *Strongyloides* too) and *18S* (codes for proteosynthesis by ribosomes, quite stable throughout evolution, used for general information such as genera differentiation). More specifically for nematodes, the mitochondrial *ND4* and *CytB* genes are used for their great variability across genera and species.

Assessment of severity of parasite burden in individuals:

- Different studies have shown that although faecal egg count (FEC) may correlate with the number of sexually mature helminths (infection level), it does not correlated with the worm burden and therefore with the impact of parasites on host. Therefore FEC was not judged useful in this study.
- Faecal antibodies IgA titers could be measured to asses gut mucosal immunity and therefore predict the baboons' health and fitness [158].
- It would also have been interesting to proceed to opportunistic necropsies of baboons in the field.

Comparing seasonal variations:

- Another set of samples could have been collected from the same individuals six months later during the opposite season. Fisher's exact texts and Mann-Whitney U tests could then be used to compare parasite prevalence and richness between dry and wet seasons [66], potentially due to environmental conditions or presence of intermediate hosts.
- Therefore, in our opinion, the results of this study possibly do not represent the entire richness that is present in the population.

Looking for correlations between parasites and hosts:

- The use of multivariate logistic regression modelling would be appropriate to observe correlations between the parasites found and the individuals tested, with variables such as : sex, age category, party, recent change of party, recent change of unit, unit size, mother, aggressive behaviour, grooming time, playing time, etc…

As one can conclude, many methods are available and possible, but some techniques are good for detecting a certain parasite but not for conservation of another, some are good for clinical work and not for general surveys (for example Zinc-sulphate flotation method is recommended if Giardiosis is suspected but many debris also float, increasing reading difficulties, Formal-ether concentration technique works for carnivorous and animal-eating hosts, but not frugivorous/folivorous, etc…). Using standardised protocols to monitor parasites would enable proper comparison of studies over time, groups, species and location, and allow clear understanding of differences [13].

2. Comparing results with previous studies & interpretations

2.1 - *Entamoeba* spp.

Entamoeba cysts were observed in the majority of individuals. Species found may be *E. hartmanni, E.histolytica, E. dispar,* or *E. chattoni.* Although previous studies named the pathogenic *E. histolytica* and the non-pathogenic *E. dispar* and *E. coli,* microscopic differentiation of these species based on cyst morphology is not reliable, and molecular analyses are needed to determine species [70,159] which could also be *E. hartmanni* or *E. chattoni*.

2.2 - *Trichuris* eggs

Although previous studies commonly show a high prevalence of *Trichuris* eggs (ranging from 28 to 92%) in faeces collected from wild baboons, only a very low prevalence was found in our samples.
Although low *Trichuris* prevalence had been observed in Guinea baboons before, it was only in captive baboons, thus leading to the assumption that captivity could alter their grooming behaviour, reducing the transmission of parasites via faeco-oral route [69]. Such a low prevalence was therefore unexpected in these highly social wild Guinea baboons.
This striking difference could be due to the number of samples examined, a larger daily travel distance and larger home-range (and therefore lower host density), or what is suspected as most probable, a

wrong interpretation of microscope viewing. Indeed, when comparing the descriptions in previous studies [23] and looking at our pseudo-parasites, we suspect that there may have been a confusion with fungi or yeast spores in previous studies.

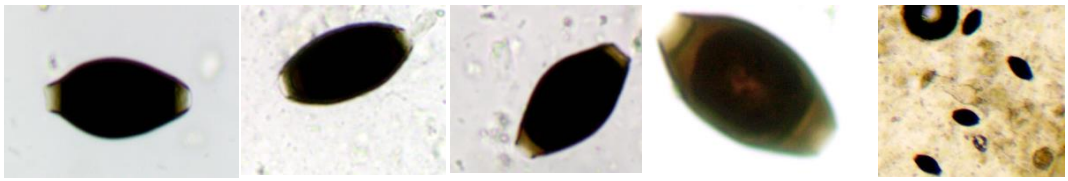


Figure 24 - Pseudoparasites found in many samples when observing fresh faeces under the microscope with flotation method.

2.3 - Spiruroidea

The identification of *Protospirura, Abbreviata* and *Physaloptera* genera can only be confirmed using molecular tools.

However, a much lower overall prevalence of Spriruroidea was observed in the Simenti baboons (24%) in comparison to Ebbert's study who found up to 70% of samples infected with *Protospirura* sp. in the Lion Valley. The exact life-cycle of these stomach nematodes is unknown, but all known spirurids have an indirect life-cycle that involves an arthropod an intermediate host [88,160]. It seems therefore highly possible that the variation in prevalence across one same species of baboon is linked to the presence of an intermediate arthropod, potentially impacted by the environmental conditions. These same conditions may also directly impact parasite survival and transmission. Sampling faeces at different seasons, comparing temperature, humidity, baboon densities, swamp presences, etc… as well as microclimate parameters would allow to study this.

2.4 - Strongylida

More strongylid eggs were found than in Ebbert's study who found 4% and 6% in LV and SV respectively but less thanMcGrew and Howells who found 38% and 29% respectively. However the latter affirmation is doubtful as many *Necator* eggs are undistinguishable by microscope examination.

When analysing the coprological results, Strongylida eggs were mainly found in male hosts (85.7% of infected individuals). This seemed congruent with a previous hypothesis that *Necator* sp. prefer male hosts. However the explanation offered was that in tropical climate areas where heavy rains and warm temperatures are characteristic, mostly men are involved in agricultural labour [161,162], which is not applicable to baboons. Additionally, when analysing the molecular results, it turned out that host distribution was nearly equal between male and female.

Under the microscope 13% of samples showed Strongylida eggs whereas PCR detected over 42% (23 out of 54 samples) infected either with *N. americanus* or *Oesophagostomum* sp..

Both these results show the limits of coprological examination and interpretation.

Following another suggestion made by Mul et al. (2007) explaining that due to more frequent physical contact female baboons and their off-springs harbour hookworms which transmitted via faeco-oral,

percutaneous or trans-mammary pathways more frequently than males. However, on top of nearly equal proportions of *N. americanus* per sex, our study PCR reveals *Oesophagostomum* infection in two males and two females. Could this be due to the higher frequency of social interactions male Guinea baboons have?
Another observation to be mentioned is that none of the yearlings, juveniles and subadults tested presented *Oesophagostomum* sp., which was seen only in adult and old individuals. Therefore one could suppose that age might play a role in *Oesophagostomum* infection.

Additionally, the *Oesophagostomum* infection prevalence found in Simenti baboons during dry season is relatively low (< 9%). It would be interesting to check if this prevalence increases with rainfall as well as if baboons ingest certain plants accordingly, suggesting similar self-medication as that seen in chimpanzees [134,135].

This molecular study did not reveal any *Trichostrongylus* with certainty, which is surprising as a high prevalence of *Trichostrongylus* was expected in these wild baboons since they feed in the same environment as many herbivores which may shed infectious *Trichostrongylus* larvae in their faeces [65,69,163].

2.5 - *Enterobius* sp.

The only pinworms to known to infect primate to date are *Enterobius* sp. and *Colobenterobius* sp. [164]. Mostly *E. vermicularis* has been reported in baboons, although the distinction with other species which have sporadically been reported in baboons, is not morphologically reliable. In contrast to Ebbert's study twenty years ago which found high prevalence (37-42%) of *Enterobius* sp. at Mount Assririk, only 1 individual was observed harbouring *Enterobius* in the Simenti baboons sampled. However, this is not so surprising as pinworm eggs are rarely shed in the faeces and more often found around the anus of the host.

2.6 - *Strongyloides* spp.

From the samples observed under the microscope, all individuals infected by *Strongyloides* were male and adult. At first sight, this could be congruent with the social male-male interactions observed in Guinea baboons as well as homosexual mounting as an alternative route to soil transmission of infectious *Strongyloides* larvae [54]. However, the molecular work reveals many more infected individuals either just with one *Strongyloides* species (17%) or with mixed infections (29%), as the microscopic work (7,4%). So interpretation based on microscopic techniques is questionable. On top of that, molecular diagnostic tools showed nearly equal portions of male and female individuals infected by one *Strongyloides* species. Therefore, the male-male anogenital interactions cannot be supported in this study, without contradicting the hypothesis either.
As another infection route is trans-mammary transmission which, although not known for *S. stercoralis*, has been reported for *S. fülleborni* in humans [54]. It would therefore be interesting to collect samples from lactating mothers and their infants, to check for a correlation of infection.

From our samples, no individuals infected with one *Strongyloides* species belonged to Party 9. However, some individuals carrying a mixed infection may also carry *Strongyloides*, so the previous observation is not interpretable.

2.7 - *Balantidium-like* ciliates

A very high number of ciliates were found, resembling *Balantioides* cysts but larger. Recently, studies conducted on primate faeces have been reporting ciliates which look like *Balantioides* cysts yet different. However very little information is available regarding intestinal parasitic and commensal ciliates, and all older studies categorise findings as "*Balantidium coli*" simply by microscopical diagnoses, which is unreliable.

In other primates similar *Balantioides*-like cyst-forming ciliates have been proven genetically close to *Buxtonella* sp, a parasite found in pigs & cattle [165]. The cysts found in these Guinea baboons had outer and inner envelope-like structure, of which the diameter was measured. Number of vacuoles and macronucleus size was not easily measurable. Somatic ridges were not visible, in contrast to other *Balantioides*-like findings which sounded similar to ours.

On top of morphological description of stained trophozoites and scanning electron microscopy, it would be interesting to apply molecular techniques to potentially identify theses mysterious cysts which - to our knowledge – belong to a yet undescribed taxon.

2.8 - Absent parasites

No trematodes such as *Schistosoma* nor *Watsonius* species were found in our samples in contrast to the previous studies on wild Guinea baboons. This may be explained that both these trematodes use snails as intermediate host [88] and that our samples were collecting during dry season, when no snails were seen and hot dry climate is not optimal for trematode survival in the environment. Also, according to McGrew (1989), some Guinea baboons have shown behavioural adaptations to avoiding infection by water-borne *S. mansoni* cercariae infection for example by drinking in shorter bouts from nonflowing rather than flowing water sources. This may also be the case here. One must also not forget that trematode eggs excretion can be irregular, so more samples per individual should be examined.
Another explanation could be that generally these baboons have very little human proximity, reducing their exposure to contaminated water [166]. Further studies comparing parasite presence and behavioural data in this field would be beneficial.

Additionally, no *Ascaris* eggs have been found. This could be due to the fact that adult worms liberate many eggs as exiting the host's anus, resulting in eggs sticking to the skin around the anus and not in the faeces. In captivity, diagnose is done by using a tape band. However in Ebbert's study collecting Guinea baboon faeces in the same national park, *Ascaris* eggs were reported. Misdiagnose is also a possibility.

3. Extrinsic factors which could influence parasite infection

3.1 - Spatial parameters

Several spatial parameters may influence parasite occurrence, prevalence, and richness. First of all, it would be interesting to study the home range of these Guinea baboons as well as daily, weekly or monthly movement of small groups. Individuals covering a larger surface on a regular basis will more likely come in direct (greeting, fighting) or indirect (through faeces) contact with a greater number of baboons, but also other animal species and diversity of habitats. This would predispose them to harbour more parasites, increasing intensity and richness of infection [167]. However, larger home-ranges may also involve spreading out of small units, lowering the infection risk with STH.
The second parameter which could be studied is the amount of time individuals/units spend on the ground and above the ground translating their individual degree or territoriality or arboreality. These have been reported to influence parasitic infections in wild animal populations [168]. Spending more time off the ground may be linked with avoidance of contaminated pathways [169,170].

3.2 - Climactic parameters

To date, studies linking seasonality with parasitic infections are diverging.
Most researchers have found a positive correlation between wet season and helminth richness [167]. Indeed, wet and warm tropical conditions favour sexual reproduction of free-living adult *Strongyloides* generation and thereby provide a relatively high number of infective larvae for host invasion. [170]. They also seem to favour *Necator* transmission rates [162], heavier worm burdens of *Bertiella* and *Oesophagostomum* [122] and higher prevalence of spirurids [171]. The higher parasitic counts has been explained by the desiccation which most nematode eggs and larvae undergo during dry season, reducing viability and therefore parasitic transmission by over 200 [16]. Another explanation offered is the reduced availability of intermediate hosts consumed by the primates during dry season [171]. A fourth cause to this pattern may be the seasonal availability of medicinal plants [135], although this behaviour has not yet been observed in baboons.

In contrast to the previous results, lower amounts of parasites have also been found during wet season, regarding *Oesophagostomum* [124] and *Trichostrongylus* [122]. This wet season attenuation of parasite transmission has been explained by the increased dung beetle activity; dispersing, burying and masticating the nematode larvae in baboon faeces [172]. A portion of the eggs left in the moist soil are also invaded by fungi [16,173]. Another explanation to the lower parasite count like spirurids in wet season could be the decreased consumption of some intermediate host [174].

Finally, some researchers conclude that despite sharp contrast between hot dry seasons and cooler wet seasons, nonstatistical comparison of parasites found shows no differences between seasons [48].

3.3 - Fauna & flora of the environment

Correlations between the vegetal habitat of baboons and their intestinal parasites have been observed. For example the development of larvae in *Trichuris* eggs in the soil requires conditions which are found in the shade, under trees [54].

The parasite community structure and richness can also be predicted by the composition and densities of other animals present in the environment, reflecting the ecological disturbance of the habitat. Positive correlations have been reported between parasite richness in baboons and parasites found in sympatric grazing animals [175] in other areas, so it would be interesting to lead similar studies in the PNNK, with its very diverse fauna.

Ecological disturbance of habitat for example through forest fragmentation can alter population densities. Monkey densities themselves have been shown to be positively correlated with prevalence and richness of parasites harboured [176] as well as to the spread of directly transmitted parasites [177]. Would the parasitism of Guinea baboons evolve according to the level of human activity over time?

Thirdly, human disturbance may also have an effect on the diet as well as the foraging, and therefore ranging, activities of baboons. During our field study, it was observed that the baboons were attracted to the new Lodge being built, feeding off the food (rice, cooked vegetables, fruit brought from the city.) left on purpose at observation points - for tourists to photograph wild animals (mostly hogs and herbivores). On several occasions it was asked the staff to stop putting food out as it disturbed the natural behaviour (less foraging, smaller home range, etc…) and diet of the troop. As the lodge opened to tourists soon after, it could be interesting to study the parasites of this same baboon population in the future to see if the modification of foraging habits and diet content associated to human proximity has an influence on their gastro-intestinal parasites.

Regardless the cause, negative impact of human activities on the microbiome of intestinal tract of primates has been demonstrated. With habitat degradation resulting from rapid global change, and the knowledge we have nowadays regarding the importance of gut microbiome in primate health, it seems important to focus on the later when conservation is of topic. [178].

4. Intrinsic factors which could be linked to parasite infection

4.1 - Immunity

While it is often suggested that environmental conditions may be parameters influencing parasitic egg survival and therefore transmission, immunity may also play its part. Although a study in rabbits showed that egg-specific antibodies are formed in response to helminth infection, they do not seem to influence egg hatchability [179]. This could be due to the co-evolution of mammalian immune systems

and helminths, leading to a “disease tolerance” [180]. However, it is possible for immunity and parasitism may still influence each other.
Several studies reveal a synergy between stress and parasitic infections in mammals and primates, for example with *Balantidium* spp. [181,182] or strongyle eggs explained by stress weakening the immune system [2].

Additionally, primate immune systems react to helminth infections via type-2 interleukin mediated responses, providing resistance towards the parasites. The cellular response seen in chronic parasitism activated the immune regulation, and together with GC prod. suppress inflammatory responses. But Glucocorticoid immunosuppression may also increase susceptibility to parasites, leading to higher infestations (for example: high levels correlated with high Trichuris egg count [180], or to infections by other para. A “vicious cycle” may appear between parasite infection and body condition, meaning that parasitism may lead to a weaker immune system, reducing the host’s ability to resist to the next infections.

Host immunity has also been proven to influence the impact of climate change on dynamics of parasitism [183]. Indeed, as discussed previously, climate warming increases the availability of infective stages of helminth species. It has been observed that immunity can reduce the severity of population infection but can also shift parasite intensity towards the younger individuals (with lower immunity) exposed to climate warming.

4.2 - Age

As seen previously, younger individuals may show increased susceptibility to parasitism due to an unexperienced immune system, but on the other hand, age could lead to cumulative exposure to parasites as well as immuno-senescence. To support this hypothesis, it has been reported that female baboon infection risks were often associated with old age for example with a positive correlation observed between baboon age and helminth burden [184,185].
In contrast, age does not seem to have a significant influence on the prevalence of Strongyloides sp. in wild orangutans [186].

4.3 - Reproductive physiology

Several studies have been and are being conducted regarding links between parasitism and reproductive history and status of primates, to understand the complex relationship as it seems that neither is fully a cause or a consequence. Indeed reproductive status may alter immune function and impose high energetic demands that should increase host susceptibility, or parasites may be to the cost of individual fitness. Directions of correlation are also parasite-dependant. For example, it has been found that female baboons with higher parasite richness exhibit longer interbirth intervals [184], but in humans *Ascaris* infections are associated with earlier first pregnancies and shorter interbirth intervals, criteria of higher fertility [187].

5. Zoonotic importance

5.1 - *Zoonotic agents of the Simenti baboons*

STH infect approximately one sixth of the world population, with highest prevalence rates in children living in sub-Saharan Africa. Although often asymptomatic, nematode infections (and sometimes protozoa infections) can lead to heavy clinical infestations, most frequently in children, impacting their development.

Six zoonotic gastrointestinal helminths were identified: *Trichuris* sp., *Trichostrongylus* sp., *Enterobius* spp. *Oesophagostomum* sp., *Strongyloides stercoralis* and *Strongyloides fülleborni.*

We could not identify the species of the *Entamoeba* cysts observed but if they were *E. histolytica* and *E. coli*, they would be considered easily transmissible from animals to humans through contaminated food, water and hands [188], causing debilitating diarrhoea in humans.

5.2 - Transmission risks

We have presented the molecular identification and phylogeny of both *S. stercoralis* and *S. fülleborni* infections of wild Guinea baboons in Senegal.
We observed that the haplotypes of *S. stercoralis* obtained in our study are shared by Guinea baboons and humans on different continents and are very similar to those found in dogs. This nucleotide polymorphism is consistent with previous reports [85] and would support the suggestion of dogs being a reservoir of zoonotic *S. stercoralis*, spreading the infection to primates and humans (whiles *S. canis* would infect only dogs) [190]. These results are of great importance as the neglected STH can cause serious life-threatening illness in immunocompromised humans [162].
The presence of hookworms in Guinea baboon populations is also of concern from a public health perspective due to its zoonotic potential: for example *Oesophagostomum* sp. is the most common species of bursate nematode infecting humans in Africa, Obanda *et al.* [167] detected *O. bifurcum* in Amboseli baboons which were very similar to isolated from humans, as well as *Trichostrongylus colubriformis*, considered zoonotic and of veterinary importance (towards livestock). However, molecular DNA analyses showed that *O. bifurcum* from humans was genetically distinct from *O. bifurcum* from the Olive baboon [191]. It would therefore be interesting to explore the GI para of humans living in the PNNK or in the surroundings, to confirm if baboons are reservoir, and if spill over or spill back is possible .

Whether the whipworm eggs found in Guinea baboons are *T. trichuria* or primate-derived *Trichuris* spp. which probably cross infect NHP and humans, they are another important causative agent of Trichuriasis, a gastro-intestinal disease affecting over half a billion people. However, it has recently been reported that *T. trichuria* consists of different genetic lineages [102] and it is yet uncertain if the primate-derived *T. trichuria* variation can cross infect humans. Identifying the precise species in our samples would be of importance concerning zoonotic potential from NHP to humans as well as the use of appropriate control strategy for different cryptic species.

Although attention is brought to parasite when transmission from wildlife to humans is suspected or confirmed, less is the case when it goes in the opposite direction; from humans to wildlife, which is at least equally as important. For example, in a study of parasites in long-tailed Macaques living in Thailand [192], no parasites present in humans were found in the macaques, but the macaques living in habitats modified by humans presented parasites (including *S. fülleborni*) that were not present in more isolated macaque groups. Proximity with humans and related activities may therefore alter the parasite community of wild-life, potential affecting their survival.

5.3 - Phylogeny: Hypothesis on Strongyloides evolution

According to the phylogenetic analysis, *S. stercoralis* and *S. fülleborni* occurring simultaneously in this baboon population are basal to one another. Regarding *S. fülleborni*, we have observed that the strains found in Guinea baboons belong to a clade containing strains that are only infectious to primates in Africa, distinct from the second clade restricted to strains that have isolated from primates in Asia. This strongly supported grouping comforts the suggestion by Barrat et al. [154] that each cluster is a potential geographic-adapted sub-variant of *S. fülleborni*, as opposed to the host-adapted variants of *S. stercoralis* which would have evolved from dog haplotypes [189], spreading across the world. Hasegawa *et al.* [193] and Obanda *et al.* [167] also observed that *S. fülleborni* populations, respectively in Tanzania and Kenya, were different from those in other countries. Moreover, distinct evolution lines are observed within the African clade, maybe due to an additional host-adapted evolution or to a more specific local haplotype.

To continue studying the complexity of *S. fülleborni*, it would be interesting to sequence the Strongyloides (sub-)species found in other baboon species to see if they belong to the same clades but different clusters as those in our study and if they share the same common ancestor, as do the *S. fülleborni* found in a yellow baboon (AB526306). More studies sequencing Strongyloides in other African primates would also be needed, to verify the hypothesis of a host-adapted evolution.

Regarding *Strongyloides stercoralis*, it had been demonstrated that strains can adapt from human to dog host and vice versa, changing its virulence (autoinfection and hyper-infection capabilities) and migratory pathways [194]. However this zoonotic potential of dog strains is now being challenged [195,196]. While zoonosis might be less common than previously proposed, the transmission possibilities between dogs and humans, human and baboons, and dogs and baboons are still not fully understood dog infections could still pose a significant transmission risk to baboons and to people [197]. This should be considered when looking at conservation-related topics.

5.4 - Prevention & control measures

Due to direct life cycles, *Strongyloides* sp. and *Necator americanus*, common in Guinea baboons, cause a high risk of zoonotic issues. Therefore direct contact between humans and NHP should be discouraged, for example through health education. In places were humans and baboons cohabite, particular attention should be drawn to the hygiene concerning food preparing areas and material, as well as sanitation of waste disposal areas, to limit faecal contamination and STH infections. Footwear,

especially in these areas is necessary to reduce *N. americanus* percutan infection. Indirect contact should also be avoided to reduce the risk of infection by parasites which use an intermediate host. One of the first things to do is to install fences to protect drinking water sources from animals, NHP and intermediate hosts included. In case of parasitic disease control programmes, control of the intermediate hosts must be taken into account, for example snail or cockroach control respectively for schistosomiasis or spirurid infections [198].

As children living in less developed countries are likely to be infected by STH and these result in impaired physical, intellectual and cognitive development [94], parasitic disease control strategies involving worm eradication has been done using mass drug administration in children [161]. School deworming programs enhance education (school attendance, and students' performances : cognition development, memory, intellectual capacity) while increasing the their quality of life as well as the health of their communities (reduced malnutrition, anaemia, stunting…) [199].
However, it does not seem to be the perfect solution. On top of funding issues, there are concerns about the sustainability of periodic deworming with benzimidazole anthelmintics and the emergence of resistance [94,197].
Additionally, it may bring to surface other medical issues in those areas. The Hygiene hypothesis states that infants and children who lack exposure to infectious agents are more susceptible to allergic diseases via modulation of immune system development as Mary Ruebush explains in her book Why Dirt is Good [200]. For example, a study in Gambia found that eradication of worms in some villages led to increased skin reactions to allergies among children [201].

On the other hand, vaccines could be administered to prevent protozoa and helminth GI parasitic infection. Their observed effect is encouraging but studies still need to be done [202].

5.5 - Use of the zoonotic potential in favour of development

Although *Necator americanus* is found in humans too, it has little pathogenic potential, and therefore the zoonotic parasites are not always a threat to human health. On the contrary, it could see its use in helmintherapy rise, due to two supported reasons: (i) improving nutritional status [203]. For example, hookworms have been reported to help people who suffer from Crohn's disease via better nutrient absorption. STH secrete particular molecules which have various therapeutical potentials [94]. It is suggested that although GI parasites can impair certain functions of the body, they can simultaneously protect against immune mediated conditions such as atopic and autoimmune disease and severe malaria [204]. The second reason supported is that the immune responses triggered by GI para may have a beneficial effect in restoring healthy gut microbiota and mucosal barrier function during inflammatory bowel disease [205] and treating food allergies [206].
Clinical trials are ongoing but there is an urgency to understand heterogeneity in immune responses against helminths in order to maximize clinical benefit [180]. On top of the therapeutical potential, the understanding of mechanisms can be used as a to study anti-inflammatory response and defining new therapeutic targets [207].

6. Behaviours

Some parasites may cause big blood loss (for example (*N. americanus* can cause up to 30µL blood loss/day [208]. The consequential iron-deficiency anaemia [209] can lead to mental retardation and growth insufficiency in children, causing abnormal learning & behaviour. Seen as baboons are not so distant from humans, one could wonder if the abnormal learning and behaviour may also occur in baboons…

Behaviours may be observed in regard to the individual towards itself, towards its environment or towards the other individuals around. Regarding the latter, frequency, length, and type of interactions could be used to define social connectedness.

Studies have suggested that high social connectedness correlates with higher helminth burdens [210,211]. This could be explained by more social interactions such as grooming, playing, anogenital greetings, etc… increasing exposure to parasites transmitted via *per cutan* and faeco-oral routes. Some researchers have linked connectedness to sex of the individuals, finding for example that a higher *T. trichuria* egg-count and parasite richness was found in male yellow baboons which were more socially connected to females, and that a higher probability of *A. caucasica* infection was found in females with high social connectedness with males. This would support the suggestions above, that (i) more social contacts increase parasitic exposure, but also that (ii) investing in mating effort may lead to trade-offs with immune function, increasing parasite susceptibility, that (iii) mating behaviour is linked to higher production of testosterone which is immunosuppressive, also increasing susceptibility [212], or that (iv) social connectedness may be associated with access to the best food resources, including intermediate hosts of parasites [184].

On the contrary it has also been suggested that social isolation leads to higher parasitism. Akyini *et al.* found higher *T. trichuris* egg counts in female yellow baboons who were more socially isolated from males as well as higher parasite richness in females who were more socially isolated from females.

With regards to dispersal, a study showed that males who disperse frequently to new social groups have higher parasite richness than males who reside stably in a group [213].

On top of the level of social contacts, the rate of gastrointestinal parasites spread in NHP can be positively corelated to group size [168] as well as intensive territorial patterns, both which would imply continuous exposure to parasites in the environment. These are parameters which could be studied in further research concerning the Simenti baboons.

Dietary behaviours of baboons may also be linked to parasitism. In many anthropoid primates, it has been shown that diet composition as well as individual ranging and foraging behaviour was positively associated with diversity of parasite, especially those with complex life cycles [168]. No research is being conducted yet on individual feeding habits of the Simenti baboons but once there is data, this would be worth exploring.

In order to avoid soil-transmitted parasites, adaptive strategies have been hypothesized. Spatial isolation of individuals or social groups has been identified as a strategy preventing infection by pathogens (Loehle, 1995). Frequent short-distance change of sleeping site by whole groups of yellow baboons has been suggested as a strategy to avoid potential infection in the build-up of faeces under the trees [16]. However a study lead by McGrew [48] revealed that it was not the case in Guinea baboons. As the study was over thirty-five years ago and accessibility and diagnostic methods have evolved, it could be interesting to check again if the correlation exists.

In any case, it remains quite delicate to state that a behaviour leads to a parasitic trend or whether parasites cause certain behaviour. For example, both spatial and social isolations could be correlated to the fact that in some NHP species, very old or ill individuals diverge from their community months, weeks or just days prior to death. Individuals may do this to avoid contaminating their community, or parasites may influence the isolation as a strategy to disperse further away… There is some evidence that parasitized mammals modify their grazing behaviour [214,215] and that other primates self-medicate [134] so variations in baboon foraging and feeding habits could also be a consequence of parasitism rather than a cause.
It is also the case regarding immunity. Is a primate more susceptible to parasites because of its weaker immune system (due to illness, age, co-infections, etc), or rather does parasite load alone weaken the immune system? It seems that parasite-host relationships are multifactorial.

These are all challenging questions and interesting suggestions, for which it would be worth to analyse the rest of the samples collected during this study as well as future samples, and in parallel to study the longitudinal behavioural data, to look for correlations.

Part 8. Conclusion

Gastro-intestinal parasites of wild Guinea baboons have been detected: a variety of helminths, a genus of amoeba and a large number unidentified *Balantioides*-like ciliates were observed.

The high prevalence of protozoa observed in this study does not support previous theories of higher nematode infections in forested habitats versus higher protozoa infections in desert environments. No significant correlations with sex, age or party were observed.
However diagnostic methods should be optimised and the rest of the collected samples should be analysed before jumping to conclusions. One must keep in mind that previous studies did not use molecular tools which are absolutely necessary to identify most parasites to species with certainty.

The molecular detection and phylogenetic analysis allow suggestions regarding *Strongyloides* spp. evolution.

Taking into consideration the optimisation of methods discussed, the presented results - along with the rest of the samples yet to be analysed - can serve as a base for longitudinal parasitological studies on this particular population.

This study will hopefully stimulate further field and laboratory pluri-disciplinary research, using the long term behavioural data available, on basic mechanisms of parasite transmission in natural primate populations and, testing various hypothesis such as parasite avoidance, self-regulation, and many more. Additionally corelations between parasitism and social connectedness of individuals and/or etho-ecology of the Guinea baboons may show selective pressure by parasites on evolution of primates as well as parasite evolution/existence strategies.

Some of the present parasites may be able to cross-infect endangered sympatric species as well as livestock and humans sharing the same ecosystem.
Studies on parasite presence and prevalence in sympatric species is also needed as changes in microfaunal patterns can serve as an ecological indicator of habitat degradation.
Research on the occurrence of these parasites in local human populations seems appropriate to confirm zoonotic risks of the strains of *Trichuris* sp., *Enterobius* sp., *Oesophagostomum* sp., *Necator* sp. and *Strongyloides* spp. found in this study.

To conclude, investigating and understanding the role of parasitism in primate ecology, behaviour and evolution is of great importance for public health and animal conservation matters. As pointed out nearly 60 years ago: “greater cooperation between parasitologists, veterinarians, and animal ecologists is strongly urged” (Yamashita, 1963).

Figure 25 - Morning sunbath, grooming and playing in Simenti, along the Niokolo river.
(Photo: A. Coles)

Part 9. Bibliography

1. Ferreira da Silva MJ, Godinho R, Casanova C, Minhós T, Sá R, Bruford MW. Assessing the impact of hunting pressure on population structure of Guinea baboons (Papio papio) in Guinea-Bissau. Conserv Genet. 2014 Dec;15(6):1339–55.
2. Chapman CA, Gillespie TR, Goldberg TL. Primates and the Ecology of their Infectious Diseases: How will Anthropogenic Change Affect Host-Parasite Interactions? Evol Anthropol. 2005 Aug 25;14(4):134–44.
3. Devaux CA, Mediannikov O, Medkour H, Raoult D. Infectious Disease Risk Across the Growing Human-Non Human Primate Interface: A Review of the Evidence. Front Public Health. 2019 Nov 5;7:305.
4. Magden ER, Mansfield KG, Simmons JH, Abee CR. Nonhuman Primates. In: Laboratory Animal Medicine [Internet]. Elsevier; 2015 [cited 2021 Mar 5]. p. 771–930. Available from: https://linkinghub.elsevier.com/retrieve/pii/B9780124095274000171
5. Knauf S, Barnett U, Maciej P, Klapproth M, Ndao I, Frischmann S, Fischer J, Zinner D, Liu H. High Prevalence of Antibodies against the Bacterium Treponema pallidum in Senegalese Guinea Baboons (Papio papio). Hassan I, editor. PLoS ONE. 2015 Nov 20;10(11):e0143100.
6. World Health Organisation. Soil-transmitted helminth infections [Internet]. [cited 2021 Feb 12]. Available from: https://www.who.int/news-room/fact-sheets/detail/soil-transmitted-helminth-infections
7. Mogaji H, Dedeke GA, Bada B, Bankole S, Adeniji A, Fagbenro M, Omitola O, Oluwole A, Odoemene S, Abe E, Mafiana C, Ekpo U. Distribution of ascariasis, trichuriasis and hookworm infections in Ogun State, Southwestern Nigeria. PLOS ONE. 2020 Jun 8;15:e0233423.
8. Brown C. Emerging Zoonoses and Pathogens of Public Health Significance—An Overview. Revue scientifique et technique (International Office of Epizootics). 2004 Sep 1;23:435–42.
9. Sapolsky RM, Share LJ. A Pacific Culture among Wild Baboons: Its Emergence and Transmission. PLOS Biology. 2004 avr;2(4):e106.
10. Gillespie TR, Lonsdorf EV, Canfield EP, Meyer DJ, Nadler Y, Raphael J, Pusey AE, Pond J, Pauley J, Mlengeya T, Travis DA. Demographic and ecological effects on patterns of parasitism in eastern chimpanzees (Pan troglodytes schweinfurthii) in Gombe National Park, Tanzania. Am J Phys Anthropol. 2010 Dec;143(4):534–44.
11. Kalema-Zikusoka G, Rothman JM, Fox MT. Intestinal parasites and bacteria of mountain gorillas (Gorilla beringei beringei) in Bwindi Impenetrable National Park, Uganda. Primates. 2005 Jan;46(1):59–63.
12. Medkour H, Amona I, Laidoudi Y, Davoust B, Bitam I, Levasseur A, Akiana J, Diatta G, Pacheco L, Gorsane S, Sokhna C, Hernandez-Aguilar RA, Barciela A, Fenollar F, Raoult D, Mediannikov O. Parasitic Infections in African Humans and Non-Human Primates. Pathogens. 2020 Jul 11;9(7):561.
13. Howells ME, Pruetz J, Gillespie TR. Patterns of gastro-intestinal parasites and commensals as an index of population and ecosystem health: the case of sympatric western chimpanzees (Pan troglodytes verus) and guinea baboons (Papio hamadryas papio) at Fongoli, Senegal. Am J Primatol. 2010;73(2):173–9.
14. Paciência FMD, Rushmore J, Chuma IS, Lipende IF, Caillaud D, Knauf S, Zinner D. Mating avoidance in female olive baboons (*Papio anubis*) infected by *Treponema pallidum.* Sci Adv. 2019 Dec;5(12):eaaw9724.
15. Fischer J, Higham JP, Alberts SC, Barrett L, Beehner JC, Bergman TJ, Carter AJ, Collins A, Elton S, Fagot J, Ferreira da Silva MJ, Hammerschmidt K, Henzi P, Jolly CJ, Knauf S, Kopp GH, Rogers J, Roos C, Ross C, Seyfarth RM, Silk J, Snyder-Mackler N, Staedele V, Swedell L, Wilson ML, Zinner D. Insights into the evolution of social systems and species from baboon studies. eLife. 2019 Nov 12;8:e50989.
16. Hausfater G, Meade BJ. Alternation of sleeping groves by yellow baboons (Papio cynocephalus) as a strategy for parasite avoidance. Primates. 1982 Apr;23(2):287–97.
17. Lozano G. Parasitic Stress and Self-Medication in Wild Animals. In: Advances in The Study of Behavior - ADVAN STUDY BEHAV. 1998. p. 291–317.
18. Whitney RA. Chapter 3 - Taxonomy. In: Bennett BT, Abee CR, Henrickson R, editors. Nonhuman Primates in Biomedical Research [Internet]. San Diego: Academic Press; 1995 [cited 2020 Apr 3]. p. 33–47. (American College of Laboratory Animal Medicine). Available from: https://www.sciencedirect.com/science/article/pii/B9780120886616500053
19. Rogers J, Raveendran M, Harris RA, Mailund T, Leppälä K, Athanasiadis G, Schierup MH, Cheng J, Munch K, Walker JA, Konkel MK, Jordan V, Steely CJ, Beckstrom TO, Bergey C, Burrell A, Schrempf D, Noll A, Kothe M, Kopp GH, Liu Y, Murali S, Billis K, Martin FJ, Muffato M, Cox L, Else J, Disotell T, Muzny DM, Phillips-Conroy J, Aken B, Eichler EE, Marques-Bonet T, Kosiol C, Batzer MA, Hahn MW, Tung J, Zinner D, Roos C, Jolly CJ, Gibbs RA, Worley KC, Baboon Genome Analysis Consortium. The comparative genomics and complex population history of *Papio* baboons. Sci Adv. 2019 Jan;5(1):eaau6947.
20. Fleagle JG. Primate Taxonomy. By Colin Groves. The Quarterly Review of Biology. 2002 Mar 1;77(1):81–81.
21. IUCN. Papio papio: Wallis, J., Alonso, C., Barlow, C., Brito, J., Ferreira da Silva, M.J., Hernansaiz, A., Kopp, G.H., Vale, C. & Zinner, D.: The IUCN Red List of Threatened Species 2020: e.T16018A17952926 [Internet]. International Union for Conservation of Nature; 2018 [cited 2021 Apr 3]. Available from: https://www.iucnredlist.org/species/16018/17952926

22. Ferreira da Silva MJ, Casanova C, Godinho R. On the western fringe of baboon distribution: mitochondrial D-loop diversity of Guinea Baboons (Papio papio Desmarest, 1820) (Primates: Cercopithecidae) in Coastal Guinea-Bissau, western Africa. J Threat Taxa. 2013 Jun 26;5(10):4441–50.
23. Ebbert MA, McGrew WC, Marchant LF. Community composition, correlations among taxa, prevalence, and richness in gastrointestinal parasites of baboons in Senegal, West Africa. Primates. 2013 Apr;54(2):183–9.
24. McGrew WC, Tutin CEG, Collins DA, File SK. Intestinal parasites of sympatric Pan troglodytes and Papio Spp. at two sites: Gombe (Tanzania) and Mt. Assirik (Senegal). Am J Primatol. 1989;17(2):147–55.
25. Sharman MJ. Feeding, ranging and social organisation of the Guinea baboon [Internet] [Thesis]. University of St Andrews; 1982 [cited 2021 Apr 3]. Available from: https://research-repository.st-andrews.ac.uk/handle/10023/2833
26. McGrew W. Primates in caves: two new reports of Papio spp. Journal of Human Evolution. 2003 Apr;44(4):521–6.
27. Patzelt A, Kopp GH, Ndao I, Kalbitzer U, Zinner D, Fischer J. Male tolerance and male-male bonds in a multilevel primate society. Proceedings of the National Academy of Sciences. 2014 Oct 14;111(41):14740–5.
28. Goffe AS, Zinner D, Fischer J. Sex and friendship in a multilevel society: behavioural patterns and associations between female and male Guinea baboons. Behav Ecol Sociobiol. 2016 Mar;70(3):323–36.
29. Goffe AS, Fischer J. Meat sharing between male and female Guinea baboons (<i>Papio papio</i>). Primate Biol. 2016 Feb 4;3(1):1–8.
30. Patzelt A, Zinner D, Fickenscher G, Diedhiou S, Camara B, Stahl D, Fischer J. Group Composition of Guinea Baboons (Papio papio) at a Water Place Suggests a Fluid Social Organization. Int J Primatol. 2011 Jun;32(3):652–68.
31. Ebbert MA, McGREW WC, Marchant LF. Differences between chimpanzee and baboon gastrointestinal parasite communities. Parasitology. 2015 Jun;142(7):958–67.
32. Kopp GH, Fischer J, Patzelt A, Roos C, Zinner D. Population genetic insights into the social organization of Guinea baboons (*Papio papio*): Evidence for female-biased dispersal. Am J Primatol. 2015 Aug;77(8):878–89.
33. Fischer J, Kopp GH, Dal Pesco F, Goffe A, Hammerschmidt K, Kalbitzer U, Klapproth M, Maciej P, Ndao I, Patzelt A, Zinner D. Charting the neglected West: The social system of Guinea baboons. Am J Phys Anthropol. 2017 Jan;162(S63):15–31.
34. Zinner D, Klapproth M, Schell A, Ohrndorf L, Chala D, Fischer J. Comparative Ecology of Guinea Baboons (Papio papio). :22.
35. HILTON-TAYLOR C. IUCN Red List of threatened species. IUCN http://www.redlist.org [Internet]. 2000 [cited 2021 Apr 3]; Available from: https://ci.nii.ac.jp/naid/10013249465/
36. Kingdon J. The Kingdon Field Guide to African Mammals. Journal of Mammalogy. 1999;80(2):692–3.
37. Byrne RW. Distance Vocalisations of Guinea Baboons (Papio papio) in Senegal: An Analysis of Function. Behaviour. 1981;78(3/4):283–313.
38. Melnick D, Pearl M. Cercopithecines in multi-male groups: Genetic diversity and population structure. In 1987.
39. Hamilton WJ. Baboon sleeping site preferences and relationships to primate grouping patterns. Am J Primatol. 1982;3(1–4):41–53.
40. Cowlishaw G. Vulnerability To Predation in Baboon Populations. Behav. 1994;131(3–4):293–304.
41. Barton RA, Byrne RW, Whiten A. Ecology, Feeding Competition and Social Structure in Baboons. Behavioral Ecology and Sociobiology. 1996;38(5):321–9.
42. Casanova CCN, Costa S. Are animals and forests forever? Perceptions o wildlife at Cantanhez Forest National Park, Guinea-Bissau. [cited 2021 Apr 3]; Available from: https://www.academia.edu/23622522/Are_animals_and_forests_forever_Perceptions_o_wildlife_at_Cantanhez_Forest_National_Park_Guinea_Bissau
43. Shefferly N. Papio papio (Guinea baboon) [Internet]. Animal Diversity Web. 2004 [cited 2020 Mar 8]. Available from: https://animaldiversity.org/accounts/Papio_papio/
44. Jolly C. Species, Subspecies, and Baboon Systematics. Species, Subspecies, and Baboon Systematics. 1993 Jan 1;67–107.
45. Bronikowski AM, Alberts SC, Altmann J, Packer C, Carey KD, Tatar M. The aging baboon: Comparative demography in a non-human primate. Proceedings of the National Academy of Sciences. 2002 Jul 9;99(14):9591–5.
46. Gauthier CA. Reproductive parameters and paracallosal skin color changes in captive female guinea baboons, Papio papio. Am J Primatol. 1999;47(1):67–74.
47. Dunbar R. Primate Social System. Bioscience. 1988 Jan 1;39.
48. Anderson JR, McGrew WC. Guinea baboons (Papio papio) at a sleeping site. Am J Primatol. 1984;6(1):1–14.
49. Barrett L, Gaynor D, Rendall D, Mitchell D, Henzi SP. Habitual cave use and thermoregulation in chacma baboons (Papio hamadryas ursinus). Journal of Human Evolution. 46(2):215–22.
50. Brain C. The Hunters or the Hunted? An Introduction to African Cave Taphonomy. Vol. 48, American Antiquity. 1981.
51. Matsumoto-Oda A. How surviving baboons behaved after leopard predation: a case report. Anthropological Science. 2015;123(1):13–7.
52. Redmond IM, Shoshani J. Mount Elgon's Elephants Are in Peril. Elephant. 1987 Dec 20;2(3):46–66.

53. Nunn C, Altizer S, Altizer SM. Infectious Diseases in Primates: Behavior, Ecology and Evolution. OUP Oxford; 2006. 397 p.
54. Appleton CC, Brain C. Gastro-intestinal parasites of Papio cynocephalus ursinus Iiving in the central Namib desert, Namibia. Afr J Ecol. 1995;33:257–65.
55. Bezjian M, Gillespie TR, Chapman CA, Greiner EC. Coprologic evidence of gastrointestinal helminths of Forest baboons, Papio anubis, in Kibale National Park, Uganda. Journal of Wildlife Diseases. 2008 Oct;44(4):878–87.
56. Gillespie TR. Noninvasive Assessment of Gastrointestinal Parasite Infections in Free-Ranging Primates. Int J Primatol. 2006 Oct 6;27(4):1129–43.
57. Hahn NE, Proulx D, Muruthi PM, Alberts S, Altmann J. [No title found]. International Journal of Primatology. 2003;24(2):271–9.
58. Müller-Graf CDM, Collins DA, Woolhouse MEJ. Intestinal parasite burden in five troops of olive baboons (*Papio cynocephalus anubis*) in Gombe Stream National Park, Tanzania. Parasitology. 1996 May;112(5):489–97.
59. Hope K, Goldsmith ML, Graczyk T. Parasitic health of olive baboons in Bwindi Impenetrable National Park, Uganda. Veterinary Parasitology. 2004 Jun;122(2):165–70.
60. Larbi JA, Akyeampong S, Abubakari A, Offei Addo S, Okoto D, Hanson H. Zoonotic Gastrointestinal Parasites of Baboons (*Papio anubis*) in the Shai Hill Reserve in Ghana. BioMed Research International. 2020 Mar 11;2020:1–6.
61. Weyher AH, Ross C, Semple S. Gastrointestinal Parasites in Crop Raiding and Wild Foraging Papio anubis in Nigeria. Int J Primatol. 2006 Dec 12;27(6):1519–34.
62. Mafuyai H, Barshep Y, Audu B, Kumbak D, Ojobe T. Baboons as potential reservoirs of zoonotic gastrointestinal parasite infections at Yankari National Park, Nigeria. Afr H Sci. 2013 Sep 5;13(2):252–4.
63. UNIVERSITY OF DAR ES SALAAM. 2020;46:14.
64. Prenant T. Les parasites digestifs des primates non-humains en captivité dans les parcs zoologiques en France: réalisation d'un atlas d'aide à la diagnose [Thèse de doctorat vétérinaire]. [France]: Ecole nationale vétérinaire d'Alfort; 2018.
65. Kouassi RYW, McGraw SW, Yao PK, Abou-Bacar A, Brunet J, Pesson B, Bonfoh B, N'goran EK, Candolfi E. Diversity and prevalence of gastrointestinal parasites in seven non-human primates of the Taï National Park, Côte d'Ivoire. Parasite. 2015;22:1.
66. Petrášová J, Modrý D, Huffman MA, Mapua MI, Bobáková L, Mazoch V, Singh J, Kaur T, Petrželková KJ. Gastrointestinal Parasites of Indigenous and Introduced Primate Species of Rubondo Island National Park, Tanzania. Int J Primatol. 2010 Oct;31(5):920–36.
67. Lilly A, Mehlman P, Doran D. Intestinal Parasites in Gorillas, Chimpanzees, and Humans at Mondika Research Site, Dzanga-Ndoki National Park, Central African Republic. International Journal of Primatology. 2002 Jan 6;23:555–73.
68. Kooriyama T, Hasegawa H, Shimozuru M, Tsubota T, Nishida T, Iwaki T. Parasitology of five primates in Mahale Mountains National Park, Tanzania. Primates. 2012 Oct;53(4):365–75.
69. Munene E, Otsyula M, Mbaabu DAN, Mutahi WT, Muriuki SMK, Muchemi GM. Helminth and protozoan gastrointestinal tract parasites in captive and wild-trapped African non-human primates. Veterinary Parasitology. 1998 Aug;78(3):195–201.
70. Deere JR, Parsons MB, Lonsdorf EV, Lipende I, Kamenya S, Collins DA, Travis DA, Gillespie TR. Entamoeba histolytica infection in humans, chimpanzees and baboons in the Greater Gombe Ecosystem, Tanzania. Parasitology. 2019 Aug;146(9):1116–22.
71. Ravasi DF, O'Riain MJ, Davids F, Illing N. Phylogenetic Evidence That Two Distinct Trichuris Genotypes Infect both Humans and Non-Human Primates. Parkinson J, editor. PLoS ONE. 2012 Aug 28;7(8):e44187.
72. Adenusi AA, Akinyemi MI, Akinsanya D. Domiciliary Cockroaches as Carriers of Human Intestinal Parasites in Lagos Metropolis, Southwest Nigeria: Implications for Public Health. J Arthropod Borne Dis. 2018 Jun;12(2):141–51.
73. Pedersen AB, Altizer S, Poss M, Cunningham AA, Nunn CL. Patterns of host specificity and transmission among parasites of wild primates. International Journal for Parasitology. 2005 May 1;35(6):647–57.
74. Levine ND. Protozoan Parasites of Domestic Animals and of Man. Burgess Publishing Company; 1973. 424 p.
75. Levine ND. Protozoan parasites of nonhuman primates as zoonotic agents. Lab Anim Care. 1970 Apr;20(2):377–82.
76. Strait K, Else JG, Eberhard ML. Chapter 4 - Parasitic Diseases of Nonhuman Primates. In: Abee CR, Mansfield K, Tardif S, Morris T, editors. Nonhuman Primates in Biomedical Research (Second Edition) [Internet]. Boston: Academic Press; 2012 [cited 2021 Apr 3]. p. 197–297. (American College of Laboratory Animal Medicine). Available from: https://www.sciencedirect.com/science/article/pii/B9780123813664000043
77. Romain L. Les parasites intestinaux chez le Macaque Crabier (Macaca fascicularis). Ecole nationale vétérianire d'Alfort; 2009.
78. Laughlin RC, Temesvari LA. Cellular and molecular mechanisms that underlie Entamoeba histolytica pathogenesis: prospects for intervention. Expert Rev Mol Med. 2005 Jul 18;7(13):1–19.
79. Copeland BE, Kimber J. Nuclear size in diagnosis of Entamoeba histolytica on stained smears. Tech Bull Regist Med Technol. 1968 Oct;38(10):282–6.
80. Wilson DW, Day PA, Brummer MEG. Diarrhea Associated with Cryptosporidium spp in Juvenile Macaques. Vet Pathol. 1984 Jul 1;21(4):447–50.

81. Griffiths J, Maguire JH, Heggenhougen K, Quah SR. Public Health and Infectious Diseases. Elsevier; 2010. 504 p.
82. Eberhard ML, da Silva AJ, Lilley BG, Pieniazek NJ. Morphologic and molecular characterization of new Cyclospora species from Ethiopian monkeys: C. cercopitheci sp.n., C. colobi sp.n., and C. papionis sp.n. Emerg Infect Dis. 1999 Oct;5(5):651–8.
83. Smith HV, Paton CA, Girdwood RW, Mtambo MM. Cyclospora in non-human primates in Gombe, Tanzania. Vet Rec. 1996 May 25;138(21):528.
84. Lynn D. The Ciliated Protozoa: Characterization, Classification, and Guide to the Literature [Internet]. 3rd ed. Springer Netherlands; 2008 [cited 2021 Apr 3]. Available from: https://www.springer.com/gp/book/9781402082382
85. Schuster FL, Ramirez-Avila L. Current World Status of Balantidium coli. Clinical Microbiology Reviews. 2008 Oct 1;21(4):626–38.
86. Strait K, Else JG, Eberhard ML. Parasitic Diseases of Nonhuman Primates. In: Nonhuman Primates in Biomedical Research [Internet]. Elsevier; 2012 [cited 2021 Mar 5]. p. 197–297. Available from: https://linkinghub.elsevier.com/retrieve/pii/B9780123813664000043
87. Lankester F, Mätz-Rensing K, Kiyang J, Jensen SA, Weiss S, Leendertz FH. Fatal ulcerative colitis in a western lowland gorilla (gorilla gorilla gorilla). Journal of Medical Primatology. 2008;37(6):297–302.
88. Roberts LS, Schmidt GD, Janovy J. Gerald D. Schmidt & Larry S. Roberts' foundations of parasitology. 8th ed. Boston: McGraw-Hill Higher Education; 2009. 701 p.
89. Yamashita J. Ecological relationships between parasites and primates: I. Helminth Parasites and Primates. Primates. 1963 Mar;4(1):1–96.
90. Myers BJ, Kuntz RE. A checklist of parasites reported for the baboon. Primates. 1965 Oct;6(2):137–94.
91. Appleton CC, Henzi SP, Whiten A, Byrne R. The gastrointestinal parasites ofPapio ursinus from the Drakensberg Mountains, Republic of South Africa. Int J Primatol. 1986 Oct;7(5):449–56.
92. Myers P. Nematoda (roundworms) [Internet]. Animal Diversity Web. [cited 2020 Oct 3]. Available from: https://animaldiversity.org/accounts/Nematoda/
93. Brusca RC, Brusca GJ. Invertebrates. [Internet]. Vol. 53. Sunderland, Massachusetts: Sinauer Associates; 2003 [cited 2021 Mar 11]. Available from: https://doi.org/10.1080/10635150490472968
94. Bethony J, Brooker S, Albonico M, Geiger SM, Loukas A, Diemert D, Hotez PJ. Soil-transmitted helminth infections: ascariasis, trichuriasis, and hookworm. The Lancet. 2006 May;367(9521):1521–32.
95. Polderman AM, Blotkamp J. Oesophagostomum infections in humans. Parasitol Today. 1995 Dec;11(12):451–6.
96. Nissen S, Al-Jubury A, Hansen TVA, Olsen A, Christensen H, Thamsborg SM, Nejsum P. Genetic analysis of Trichuris suis and Trichuris trichiura recovered from humans and pigs in a sympatric setting in Uganda. Vet Parasitol. 2012 Aug 13;188(1–2):68–77.
97. Cutillas C, Rocio C, Rojas M, Tewes B, Ubeda JM, Ariza C, Guevara D. Trichuris suis and Trichuris trichiura are different nematode species. Acta tropica. 2009 Jun 1;111:299–307.
98. Rivero J, García-Sánchez ÁM, Zurita A, Cutillas C, Callejón R. Trichuris trichiura isolated from Macaca sylvanus: morphological, biometrical, and molecular study. BMC Veterinary Research. 2020 Nov 17;16(1):445.
99. García-Sánchez ÁM, Callejón R, Zurita A, Reguera-Gomez M, Valero MA, Cutillas C, Rivero J. Differentiation of *Trichuris* species using a morphometric approach. Int J Parasitol Parasites Wildl. 2019 May 31;9:218–23.
100. Callejón R, Halajian A, Cutillas C. Description of a new species, Trichuris ursinus n. sp. (Nematoda: Trichuridae) from Papio ursinus Keer, 1792 from South Africa. Infection, Genetics and Evolution. 2017 Jul;51:182–93.
101. Hawash MBF, Andersen LO, Gasser RB, Stensvold CR, Nejsum P. Mitochondrial Genome Analyses Suggest Multiple Trichuris Species in Humans, Baboons, and Pigs from Different Geographical Regions. PLoS Negl Trop Dis. 2015;9(9):e0004059.
102. Rivero J, Cutillas C, Callejón R. Trichuris trichiura (Linnaeus, 1771) From Human and Non-human Primates: Morphology, Biometry, Host Specificity, Molecular Characterization, and Phylogeny. Front Vet Sci. 2021 Feb 9;7:626120.
103. Parija SC, Chidambaram M, Mandal J. Epidemiology and clinical features of soil-transmitted helminths. Trop Parasitol. 2017 Dec;7(2):81–5.
104. Speich B, Ame SM, Ali SM, Alles R, Huwyler J, Hattendorf J, Utzinger J, Albonico M, Keiser J. Oxantel pamoate-albendazole for Trichuris trichiura infection. N Engl J Med. 2014 Feb 13;370(7):610–20.
105. Nejsum P, Andersen KL, Andersen SD, Thamsborg SM, Tejedor AM. Mebendazole treatment persistently alters the size profile and morphology of Trichuris trichiura eggs. Acta Trop. 2020 Apr;204:105347.
106. Thamsborg SM, Ketzis J, Horii Y, Matthews JB. Strongyloides spp. infections of veterinary importance. Parasitology. 2017 Mar;144(3):274–84.
107. Olsen A, van Lieshout L, Marti H, Polderman T, Polman K, Steinmann P, Stothard R, Thybo S, Verweij JJ, Magnussen P. Strongyloidiasis – the most neglected of the neglected tropical diseases? Transactions of the Royal Society of Tropical Medicine and Hygiene. 2009 Oct 1;103(10):967–72.
108. Pampiglione S, Ricciardi ML. The presence of Strongyloides fülleborni von Linstow, 1905, in man in Central and East Africa. Parassitologia. 1971 Aug;13(1):257–69.

109. Nutman TB. Human infection with *Strongyloides stercoralis* and other related Strongyloides species. Parasitology. 2017 Mar;144(3):263–73.
110. Ashford RW, Barnish G, Viney ME. Strongyloides fuelleborni kellyi: infection and disease in Papua New Guinea. Parasitol Today. 1992 Sep;8(9):314–8.
111. Page W, Judd JA, Bradbury RS. The Unique Life Cycle of Strongyloides stercoralis and Implications for Public Health Action. Tropical Medicine and Infectious Disease. 2018 Jun;3(2):53.
112. Machida M, Araki J, Koyama T, Kumada M, Horii Y, Imada I, Takasaka M, Honjo S, Matsubayashi K, Tiba T. The life cycle of Streptopharagus pigmentatus (Nematoda, Spiruroidea) from the Japanese monkey. Bull Natn Sci Mus Ser A (Zool). 1978 Jan 1;4:1–9.
113. Baylis HA. On the nematode genus Streptopharagus, with some remarks on the genus Spirocerca. Transactions of the Royal Society of Tropical Medicine and Hygiene. 1923 Feb 15;16(8):486–97.
114. Ortlepp RJ. A Review of the Members of the Genus Streptopharagus Blanc, 1912. Journal of Helminthology. 1925 Dec;3(5):209–16.
115. Chitwood MB. Systema Helminthum. vol. 3, The Nematodes of Vertebrates. Satyu Yamaguti. Interscience (Wiley), New York, 1962. 1261 pp. Illus. $90. Science. 1963 Sep 6;141(3584):897–897.
116. Chardome M, Peel E. Note complémentaire sur des filarides de chimpanzés, Pan panicus et Pan satyrus au Congo Belge. Ann Soc Belg Med Trop. 1947;
117. Kreis HA. Beiträge zur Kenntnis parasitischer Nematoden. IX. Zentralblatt für Bakteriologie Abt 1. 1940;9.
118. Soulsby EJL. Textbook of Veterinary Clinical Parasitology. F. A. Davis; 1965. 1160 p.
119. Quentin J-C. Cycle biologique de Pterygodermatites desportesi (Chabaud et Rousselot, 1956) Nematoda Rictulariidae. Ann Parasitol Hum Comp. 1969;44(1):47–58.
120. Sakakibara I. Naturally occurring diseases in cynomolgus monkeys. Jpn J Med Sci Biol. 1981 Aug;34(4):263–7.
121. GUTIERREZ Y. Other tissue nematode infections. Tropical Infectious Diseases : Principles, Pathogens, and Practice. 2006;1240–1.
122. Pettifer HL. The helminth fauna of the digestive tracts of chacma baboons, Papio ursinus, from different localities in the Transvaal. Onderstepoort J Vet Res. 1984 Sep;51(3):161–70.
123. Ghai RR, Chapman CA, Omeja PA, Davies TJ, Goldberg TL. Nodule Worm Infection in Humans and Wild Primates in Uganda: Cryptic Species in a Newly Identified Region of Human Transmission. Jex AR, editor. PLoS Negl Trop Dis. 2014 Jan 9;8(1):e2641.
124. Cibot M, Guillot J, Lafosse S, Bon C, Seguya A, Krief S. Nodular Worm Infections in Wild Non-human Primates and Humans Living in the Sebitoli Area (Kibale National Park, Uganda): Do High Spatial Proximity Favor Zoonotic Transmission? Davies SJ, editor. PLoS Negl Trop Dis. 2015 Oct 9;9(10):e0004133.
125. John DT, Petri WA. Markell and Voge's Medical Parasitology - E-Book. Elsevier Health Sciences; 2013. 487 p.
126. Hotez P, Bethony J, Bottazzi M, Brooker S, Buss P. Hookworm: "The Great Infection of Mankind." PLoS medicine. 2005 Apr 1;2:e67.
127. Steenhard NR, Storey PA, Yelifari L, Pit DSS, Nansen P, Polderman AM. The role of pigs as transport hosts of the human helminths Oesophagostomum bifurcum and Necator americanus. Acta Tropica. 2000 Sep 18;76(2):125–30.
128. STRICKLAND T. Hunter's Tropical Medicine and Emerging Infectious Diseases. Revista do Instituto de Medicina Tropical de São Paulo. 2001 Apr 1;43.
129. Larval cultures and "Strongyle" type eggs [Internet]. WormBoss. 2020 [cited 2020 Mar 19]. Available from: http://www.wormboss.com.au/sheep-goats/news/articles/tests/larval-cultures-and-strongyle-type-eggs.php
130. Blotkamp J, Krepel HP, Kumar V, Baeta S, Noordende JMV, Polderman AM. Observations on the morphology of adults and larval stages of *Oesophagostomum* sp. isolated from man in northern Togo and Ghana. J Helminthol. 1993 Mar;67(1):49–61.
131. Hotez PJ, Brooker S, Bethony JM, Bottazzi ME, Loukas A, Xiao S. Hookworm Infection. N Engl J Med. 2004 Aug 19;351(8):799–807.
132. Terio KA, Lonsdorf EV, Kinsel MJ, Raphael J, Lipende I, Collins A, Li Y, Hahn BH, Travis DA, Gillespie TR. Oesophagostomiasis in non-human primates of Gombe National Park, Tanzania: Oesophagostomiasis in Gombe Primates. Am J Primatol. 2018 Jan;80(1):e22572.
133. Chow AL. Trichostrongyliasis [Internet]. ParaSites2005, Stanford University. [cited 2020 Apr 3]. Available from: https://web.stanford.edu/group/parasites/ParaSites2005/Trichostrongyliasis/index.htm
134. Krief S, Jamart A, Mahé S, Leendertz FH, Mätz-Rensing K, Crespeau F, Bain O, Guillot J. Clinical and pathologic manifestation of oesophagostomosis in African great apes: does self-medication in wild apes influence disease progression? Journal of Medical Primatology. 2008 Aug;37(4):188–95.
135. Huffman MA, Gotoh S, Turner LA, Hamai M, Yoshida K. Seasonal trends in intestinal nematode infection and medicinal plant use among chimpanzees in the Mahale Mountains, Tanzania. Primates. 1997 Apr 1;38(2):111–25.
136. Storey PA, Faile G, Hewitt E, Yelifari L, Polderman AM, Magnussen P. Clinical epidemiology and classification of human oesophagostomiasis. Trans R Soc Trop Med Hyg. 2000 Apr;94(2):177–82.

137. Ziem JB, Spannbrucker N, Magnussen P, Olsen A, Amon-Kotey DN, Frenzel K, Nang-Beifubah A, Westendorp RGJ, Polderman AM. Oesophagostomum bifurcum-induced nodular pathology in a highly endemic area of Northern Ghana. Trans R Soc Trop Med Hyg. 2005 Jun;99(6):417–22.

138. Pineda N, Yang E. Hookworm: Ancylostoma duodenale and Necator americanus [Internet]. ParaSites2009, Stanford University. [cited 2020 Mar 4]. Available from: https://web.stanford.edu/group/parasites/ParaSites2009/PinedaANDYang_Hookworm/PinedaANDYang_Hookworm.htm

139. Lieshout L, Gruijter JM, Adu-Nsiah M, Haizel M, Verweij JJ, Brienen EAT, Gasser RB, Polderman AM. Oesophagostomum bifurcum in non-human primates is not a potential reservoir for human infection in Ghana. Trop Med Int Health. 2005 Dec;10(12):1315–20.

140. Taylor, Louise. OIE Manual of Diagnostic Tests and Vaccins for Terrestrial Animals. Office Internationale des Epizooties, Paris, France; 2014.

141. Putt SNH, Shaw APM, Woods AJ, Tyler L, James AD. Veterinary epidemiology and economics in Africa: A manual for use in the design and appraisal of livestock health policy. Vol. No.3. Berkshire, England: ILCA; 1988.

142. Euzeby J. Diagnostic expérimental des helminthoses animales (Animaux domestiques, Animaux de laboratoire, Primates); Travaux pratiques d'helminthologie vétérinaire : Livre 1 généralités, diagnostic ante-mortem. Paris, France: Ministère de l'Agriculture, édition Informations techniques des services vétérinaires; 1981. 349 p.

143. Cringoli G, Maurelli MP, Levecke B, Bosco A, Vercruysse J, Utzinger J, Rinaldi L. The Mini-FLOTAC technique for the diagnosis of helminth and protozoan infections in humans and animals. Nat Protoc. 2017 Sep;12(9):1723–32.

144. Pereira Lima J, Delgado PG. Diagnosis of strongyloidiasis: Importance of Baermann's method. Digest Dis Sci. 1961 Sep;6(9):899–904.

145. Garcia LS, Arrowood M, Kokoskin E, Paltridge GP, Pillai DR, Procop GW, Ryan N, Shimizu RY, Visvesvara G. Practical Guidance for Clinical Microbiology Laboratories: Laboratory Diagnosis of Parasites from the Gastrointestinal Tract. Clin Microbiol Reviews. 2018 Jan;31(1):e00025-17, e00025-17.

146. Dupuy AR. Le Niokolo-Koba premier grand parc national de la république du Sénégal. Dakar: GIA; 1971. 274 p.

147. Verschuren J. Ecologie du Parc National du Niokolo-Koba (Sénégal). Grands mammifères et remarques sur la conservation. Bulletin Institut Royal Sciences Naturelles de Belgique - Biologie 55. 1983;

148. Vincke X, Hornick J-L, Njikam NI, Leroy P. Gestion de la faune sauvage au Sénégal: comparaison du Parc national du Niokolo Koba et de la Réserve privée de Bandia. Ann Méd Vét. 2005;(149).

149. Rabeil T. Inventaire de la grande faune du Parc National du Niokolo Koba au Sénégal. 2015;39.

150. Mcgrew W, Baldwin P, Marchant L, Pruetz J, Tutin C. Chimpanzees (Pan troglodytes verus) and their mammalian sympatriates: Mt. Assirik, Niokolo-Koba National Park, Senegal. Primates. 2014 Oct 1;55.

151. Benoit M. Le Parc National du Niokolo Koba : livre blanc. DPN; 1993. 52 p.

152. Jessee MT, Schilling PW, Stunkard JA. Identification of intestinal helminth eggs in Old World primates. Laboratory Animal Care [Internet]. 1970 [cited 2020 Jul 4];20(1). Available from: https://www.cabdirect.org/cabdirect/abstract/19710803555

153. Ash L, Orihel TC. Ash & Orihel's atlas of human parasitology. 5th ed. 2007.

154. Barratt JLN, Lane M, Talundzic E, Richins T, Robertson G, Formenti F, Pritt B, Verocai G, Nascimento de Souza J, Mato Soares N, Traub R, Buonfrate D, Bradbury RS. A global genotyping survey of Strongyloides stercoralis and Strongyloides fuelleborni using deep amplicon sequencing. Periago MV, editor. PLoS Negl Trop Dis. 2019 Sep 16;13(9):e0007609.

155. Kearse M, Moir R, Wilson A, Stones-Havas S, Cheung M, Sturrock S, Buxton S, Cooper A, Markowitz S, Duran C, Thierer T, Ashton B, Meintjes P, Drummond A. Geneious Basic: An integrated and extendable desktop software platform for the organization and analysis of sequence data. Bioinformatics. 2012 Jun 15;28(12):1647–9.

156. Petrzelkova KJ, Hasegawa H, Moscovice LR, Kaur T, Issa M, Huffman MA. Parasitic Nematodes in the Chimpanzee Population on Rubondo Island, Tanzania. Int J Primatol. 2006 Jun;27(3):767–77.

157. Koga K, Kasuya S, Khamboonruang C, Sukhavat K, Ieda M, Takatsuka N, Kita K, Ohtomo H. A Modified Agar Plate Method for Detection of Strongyloides Stercoralis. The American Journal of Tropical Medicine and Hygiene. 1991 Oct 1;45(4):518–21.

158. Gesquiere LR, Habig B, Hansen C, Li A, Freid K, Learn NH, Alberts SC, Graham AL, Archie EA. Noninvasive measurement of mucosal immunity in a free-ranging baboon population. Am J Primatol [Internet]. 2020 Feb [cited 2021 Mar 5];82(2). Available from: https://onlinelibrary.wiley.com/doi/abs/10.1002/ajp.23093

159. Gómez A. Prevalence of Entamoeba histolytica and Entamoeba dispar in a Highly Endemic Rural Population. Archives of Medical Research [Internet]. [cited 2021 Apr 10]; Available from: https://www.academia.edu/6063274/Prevalence_of_Entamoeba_histolytica_and_Entamoeba_dispar_in_a_Highly_Endemic_Rural_Population

160. Baker DG. Parasites of non-human primates. In Flynn's Parasites of Laboratory Animals. 2nd ed. Ames, IA, USA: Blackwell Publishing; 2008.

161. Behnke JM, Clercq DD, Sacko M, Gilbert FS, Ouattara DB, Vercruysse J. The epidemiology of human hookworm infections in the southern region of Mali. Tropical Medicine & International Health. 2000;5(5):343–54.

162. Appleton CC, Maurihungirire M, Gouws E. The distribution of helminth infections along the coastal plain of Kwazulu-Natal province, South Africa. Annals of Tropical Medicine and Parasitology. 1999 Dec 1;93(8):859–68.
163. Ocaido M, Dranzoa C, Cheli P. Gastrointestinal parasites of baboons (*Papio anubis*) interacting with humans in West Bugwe Forest Reserve, Uganda: *Gastrointestinal parasites of baboons*. African Journal of Ecology. 2003 Dec;41(4):356–9.
164. Gillespie TR, Nunn CL, Leendertz FH. Integrative approaches to the study of primate infectious disease: Implications for biodiversity conservation and global health. Am J Phys Anthropol. 2008;137(S47):53–69.
165. Práca O, Tehlárová Z. Rozmanitosť gastrointestinálnych parazitov mangabejov štíhlych (Cercocebus agilis) v Bai Hokou, Stredoafrická republika. 2014;89.
166. Murray S, Goodall J. Intestinal parasites of baboons (Papio Cynocephalus Anubis) and chimpanzees (Pan Troglodytes) in Gombe National Park. :4.
167. Obanda V, Maingi N, Muchemi G, Ng'ang'a CJ, Angelone S, Archie EA. Infection dynamics of gastrointestinal helminths in sympatric non-human primates, livestock and wild ruminants in Kenya. Serrano E, editor. PLoS ONE. 2019 Jun 10;14(6):e0217929.
168. Vitone N, Altizer S, Nunn C. Body size, diet and sociality influence the species richness of parasitic worms in anthropoid primates. Evol Ecol Res. 2003 Nov 30;6.
169. Mul IF, Paembonan W, Singleton I, Wich SA, van Bolhuis HG. Intestinal Parasites of Free-ranging, Semicaptive, and Captive Pongo abelii in Sumatra, Indonesia. Int J Primatol. 2007 Apr;28(2):407–20.
170. Labes EM, Hegglin D, Grimm F, Nurcahyo W, Harrison ME, Bastian ML, Deplazes P. Intestinal parasites of endangered orangutans *(Pongo pygmaeus*) in Central and East Kalimantan, Borneo, Indonesia. Parasitology. 2010 Jan;137(1):123–35.
171. Pafčo B, Benavides JA, Pšenková-Profousová I, Modrý D, Červená B, Shutt KA, Hasegawa H, Fuh T, Todd AF, Petrželková KJ. Do habituation, host traits and seasonality have an impact on protist and helminth infections of wild western lowland gorillas? Parasitol Res. 2017 Dec;116(12):3401–10.
172. Post DG, Hausfater G, McCuskey SA. Feeding behavior of yellow baboons (Papio cynocephalus): relationship to age, gender and dominance rank. Folia Primatol (Basel). 1980;34(3–4):170–95.
173. Lýsek H. A scanning electron microscope study of the effect of an ovicidal fungus on the eggs of Ascaris lumbricoides. Parasitology. 1978 Oct;77(2):139–41.
174. Kalousová B, Piel AK, Pomajbíková K, Modrý D, Stewart FA, Petrželková KJ. Gastrointestinal Parasites of Savanna Chimpanzees (Pan troglodytes schweinfurthii) in Ugalla, Tanzania. Int J Primatol. 2014 Apr;35(2):463–75.
175. Schneider-Crease I, Beehner JC, Bergman TJ, Gomery MA, Koklic L, Lu A, Snyder-Mackler N. Ecology eclipses phylogeny as a major driver of nematode parasite community structure in a graminivorous primate. Funct Ecol. 2020 Sep;34(9):1898–906.
176. Mbora DNM, McPeek MA. Host density and human activities mediate increased parasite prevalence and richness in primates threatened by habitat loss and fragmentation. J Anim Ecol. 2009 Jan;78(1):210–8.
177. Arneberg P. Host population density and body mass as determinants of species richness in parasite communities: comparative analyses of directly transmitted nematodes of mammals. Ecography. 2002;25(1):88–94.
178. Barelli C, Pafčo B, Manica M, Rovero F, Rosà R, Modrý D, Hauffe HC. Loss of protozoan and metazoan intestinal symbiont biodiversity in wild primates living in unprotected forests. Sci Rep. 2020 Jul 2;10(1):10917.
179. Lambert KA, Pathak AK, Cattadori IM. Does host immunity influence helminth egg hatchability in the environment? J Helminthol. 2015 Jul;89(4):446–52.
180. Girgis NM, Gundra UM, Loke P. Immune Regulation during Helminth Infections. PLoS Pathog [Internet]. 2013 Apr 18 [cited 2021 Feb 8];9(4). Available from: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3630086/
181. Anargyrou K, Paterakis G, Boutsis D, Politou M, Papadhimitriou SI, Siakandaris M, Vassiliadis J, Androulakis A, Meletis J, Rombos J, Tassiopoulou A, Vaiopoulos G. An unusual case of CD4+ CD7+ CD56+ acute leukemia with overlapping features of type 2 dendritic cell (DC2) and myeloid/NK cell precursor acute leukemia. Eur J Haematol. 2003 Oct;71(4):294–8.
182. Cho H-S, Shin S-S, Park N-Y. Balantidiasis in the gastric lymph nodes of Barbary sheep (Ammotragus lervia): an incidental finding. J Vet Sci. 2006 Jun;7(2):207–9.
183. Mignatti A, Boag B, Cattadori IM. Host immunity shapes the impact of climate changes on the dynamics of parasite infections. Proc Natl Acad Sci USA. 2016 Mar 15;113(11):2970–5.
184. Akinyi MY, Jansen D, Habig B, Gesquiere LR, Alberts SC, Archie EA. Costs and drivers of helminth parasite infection in wild female baboons. Cattadori I, editor. J Anim Ecol. 2019 Jul;88(7):1029–43.
185. Moore SL, Wilson K. Parasites as a Viability Cost of Sexual Selection in Natural Populations of Mammals. Science. 2002 Sep 20;297(5589):2015–8.
186. Foitová I, Huffman MA, Wisnu N, Olšanský M. Parasites and their impacts on orangutan health. :15.
187. Blackwell AD, Tamayo MA, Beheim B, Trumble BC, Stieglitz J, Hooper PL, Martin M, Kaplan H, Gurven M. Helminth infection, fecundity, and age of first pregnancy in women. Science. 2015 Nov 20;350(6263):970–2.
188. Smith J, Meerovitch E. Primates as a Source of Entamoeba histolytica, Their Zymodeme Status and Zoonotic Potential. The Journal of parasitology. 1986 Jan 1;71:751–6.

189. Nagayasu E, Aung MPPTHH, Hortiwakul T, Hino A, Tanaka T, Higashiarakawa M, Olia A, Taniguchi T, Win SMT, Ohashi I, Odongo-Aginya EI, Aye KM, Mon M, Win KK, Ota K, Torisu Y, Panthuwong S, Kimura E, Palacpac NMQ, Kikuchi T, Hirata T, Torisu S, Hisaeda H, Horii T, Fujita J, Htike WW, Maruyama H. A possible origin population of pathogenic intestinal nematodes, Strongyloides stercoralis, unveiled by molecular phylogeny. Sci Rep. 2017 Dec;7(1):4844.
190. Sanpool O, Intapan PM, Rodpai R, Laoraksawong P, Sadaow L, Tourtip S, Piratae S, Maleewong W, Thanchomnang T. Dogs are reservoir hosts for possible transmission of human strongyloidiasis in Thailand: molecular identification and genetic diversity of causative parasite species. J Helminthol. 2020;94:e110.
191. Polderman AM, De Gruijter JM, Verweij JJ, Ziem J, Gasser RB. Genetic substructuring within the Oesiphagostomum bifurcum (Nematoda) from human and non-human primates from Ghana based on random amplified polymorphic DNA analysis. The American Journal of Tropical Medicine and Hygiene. 2004 Aug 1;71(2):227–33.
192. Wenz-Mücke A, Sithithaworn P, Petney T, Taraschewski H. Human contact influences the foraging behaviour and parasite community in long-tailed macaques. Parasitology. 2013 Jan 30;140:1–10.
193. Hasegawa H, Sato H, Fujita S, Nguema PPM, Nobusue K, Miyagi K, Kooriyama T, Takenoshita Y, Noda S, Sato A, Morimoto A, Ikeda Y, Nishida T. Molecular identification of the causative agent of human strongyloidiasis acquired in Tanzania: Dispersal and diversity of Strongyloides spp. and their hosts. Parasitology International. 2010 Sep;59(3):407–13.
194. Faust EC, Kagy ES. Experimental Studies on Human and Primate Species of Strongyloides: I. The Variability and Instability of Types. The American Journal of Tropical Medicine and Hygiene. 1933 Jan 1;s1-13(1):47–65.
195. Takano Y, Minakami K, Kodama S, Matsuo T, Satozono I. Cross infection of <I>Strongyloides</I> between humans and dogs in the Amami Islands, Japan. Tropical Medicine and Health. 2009;advpub:0911110110–0911110110.
196. Schär F, Guo L, Streit A, Khieu V, Muth S, Marti H, Odermatt P. Strongyloides stercoralis genotypes in humans in Cambodia. Parasitol Int. 2014 Jun;63(3):533–6.
197. Harhay MO, Horton J, Olliaro PL. Epidemiology and control of human gastrointestinal parasites in children. Expert Review of Anti-infective Therapy. 2010 Feb;8(2):219–34.
198. Legesse M, Erko B. Zoonotic intestinal parasites in Papio anubis (baboon) and Cercopithecus aethiops (vervet) from four localities in Ethiopia. Acta Tropica. 2004 May;90(3):231–6.
199. Hoffmann K. Parasites in the classroom: The effects of intestinal helminths on education attainment and the value of school deworming programs [Internet]. ParaSites2010, Stanford University. 2010 [cited 2021 Feb 8]. Available from: https://web.stanford.edu/group/parasites/ParaSites2010/Katherine_Hoffmann/Hoffmann%20-%20ParaSites%20Webpage.htm
200. Ruebush M. Why dirt is good : 5 ways to make germs your friends [Internet]. New York : Kaplan Pub.; 2009 [cited 2021 Feb 11]. 218 p. Available from: http://archive.org/details/whydirtisgood5wa0000rueb
201. Cooper PJ. Intestinal worms and human allergy. Parasite Immunology. 2004;26(11–12):455–67.
202. Asojo OA, Goud G, Dhar K, Loukas A, Zhan B, Deumic V, Liu S, Borgstahl GEO, Hotez PJ. X-ray Structure of Na-ASP-2, a Pathogenesis-related-1 Protein from the Nematode Parasite, Necator americanus, and a Vaccine Antigen for Human Hookworm Infection. Journal of Molecular Biology. 2005 Feb;346(3):801–14.
203. Helminthic therapy and nutritional deficiencies [Internet]. Helminthic Therapy wiki. [cited 2021 Feb 8]. Available from: https://helminthictherapywiki.org/wiki/index.php/Helminthic_therapy_and_nutritional_deficiencies
204. Elliott A, Kizza M, Quigley M, Ndibazza J, Nampijja M, Muhangi L, Morison L, Namujju P, Muwanga M, Kabatereine N, Whitworth J. The impact of helminths on the response to immunization and on the incidence of infection and disease in childhood in Uganda: design of a randomized, double-blind, placebo-controlled, factorial trial of deworming interventions delivered in pregnancy and early childhood [ISRCTN32849447. Clinical trials (London, England). 2007 Feb 1;4:42–57.
205. Wolff MJ, Broadhurst MJ, Loke P. Helminthic therapy: improving mucosal barrier function. Trends Parasitol. 2012 May;28(5):187–94.
206. Jouvin M-H, Kinet J-P. Trichuris suis ova: testing a helminth-based therapy as an extension of the hygiene hypothesis. J Allergy Clin Immunol. 2012 Jul;130(1):3–10; quiz 11–2.
207. Harnett W, Harnett MM. Helminth-derived immunomodulators: can understanding the worm produce the pill? Nat Rev Immunol. 2010 Apr;10(4):278–84.
208. St. Georgiev V. Parasitic infections. Treatement and developmental therapeutics. 1. Necatoriasis. In: Current Pharmaceutical Design. Bentham Science Publishers; 1999. p. 545–54.
209. Necator americanus [Internet]. MSDSonline. [cited 2021 Feb 11]. Available from: https://www.msdsonline.com/resources/sds-resources/free-safety-data-sheet-index/necator-americanus/
210. MacIntosh AJJ, Jacobs A, Garcia C, Shimizu K, Mouri K, Huffman MA, Hernandez AD. Monkeys in the Middle: Parasite Transmission through the Social Network of a Wild Primate. Korb J, editor. PLoS ONE. 2012 Dec 5;7(12):e51144.
211. Rimbach R, Bisanzio D, Galvis N, Link A, Di Fiore A, Gillespie TR. Brown spider monkeys (*Ateles hybridus*): a model for differentiating the role of social networks and physical contact on parasite transmission dynamics. Phil Trans R Soc B. 2015 May 26;370(1669):20140110.

212. Klein SL. Hormonal and immunological mechanisms mediating sex differences in parasite infection. Parasite Immunology. 2004;26(6–7):247–64.
213. Habig B, Jansen DAWAM, Akinyi MY, Gesquiere LR, Alberts SC, Archie EA. Multi-scale predictors of parasite risk in wild male savanna baboons (Papio cynocephalus). Behav Ecol Sociobiol. 2019 Oct;73(10):134.
214. VanderWaal KL, Obanda V, Omondi GP, McCowan B, Wang H, Fushing H, Isbell LA. The "strength of weak ties" and helminth parasitism in giraffe social networks. BEHECO. 2016;27(4):1190–7.
215. Hutchings MR, Kyriazakis I, Anderson DH, Gordon IJ, Coop RL. Behavioural strategies used by parasitized and non-parasitized sheep to avoid ingestion of gastro-intestinal nematodes associated with faeces. Animal Science. 1998 Aug;67:97–106.

Acknowledgements

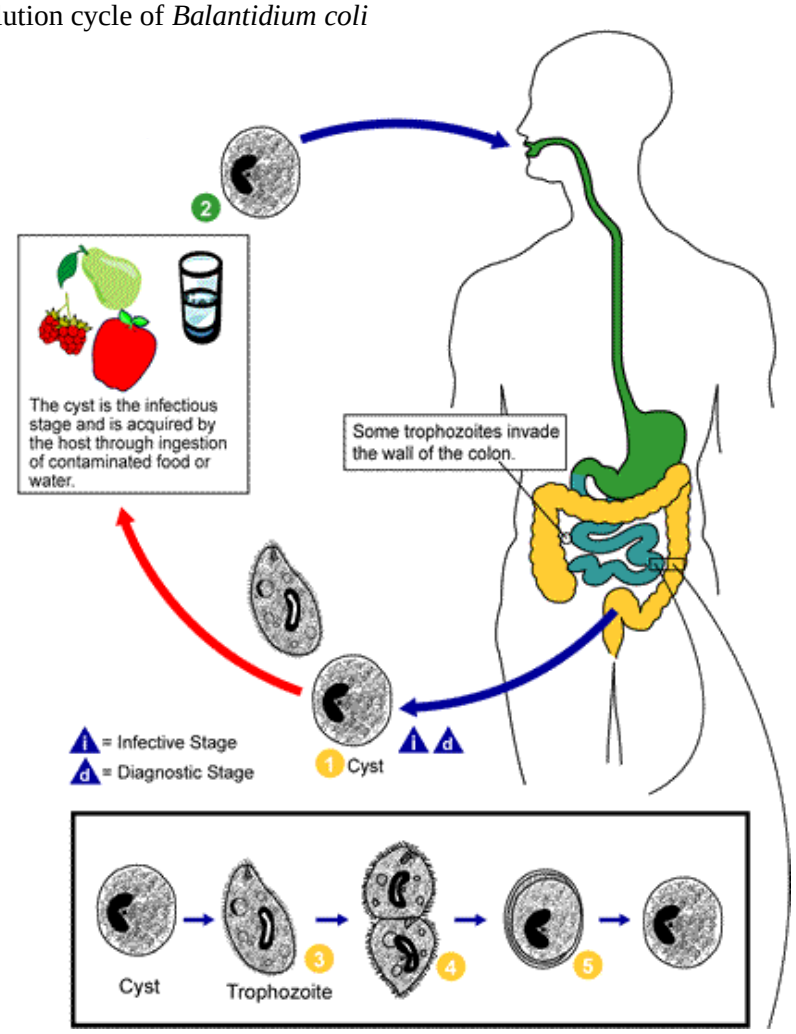
I would like to thank the Direction des Parcs Nationaux and Ministère de l'Environnement et de la Protection de la Nature de la République du Sénégal and the Conservateur for permission to work in the PNNK. I thank the park agents of the Centre de Recherche de Primatologie, Simenti, for sharing their knowledge and culture with me, especially Coli for the profound discussions and the linguistic exchanges. Thank you to Julia for giving me the opportunity to set foot in the fascinating baboon world and Dietmar for sending me many interesting articles and helpful comments on parts of my work. I also appreciate the help of my colleagues Irene, Brenda, Rachel to identify the baboons during my first weeks in the field and the help of William to stay physically and mentally healthy until the end (and for letting me cramp the fridge with fresh local fruit and vegetables)! Thank you David Modry for saving my thesis from the sanitary crisis and sharing useful knowledge about primate parasites, as well as Evca, Terka, Anna, Niki & Bara for helping me out in the lab.

Finally, I gratefully acknowledge the support from my parents Nigel and Michèle, my sister Eli, my aunt Ali, Johannes, Alizée, Moka, and all the human and non human creatures with who I've shared adventures on the road to becoming a vet !

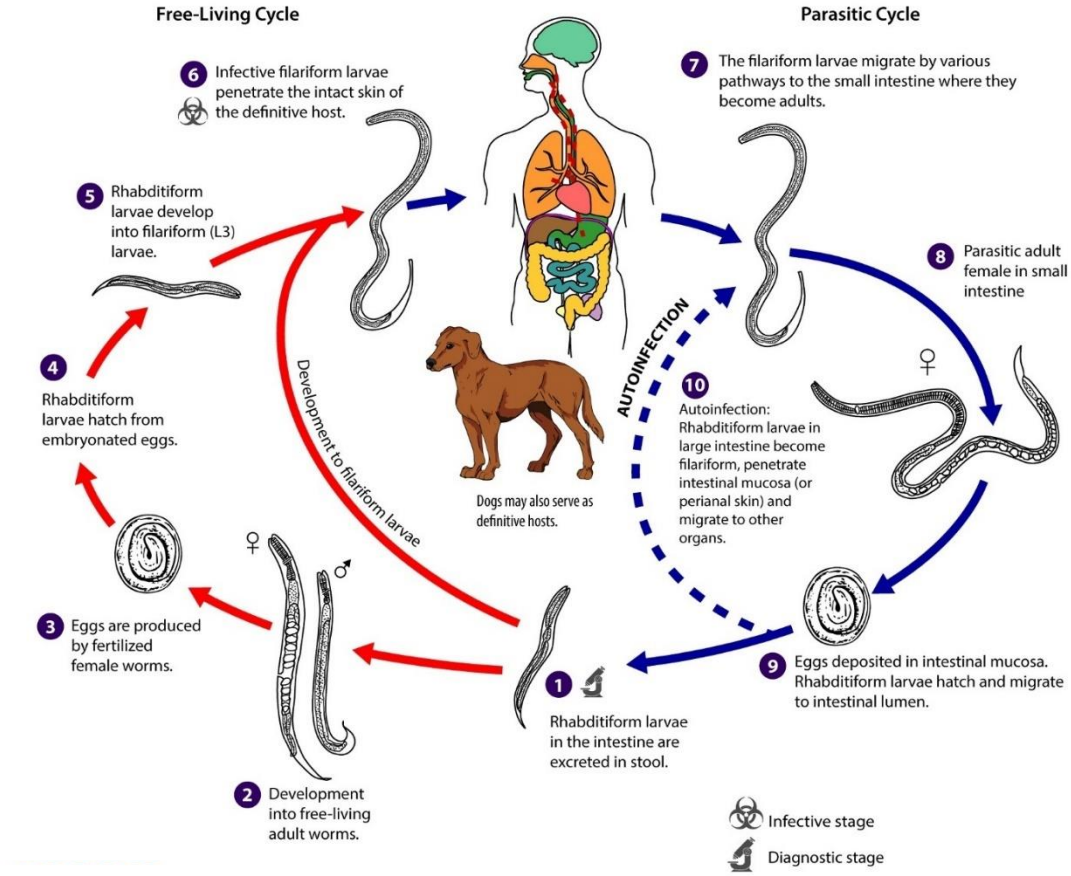
Juvenile male Guinea baboon at Simenti, PNNK *(Photo: A. Coles)*

Annexes

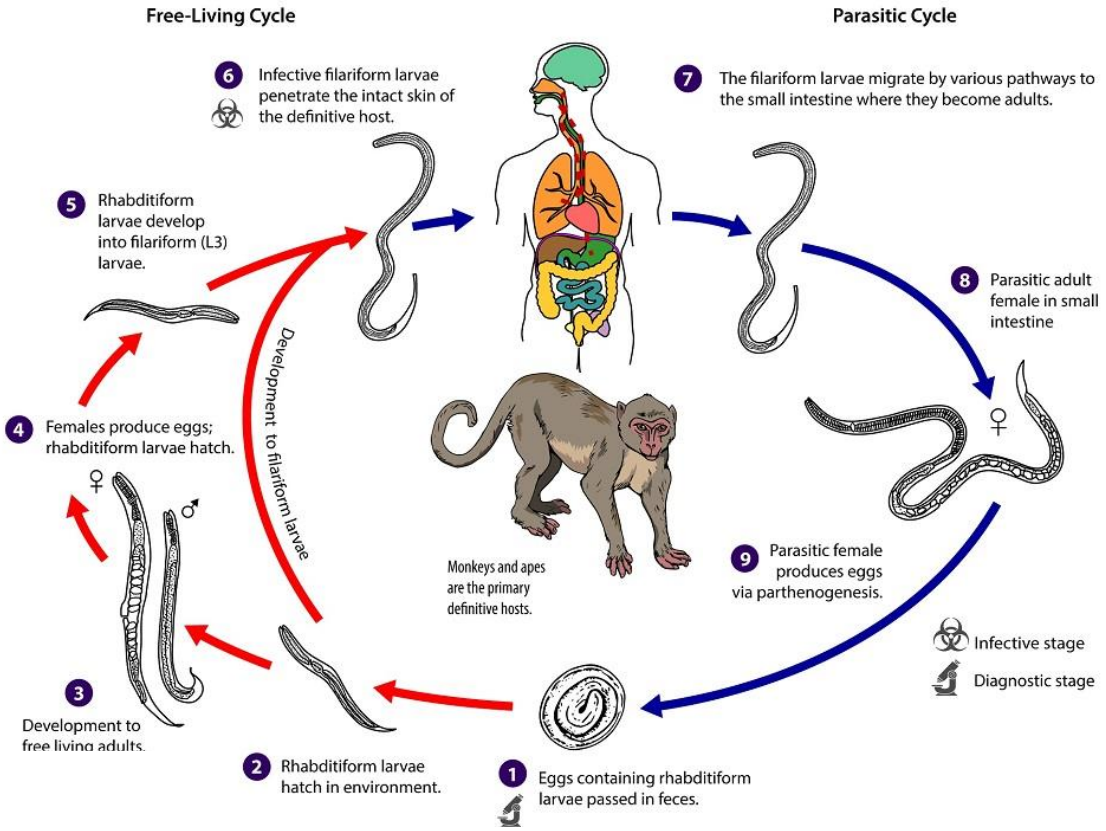
Annex 1 - Evolution cycle of *Balantidium coli*



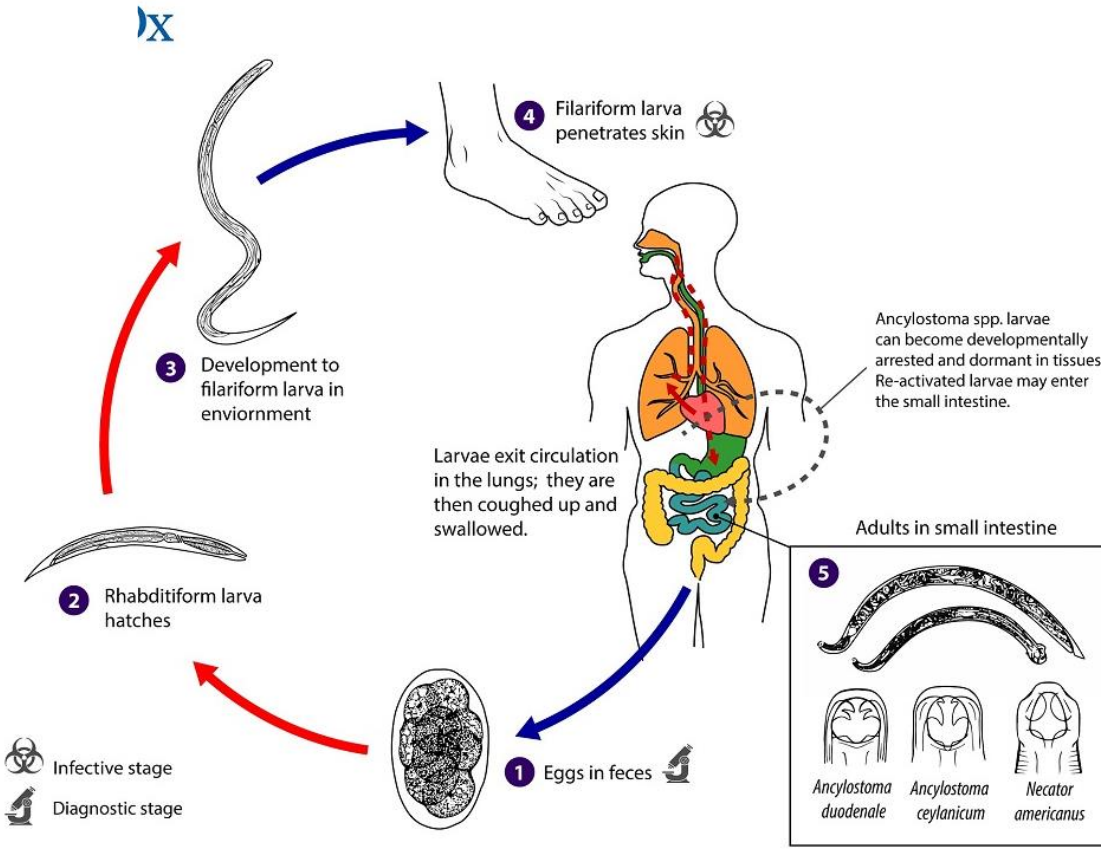
Annex 2 – Life cycle of *Strongyloides stercoralis*



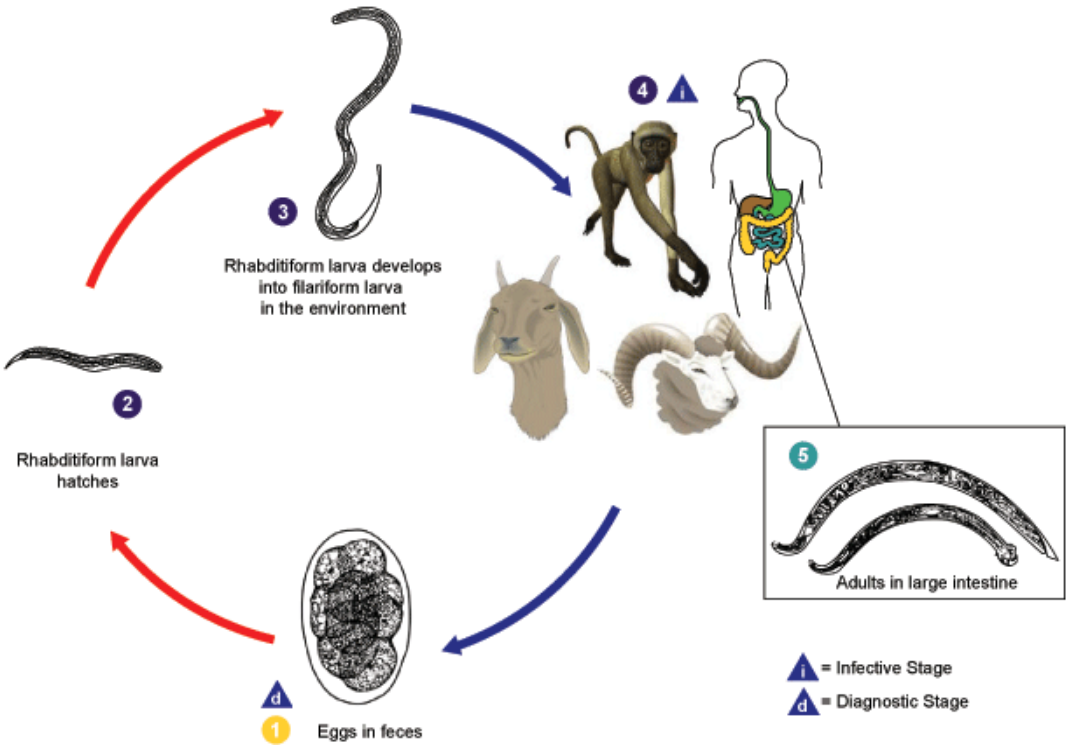
Annex 3 – Life cycle of *Strongyloides fülleborni*



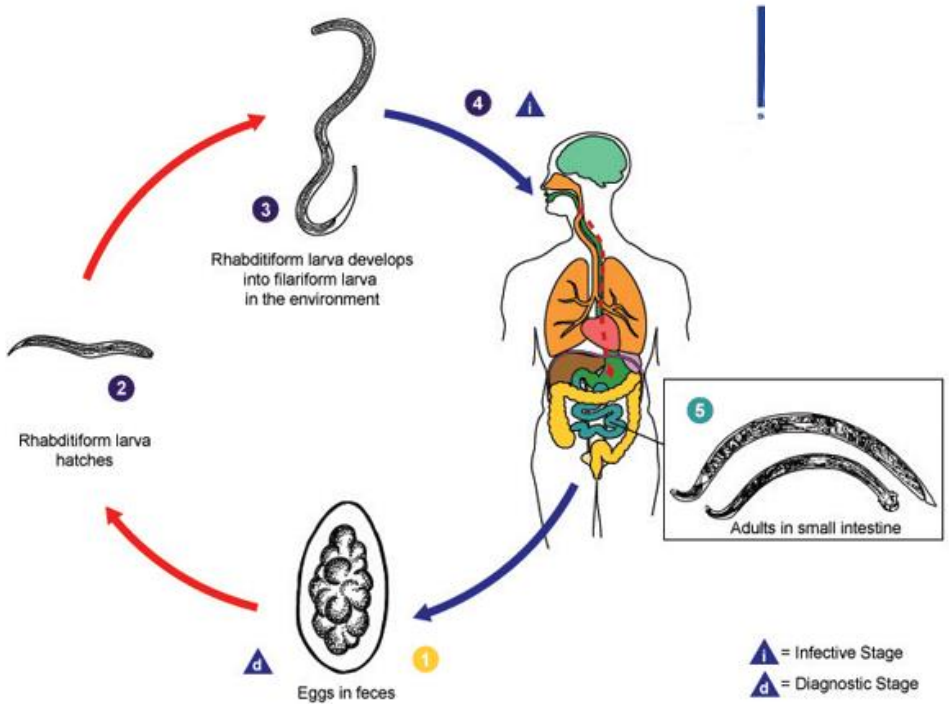
Annex 4 - Life cycle of intestinal hookworms such as *Necator americanus.* The human silhouette can be replaced by a baboon, the cycle is identical.



Annex 5 - Life cycle of intestinal hookworms such as *Oesophagostomum* species, usually completed in less than 60 days.



Annex 6 - Life cycle of intestinal hookworms such as *Trichostrongylus* species. The human silhouette can be replaced by a baboon, the cycle is identical.



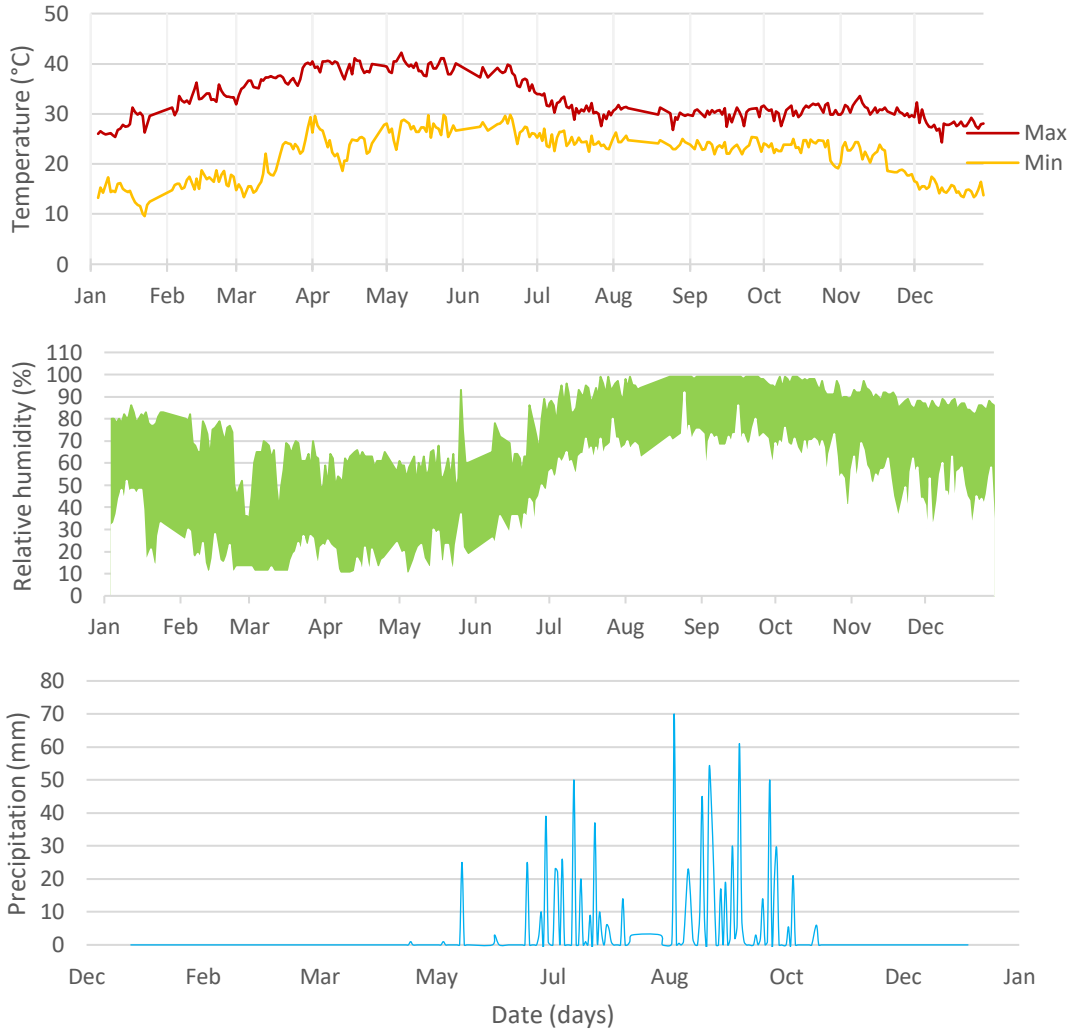
Annex 7 - Microscopic morphological differences between helminthic parasites eggs present in baboon faeces, useful as a diagnostic tool. *Modified from: Centres for Disease Control and Prevention, 2016.*

EGGS					
NEMATODES	**Size range (µm)**	**Shape**	**Colour**	**Development stage when passed**	**Details**
Enterobius vermicularis	20-32 x 50-60	Elongated, asymmetrical with one convex side and one flattened side	None	Embryonated, often with C shaped embryo	Smooth, thin eggshell. Rarely a fully developed larva is found. Best seen on tape / anal swab.
Ascaris lumbricoides	Fertile egg: 35-45 x 45-70	Round or ovoidal. Thick shell	Brown or yellowish brown	1 cell, separated from the shell at both ends	Mamillated albuminous outer shell (resembles a golf ball). Occasionally contains more cells or developed larva
	Infertile egg: 35-45 x 85-95	Elongated, sometimes triangular or kidney shaped. Very thin shell	Brown	Internal material is an irregular granulated mass	Mamillated covering is often reduced
Trichuris trichuria	20-29 x 49-65	Elongated, barrel-shaped with polar "plug" at each end	Yellow to brown. Colourless plugs.	1 cell or unsegmented	Rarely, untypical eggs may lack polar plugs
Ancylostoma duodenale / ***Necator americanus***	35-47 x 55-76	Oval or ellipsoid. Thin shell.	Colourless or with grey embryo	4-to-8 cell stage	Eggs should be reported as "hookworm" and identification needs copro-culture or molecular tools
TREMATODES					
Schistosoma mansoni	45-73 x 114-180	Elongated with lateral spine near posterior end. Anterior end tapered and slightly curved	Yellow or yellow brown	Embryonated. Contains mature miracidium	Eggs are discharged in stool at irregular intervals….
Fasciola hepatica, F. buski	63-90 x 120-150	Ellipsoidal, thin shell. Small operculum.	Yellow to light brown	Un-embryonated. Germinal cell is imbedded in yolk cells	Large size, broad ovals. *F. buski* slightly larger (by 10 µm)

Annex 8 - Microscopic morphological differences between helminthic parasites larvae present in baboon faeces, useful as a diagnostic tool. *Modified from: Centres for Disease Control and Prevention in 2016.*

Species	**L1 RHABDITIFORM LARVA**				**L3 FILIFORM LARVA**		
	Size (*µm*)	**Buccal cavity**	**Oesophagus**	**Genital primordium**	**Size (*µm*)**	**Oesophagus**	**Tail**
Strongyloides stercoralis	16-20 x 200-300	Short. 1/3-1/2 the width of anterior body end.	Bulbed. 1/3 the body length	Prominent. Elongate, tapered or pointed structure located along ventral wall.	20-24 x 500-550	No bulb. ½ the body length	Notched
Hookworm	14-17 x 200-300	Long. Approximately as long as body width.	Bulbed. 1/3 the body length	Inconspicuous. Rarely distinct. Small, nearer the tail region.	20-24 x 500-700	No bulb. ¼ the body length	Pointed

Annex 9 - Climate at Simenti field station in 2019, summary of daily measurements.
From top to bottom: Temperature (minimum and maximum), relative humidity (mininum and maximum), rainfall.



Annex 10 - Demography of main parties of habituated baboons at time of study. *F*: female, *M*: male.

	Age cat.	Old			Adults			Subadults			Juveniles			Yearlings			Total		
Gang	**Party name**	***F***	***M***	**total**	***F***	***M***	**total**	***F***	***M***	**total**	***F***	***M***	**total**	***F***	***M***	**total**	***F***	***M***	**all**
Simenti	**Party 5**	*1*	*1*	2	*9*	*4*	13				*7*	*10*	17	*1*	*1*	2	18	16	34
	Party 6I		*1*	1	*2*	*2*	4		*1*	1	*1*	*3*	4				3	7	10
	Party 6W	*2*		2	*3*	*4*	7	*4*	*1*	5	*9*	*2*	11	*1*		1	19	7	26
Mare	**Party 9**	*1*	*1*	2	*4*	*6*	10	*1*	*1*	2	*4*	*6*	10				10	14	24
	Party 9B	*1*	*2*	3	*1*	*2*	3	*1*		1					*1*	1	3	5	8
	Grand Total			**10**			**37**			**9**			**42**			**4**			**102**

I grant in-house access (namely, reading the hard copy version of the document) at the Library.

If the preparation of the document to be uploaded was supported or sponsored by a firm or an organization, you also declare that you are entitled to sign the present Agreement concerning the document.

The operators of HuVetA do not assume any legal liability or responsibility towards the author/copyright holder/organizations in case somebody uses the material legally uploaded to HuVetA in a way that is unlawful.

Date: Budapest, 30 day 03 month 2021 year

Author/copyright owner signature

HuVetA Magyar Állatorvos-tudományi Archívum – Hungarian Veterinary Archive *is an online veterinary repository operated by the Ferenc Hutÿra Library, Archives and Museum. It is an electronic knowledge base which aims to collect, organize, store documents regarding Hungarian veterinary science and history, and make them searchable and accessible in line with current legal requirements and regulations.*

HuVetA relies on the latest technology in order to provide easy searchability (by search engines, as well) and access to the full text document, whenever possible.
Based on the above, HuVetA aims to:

- *increase awareness of Hungarian veterinary science not only in Hungary, but also internationally;*
- *increase citation numbers of publications authored by Hungarian veterinarians, thus improve the impact factor of Hungarian veterinary journals;*
- *present the knowledge base of the University of Veterinary Medicine Budapest and its partners in a focussed way in order to improve the prestige of the Hungarian veterinary profession, and the competitiveness of the organizations in question;*
- *facilitate professional relations and collaboration;*
- *support open access.*

Appendix 6. Electronic License Agreement and Copyright Declaration

HuVetA

ELECTRONIC LICENSE AGREEMENT AND COPYRIGHT DECLARATION*

Name: COLES Alexia Ivy

Contact information (e-mail): a.coles@outlook.fr

Title of document (to be uploaded): Diversity of gastro-intestinal parasites of wild Guinea Baboons in Senegal

Publication data of document: 2021

Number of files submitted: 1

By accepting the present agreement the author or copyright owner grants non-exclusive license to HuVetA over the above mentioned document (including its abstract) to be converted to copy protected PDF format without changing its content, in order to archive, reproduce, and make accessible under the conditions specified below.

The author agrees that HuVetA may store more than one copy (accessible only to HuVetA administrators) of the licensed document exclusively for purposes of secure storage and backup, if necessary.

You state that the submission is your original work, and that you have the right to grant the rights contained in this license. You also state that your submission does not, to the best of your knowledge, infringe upon anyone's copyright. If the document has parts which you are not the copyright owner of, you have to indicate that you have obtained unrestricted permission from the copyright owner to grant the rights required by this Agreement, and that any such third-party owned material is clearly identified and acknowledged within the text of the licensed document.

The copyright owner defines the scope of access to the document stored in HuVetA as follows **(mark the appropriate box with an X)**:

- [ ] I grant unlimited online access,
- [ ] I grant access only through the intranet (IP range) of the University of Veterinary Medicine,
- [ ] I grant access only on one dedicated computer at the Ferenc Hutÿra Library,
- [x] I grant unlimited online access only to the bibliographic data and abstract of the document.

Please, define the **in-house accessibility of the document** by marking the below box with an **X**:

- [x]

Appendix 6. Electronic License Agreement and Copyright Declaration

HuVetA

ELECTRONIC LICENSE AGREEMENT AND COPYRIGHT DECLARATION*

Name: COLES Alexia Ivy

Contact information (e-mail): a.coles@outlook.fr

Title of document (to be uploaded): Diversity of gastro-intestinal parasites of wild Guinea Baboons in Senegal

Publication data of document: 2021

Number of files submitted: 1

By accepting the present agreement the author or copyright owner grants non-exclusive license to HuVetA over the above mentioned document (including its abstract) to be converted to copy protected PDF format without changing its content, in order to archive, reproduce, and make accessible under the conditions specified below.

The author agrees that HuVetA may store more than one copy (accessible only to HuVetA administrators) of the licensed document exclusively for purposes of secure storage and backup, if necessary.

You state that the submission is your original work, and that you have the right to grant the rights contained in this license. You also state that your submission does not, to the best of your knowledge, infringe upon anyone's copyright. If the document has parts which you are not the copyright owner of, you have to indicate that you have obtained unrestricted permission from the copyright owner to grant the rights required by this Agreement, and that any such third-party owned material is clearly identified and acknowledged within the text of the licensed document.

The copyright owner defines the scope of access to the document stored in HuVetA as follows **(mark the appropriate box with an X)**:

- [ ] I grant unlimited online access,
- [ ] I grant access only through the intranet (IP range) of the University of Veterinary Medicine,
- [ ] I grant access only on one dedicated computer at the Ferenc Hutÿra Library,
- [x] I grant unlimited online access only to the bibliographic data and abstract of the document.

Please, define the **in-house accessibility of the document** by marking the below box with an **X**:

- [x]

I grant in-house access (namely, reading the hard copy version of the document) at the Library.

If the preparation of the document to be uploaded was supported or sponsored by a firm or an organization, you also declare that you are entitled to sign the present Agreement concerning the document.

The operators of HuVetA do not assume any legal liability or responsibility towards the author/copyright holder/organizations in case somebody uses the material legally uploaded to HuVetA in a way that is unlawful.

Date: Budapest, 30 day 03 month 2021 year

Author/copyright owner signature

HuVetA Magyar Állatorvos-tudományi Archívum – Hungarian Veterinary Archive *is an online veterinary repository operated by the Ferenc Hutÿra Library, Archives and Museum. It is an electronic knowledge base which aims to collect, organize, store documents regarding Hungarian veterinary science and history, and make them searchable and accessible in line with current legal requirements and regulations.*

HuVetA relies on the latest technology in order to provide easy searchability (by search engines, as well) and access to the full text document, whenever possible.
Based on the above, HuVetA aims to:

- *increase awareness of Hungarian veterinary science not only in Hungary, but also internationally;*
- *increase citation numbers of publications authored by Hungarian veterinarians, thus improve the impact factor of Hungarian veterinary journals;*
- *present the knowledge base of the University of Veterinary Medicine Budapest and its partners in a focussed way in order to improve the prestige of the Hungarian veterinary profession, and the competitiveness of the organizations in question;*
- *facilitate professional relations and collaboration;*
- *support open access.*