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M. Sc. Thesis

The parasite fauna of some domestic and wild ruminant species in Bandia and Fathala Reserve in Senegal

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Declaration

I declare that I have written my diploma thesis titled "The parasite fauna of some domestic and wild ruminant species in Bandia and Fathala Reserve in Senegal" on my own with a help of literature listed in References.

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Abstract:

Within the long-term project is *ex situ* breeding Programme of semi-captive *T. derbianus derbianus* from 13 February to 22 March 2011 the parasitofauna of ruminant (Artiodactyla, Ruminantia) species were studied inhabiting in the Bandia and Fathala Reserves from the Republic of Senegal (Western Africa). Parasitological research was made between February and March 2011 (dry season) on eleven common game ruminant species, namely Impala (*Aepyceros melampus* Lichtenstein), Giraffe (*Giraffa camelopardalis giraffa* von Schreber), Roan antelope (*Hippotragus equinus koba* Gray), Waterbuck (*Kobus ellipsyprimnus defassa* Rüppell), Kob (*Kobus kob kob* Erxleben), Gemsbok (*Oryx gazella* Linnaeus), African buffalo (*Syncerus caffer brachyceros* Gray), western Derby eland (*Taurotragus derbianus derbianus* Gray), Common eland (*Taurotragus oryx* Pallas), Bushbuck (*Tragelaphus strepsiceros* Pallas) and two species of domestic animals (zebu cattle and goats) from the neighbouring areas of the Bandia Reserve.

Coprological examination based on 216 freshly faecal samples, 22 blood smears and 3 examinations of tissues post-mortem revealed the ocurrence of oocyst of *Eimeria* spp. (Apicomplexa: Eimeriidae), eggs of *Moniezia* spp.(Plathelmintha: Anoplocephalidae), Strongyle-type eggs and *Trichostrongylus* sp. (Nemathelmintha: Strongylida), *Trichocephalus* spp. (Nemathelmintha: Trichuridae).

The protozoa at *H. equinus*, *T. derbianus derbianus*, *T. oryx* and *K. ellipsyprimnus defassa* as in domestic animals were found coccidia oocyst genus Eimeria with prevalence from 1.4 to 76.9%. From the faeces of a *T. derbianus derbianus* were isolated oocysts with an average size of the $27.6 \times 21.5 \mu m$. Detailed description of morphology was made, the sporulation time was determined and documented. By comparing literature details was found that this is a new species of *Eimeria* for this host, which was named *E. derbiani* (Máca, 2012 in press).

From the helminth infection were found eggs genus *Moniezia* (Cestoda). Strongyletype nematode eggs were most often found on both reserves with the prevalence from 14.3% to 100% in wild and domestic hosts. The second most commonly occurring nematode eggs were *Trichocephalus* spp. (prevalence from 8.3% to 100%). *Trichostrongylus* spp. eggs were detected only very sporadically in 15.4 % in cattle. The intensity of the eggs found by qualitative coprological examination method using McMaster counting chamber was in most cases low in the range from 11 e.p.g. (eggs per gram) (Strongyle-type eggs from *T. derbianus derbianus*) to 8624 o.p.g.(oocysts per gram) (*Eimeria* spp. ex goat) from 1 g feces. Overall prevalence of 50% were in Bandia Reserve (four parasite groups), 90% for domestic animals bred in the immediate proximity (five parasite groups), 74.2% in Fathala Reserve (three parasite groups). From post-mortem material were recorded three genera, namely: *Cooperioides, Cooperia* and *Impalaya*. Two genera of Ixodes ticks *Amblyomma* and *Hyalomma* were collected. These findings were photographically documented, adult specimens were fixed in the normal way to further study for their eventual closer destination.

Lung worms, trematodes and blood parasites were not found in the investigated material.

Using two nonparametric methods, the Mann-Whitney U test and the Kruskal-Wallis analysis, statistical evaluation was performed and results achieved by selecting a few basic parameters of their relationship was statistically significant, some interesting results are difference among groups of *T. derbianus derbianus* in burden of Strongyle-type parasite group; parasite burden differs with host species feed strategy, some were not significant e.g. burden differs with age groups *T. derbianus derbianus; Moniezia* sp. was not significantly different among hosts species.

Key words: parasite fauna, ruminants, Senegal, faecal sample

Abstrakt:

V rámci probíhajícího *ex situ* projektu na ochranu antilopy Derbyho (*Taurotragus derbianus derbianus*) v Senegalu se v období od 13. února do 22. března 2011 uskutečnila expedice do oblasti rezervací Bandia a Fathala ve státě Senegal (Západní Afrika). Jejím cílem bylo poprvé prostudovat parazitofaunu u 11 druhů přežvýkavců (Artiodactyla, Ruminantia) a sice Impala (*Aepyceros melampus* Lichtenstein), Girafa (*Giraffa camelopardalis giraffa* von Schreber), Antilopa koňská (*Hippotragus equinus koba* Gray), Voduška jelenovitá (*Kobus ellipsyprimnus defassa* Rüppell), Voduška kob (*Kobus kob kob* Erxleben), Přímorožec jihoafrický (*Oryx gazella* Linnaeus), Buvol krátkorohý (*Syncerus caffer brachyceros* Gray), Antilopa Derbyho (*Taurotragus derbianus derbianus* Gray), Antilopa losí (*Taurotragus oryx* Pallas), Lesoň pestrý (*Tragelaphus scriptus* Pallas), Kudu velký (*Tragelaphus strepsiceros* Pallas) a druhů (skot, kozy) vyskytujících se v blízkosti oplocení rezervace Bandia.

Koprologicky bylo vyšetřeno celkem 216 vzorků čerstvého trusu a u třech zvířat (A. *melampus, T. derbianus derbianus*) byla parazitofauna zjišťována postmortálně. Ve vyšetřeném materiálu byli zjištěni prvoci - oocysty kokcidií rodu *Eimeria* spp. (Apicomplexa: Eimeriidae), z helmintů vajíčka *Moniezia* spp.(Plathelmintha: Anoplocephalidae), Strongyle-typu vajíčka a *Trichostrongylus* sp. (Nemathelmintha: Strongylida), *Trichocephalus* spp. (Nemathelmintha: Trichuridae).

Eimerie se vyskytovaly u divokých zvířat (*H. equinus koba, T. derbianus derbianus, T. oryx* a *K. ellipsyprimnus defassa*) s prevalencí 1,4% až 30%. U domácích zvířat (skot, kozy) s prevalencí od 71,4% až 76,9%. Z trusu antilopy *T. derbianus derbianus* byly izolovány oocysty s průměrnou velikostí $27,6 \times 21,5 \mu m$. Byla provedena jejich detailní morfometrická charakteristika, stanovena doba sporulace a nález byl zdokumentován. Porovnáním s literárními údaji lze konstatovat, že se jedná o nový druh kokcidie rodu *Eimeria* u tohoto hostitele a byl pojmenován *Eimeria derbiani* (Máca, 2012 v tisku).

Z helmintozních infekcí byla nalezena vajíčka rodu *Moniezia* (Cestoda) u rodu *Taurotragus* (prevalence 10% až 33,3%) a domácích zvířat (14,3% až 15,4%). Na obou lokalitách se u zvířat vyskytovala nejčastěji vajíčka Strongyle-typu s prevalencí 14,3% až

100% a také vajíčka *Trichocephalus* spp. (s prevalencí od 8,3% do 100%). Velmi ojediněle, pouze v 15,4% vzorků trusu skotu byla detekována vajíčka *Trichostrongylus* spp. Při kvantitativním vyšetřování flotační centrifugační metodou s použitím McMasterovi počítací komůrky byla intenzita nalezených vajíček zjištěných parazitárních zárodků ve většině případů poměrně nízká. Hodnoty se pohybovaly v rozmezí od 11 e.p.g. (vajíčka Strongyle-type z *T. derbianus derbianus*) až 8624 o.p.g. (*Eimeria* spp. z kozy). Celková prevalence diagnostikovaných endoparazitů byla 50% v rezervaci Bandia (čtyři zástupci parasitů), 90% u domácích zvířat chovaných v její těsné blízkosti (pět zástupců parasitů), 74.2% v rezervaci Fathala (tři zástupci parasitů). Při postmortálním vyšetřování zvířat byla zjištěna nákaza dospělými nematody rodů *Cooperioides, Cooperia* a *Impalaya. Z A. melampus, T. derbianus derbianus* a *Hippotragus equinus koba* byly izolovány dva rody klíšťat, *Amblyomma* a *Hyalomma*. Veškeré nálezy byly fotograficky zdokumentovány, dospělé exempláře běžným způsobem fixovány k eventuálnímu dalšímu studiu a jejich bližšímu určení.

Výskyt krevních parasitů, plicní červivost a motoličnatost nebyla u žádného z vyšetřovaných zvířat prokázána.

Vyhodnocení dosažených výsledků bylo provedeno pomocí dvou neparametrických statistických metod, Mann-Whitneyova U testu a Kruskal-Wallisova testu vybráním několika základních parametrů. Statisticky signifikantní byl zejména vztah mezi různým způsobem chovaných skupin *T. derbianus derbianus* a výskytem Strongyle-typu parasitů. Významný byl také vztah výskytu parazitů v závislosti na potravní strategii hostitelů. Nesignifikantní byl např. výskyt helmintů v závislosti na věku antilopy *T. derbianus derbianus*, stejně jako i *Moniezia* spp. u všech druhů zvířat.

Klíčová slova: parazitofauna, přežvýkavci, Senegal, trus

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1 Introduction

Reserves of Bandia and Fathala were established to promote the survival of the native fauna of Senegal based on population management as a tool in the recovery of the critically endangered species, e.g. ruminants. One of many studies in this geographical zone of Senegal was organized by Derbianus Czech Society for African Wildlife Czech University of Life Sciences in Prague headed by Dr Koláčková in cooperation with Society for the Protection of Environment and Fauna in Senegal, Directorate of National Parks in Senegal and others on the issue of parasitofauna on ruminants initially studied by Antonínová (2002) in her diploma thesis derived from the National Park Niokolo Koba (NPNK). The content and aims of the present thesis are attempting to extend the knowledge already established in NPNK and study for the first time the area of the Bandia and Fathala reserves. In recent years, various questions regarding host-parasite transmission have been arised from these reserves, i.e. whether transmission of parasites from domestic animals to wild animals and/or viceversa have ocurred and what would be its ecological impact on these animals. Therefore, the starting point to carry out present study was develop an inventory about the parasitofauna on eleven previously selected species of ruminants as source of reference for future monitoring strategies of parasites on these animals inhabiting into the reserves.

Fenced area with the western Derby elands (*Taurotragus derbianus derbianus*) part of the reserve of Bandia and as mentioned above, it represents an important animal group to be study for parasites since elands are species that are on the Red list of threatened species with a status of critically endangered (IUCN, 2011) in comparison with other species inhabiting in the same area, most elands do not share a common reserve area and waterholes.

2 Literature overview

2.1 Current knowledge on the occurrence of parasites on wild animals in Africa

There are evidence of experts dealing with the parasitofauna fauna across Africa, however, available literature or information on this issue from the West Africa from which are located reserves is actually fragmented. At least, fourteen most important families of endoparasites have been reported from central Europe and African ruminants (Koch, 2005). By collecting samples and studying the internal parasites of animals from reserves based on present thesis, it is possible to complement, extend or bring out information regarding parasite composition and diversity. Only, Antonínová (2002) determined the prevalence and species spectrum of parasites in NPNK of 8 host ungulate species. In the literature overview of the present thesis, will focus mainly on works from West Africa, but also from other areas due to the poor accessibility of literature.

2.1.1. Blood parasite

Blood parasites are associated with the occurrence of vectors. Main tick-borne pathogens of domestic and game animals are of the genera *Babesia, Theileria, Anaplasma,* and *Ehrlichia* (Thomas et al., 1982; McInnes et al., 1991; Gueye et al. 1993; Oosthuizen et al., 2009; Tonetti et al., 2009; Pfitzer et al., 2010;). Gueye et al. (1993) found 10 species of blood parasites from 200 blood smears (goats, cattle, sheep) in Senegal, tick-born and there is presence of *Trypanosoma vivax* in cattle and *Trypanosoma congolense* in sheep transmited by tsetse flies. Among others studies. Fa11 et al. (1999) found *Glossina morsitans submorsitans* and *Glossina nalnalis nambiensis* with density 5.4 flies/trap/days and infection rate of 2.4% by *T. vivax* and *T. congolense* in Ndama cattle in southern Senegal.

2.1.2. Coccidia (Leuckart, 1879)

The genus *Eimeria* Schneider, 1875 belong to the phylum Apicomplexa Levine, 1970. World-wide protozoan coccidia parasitize mostly the intestine of vertebrates and among them are the must common species with important cost to cause disease coccidiosis. Most species infecting domestic or wild ruminants belong to the genus *Eimeria* (oocysts with 4 sporocysts, each with 2 sporozoites) are often restricted to a certain host species and vary in their pathogenicity (Levine, 1985). According to Turner et al. (2010) results showed, that *Eimeria* spp. are strongly seasonal from which majority of hosts are infected during rainy season rather in dry season. Concerning this parasite group, it is known the highest intensity in juvenils (Harper & Penzhorn, 1999; Matjila & Penzhorn, 2002), and also Turner et al. (2010) promote this well-known thing in terms of Africa.

According to Kusiluka & Kambarage (1996) pointed that studies implemented in Senegal and other neighbouring countries have indicated that coccidiosis is an important subclinical disease, faced to significant economic losses in the small ruminant industry with prevalence range between 40 and 90%. In goats the following species were found by Vercruysse (1982) in Senegal during 12-month period: *Eimeria ahsata, E. arloingi, E. christenseni, E. crandallis, E. faurei, E. intricata, E. ninakohlyakimovae* and *E. parva*, who investigated faecal samples in 577 goats. The prevalence was 85% and mostly multiple parasitism, no seasonality with oocyst output in the range of 1000–5000 o.p.g.(oocysts per gram) of faeces. Levine (1985) detected that the most pathogenic species in goats are *E. arloingi* and *E. ninakohlyakimovae*, in the case of the cattle *E. bovis* and *E.zuernii*. Unfortunately, with some exceptions only coccidia from domestic animals have been extensively studied (Levine, 1985; Levine & Ivens, 1986; Pellérdy, 1974).

According to Antoníová et. al. (2002) coccidia oocyst genus *Eimeria* are present in four species of examinated ungulates from NKNP Senegal with prevalence 4.65% (10–507 o.p.g.), namely *Hippotragus equinus*, *Alcelaphus buselaphus*, *Kobus ellipsyprimnus defassa* and *Taurotragus derbianus derbianus*.

2.1.3. Digenea (Carus, 1863)

Trematodes are very important endoparasites and belong to the phylum Platyhelminthes Gegenbaur, 1859. In the present thesis are mentioned representatives of the class Digenea, which have indirect life cycle and may include one or more intermediate hosts. At worldwide level, the most infectious species is the liver-fluke *Fasciola hepatica* (Lapage, 1956), parasite of cattle, sheep and some species of *Fasciola* spp. are present also in wildlife ruminants. For example, Hammond (1972) found *Fasciola* spp. in the *Giraffa* spp., *Taurotragus oryx, Kobus defassa* and based on other studies in *Aepyceros melampus* (e.g. Horak, 1981). Other common genera are *Schistosoma, Paramphistomum, Calicophoron* and *Cotylophoron* (e.g. Anderson, 1983; Boomker et al., 1984).

Members of another parasite class found in African animals, is Cestoda which contains genera of *Moniezia, Avitellina, Stilesia, Thysaniezia*. In cattle, goats and sheep are ocurring two parasite species of the genus *Moniezia* that are the most prevalent, they are *Moniezia expanza* and *M. benedeni* (Ryšavý & Erhardová, 1953; Kaufmann, 1996; Foreyt, 2001). For example, *M. expanza* and *M. benedeni* (e.g. *M. expanza* ex *H. equinus koba* with 42.7% from no. animals 14; *M. benedeni* ex *Taurotragus derbianus*) and other (e.g. *Moniezia monardi* ex *Hippotragus equinus* and *Kobus defassa*) (Belem & Bakoné, 2009) may occur in wild ruminants (e.g. Graber & Thal, 1979). Ba et al. (1993) presented a population genetic study of *M. expansa, M. benedeni*, members of the Cestoda collected from the small intestine of cattle, sheep and goats in Senegal and compared with samples from France; in this study, they found *M. expanza* in sheep and goats, never in cattle. Species of other genera are also well studied in Central Africa, for example *Avitellina edifontaineus* from *Hippotragus equinus koba* with prevalence of infection of 21.4% on fourteen animals (Belem & Bakoné, 2009).

Graber et. al. (1973) investigated the presence of cysticercosis of wild herbivorous animals in Central Africa and found two species of tapeworms, namely, *Taenia hyaenae* and *T. crocutae* (both in Cestoda). Intermediate hosts for *T. hyaenae* are *Syncerus caffer*, *Adenota kob, Alcelaphus lelwel, Hippotragus equinus, Taurotragus derbianus* and *Tragelaphus scriptus*; hosts for *T. crocuta*are are *Syncerus caffer, Alcelaphus lelwel, Hippotragus equinus.* Schandevyl & Vercruysse (1982) studied cysticercosis of domestic animals, i.e., 51 cattle in Senegal (Dakar) and found 13.7% hearts and 25.5% (masticatory musles) of the total of worm specimens as *Taenia saginata* cysticerci.

According to Antoníová et. al. (2002) ungulates in NKNP are attacked by species of five genera of cestodes. Examination of samples from this latter study revealed the ocurrence of the liver-fluke genus *Fasciola* in three groups of hosts (*H. equinus, Ourebia ourebi*) with prevalence 2.33%; genus *Dicrocoelium* with prevalence 8.14% (*H. equinus, Kobus kob, T. derbianus derbianus, T. scriptus*); *Paramphistomum* with prevalence 6.98% (*Cephalophus rufilatus, H. equinus, T. derbianus derbianus, K. ellipsyprimnus defassa*) and one unknown species of trematod 2.33% (*K. ellipsyprimnus defassa, K. kob*). The same research showed the presence of *Moniezia* spp. with prevalence of 5.81% (*C. rufilatus, H. equinus, T. scriptus*) for all genera with quantitativ value 4–24 e.p.g. (eggs per gram).

2.1.4. Nematoda (Diesing, 1861)

Most available literature is devoted to this parasite group which reflects more attention on the group as many published works are derived mainly from South Africa by scientists as Boomker and Horak (University of Pretoria, South Africa) with colleagues and students who have focused on the parasitofauna of domestic and free-ranging ungulates. This research, has allowed use post-mortem material (i.e., infected tissues) to isolate and classify the types (i.e. holotype and paratypes) of the parasites specimens and determine very accurately morphological traits over all other findings.

Among the most common species from most African ungulates are all those belong to the group of Strongylid-type nematods. It include the stomach and intestinal nematods. Order Strongyles consists of 5 families of parasites in Africa (Chabertiidae, Ancylostomatidae, Trichostrongylidae, Molineidae, Dictyocaulidae). This is a large group, and therefore it is impossible to mention each of them and not even concern of this work. So, present thesis is limited to show only examples of the extent on this concern or most often when occurs Strongyle-type eggs or larvae of these parasites with faeces during examinations derived of the present study.

In species with eggs that are morphometrically different from other species (e.g. genus *Nematodirus*) is possible to do a diagnosis (identity at level of genus), but with some groups it is very difficult to distinguish the genus and not to mention the species. For the purposes of a close taxonomic identification is crucial to obtain, if it possible, just the above mentioned post-mortem material and based on morphometric, morphology or molecular parameters to identify the adult worms to level of species (e.g. accessory copulatory organs of males). All this is possible if the acquisition of knowledge and literature of the monitored area is available. Diversity of parasites is very well described in the species globally wide spread in domestic animals (e.g. cattle, goats, sheep, horse) (e.g. Skrjabin et al., 1952; Lapage, 1956) but still is need to deal with endemic species.

There are not many works dealing with nematodes in Senegal, for instance, according to Vercruysse (1985) who examined abomasums over thousand of sheeps through thirteen months, found evidence to explain the survival of the hypobiotic larvae and adult form of the nematode *Haemonchus* during dry season and Hypobiosisis, a very interesting factor in the defense and survival of these parasites in the unfavorable period for parasite stages (eggs, larvae) during dry season according to e.g. Ndao et al. (1995a).

From wild animals information about 5 parasite groups (namely, *Strongyloides*, *Chabertia, Toxocata, Trichuris, Bunostomum*) from hosts in NKNP is available from Antonínová (2002). This study revealed the dominance of the genus *Toxocara* and *Trichocephalus* (prevalence 13.95%, 9.3%). About nematodes of Central Africa is worth mentioning that study of Belem & Bakoné (2009), who found five species of ruminant monitor load and prevalence of gastrointestinal nematodes (namely *Hippotragus equinus koba, Kobus ellipsiprymnus defassa, Ourebia ourebi quadriscopa, Alcelaphus buselaphus major* and *Syncerus caffer brachyceros*) with results of nine different nematode species burden (*Cooperia spp., Haemonchus contortus, Trichostrongylus sp., Skrjabinema sp., Trichuris ovis, Bunostomum phlebotomum* and *Oesophagostomum* sp.). The largest nematode group was that of *Haemonchus* and *Cooperia* with prevalence of infection of 50-

100% and 16.7-100%, respectively, *Trichostrongylus* spp. and *Bunostomum* only 7.1-50% and 7.1 to 12.5% respectively. *Trichuris ovis* was found in *H. equinus koba* with prevalence 35.7%, *Oesophagostomum* sp. only in 33% of *S. cafer brachyceros*. Host of *H. equinus koba* had the highest number of the infected parasite groups (seven groups of nematodes) Belem & Bakoné (2009).

For instance many species of *Cooperia* may be found in the small intestine of domestic and wildlife animals. They are usually reddish (Fig 4.13), thus they are clearly visible in the small intestinal content. According to Horak (1978) it is the first of all well-known parasites of *Aepyceros melampus* and very common in other wild antelopes.

2.1.5. Ectoparasite

Ticks are the most important group of ectoparasites in human and veterinary medicine, among others because they are vectors of the wide range of pathogenic agents that they transmit (Irvin et al., 1996; Kaufmann, 1996; Samuel et al., 2001; Mediannikov et al. 2010). Acarology is a very active discipline at world-wide level. For instance, the subsaharan West Africa have been studied for ticks infected with Crimean-Congo hemorrhagic fever sampled periodically on a herd of 10-20 animal species (Camitas et al., 1990); ticks from Senegal (e.g. Bandia) on namely Amblyomma variegatum, Hyalomma impeltatum, H. marginatum rufipes, H. truncatum, and Rhipicephalus guilhoni. In Senegal as the dry season proceed adult A. variegatum disappear with the beginning of this season, for H. impeltatum, H. truncatum, and R. guilhoni, adults gobble on ruminants during the first half of the dry season. H. marginatum rufipes is never very abundant, in comparison with A. variegatum and H. truncatum. with multivalent seasonal cycle (Camitas et al., 1990). The same species plus Boophilus geigyi, Rhipicephalus sulcatus, R. senegalensis and R. *lunulatus* found Gueye et al. (1993) in Senegal after examination of 120 domestic animals. Presence of seventeen species of 6 genera of ticks in West Africa confirmed previous identification study according to Walker et al. (2003).

2.2 Influence of some transmission patterns

Currently, there is a great interest at global level to protect and develop actions on conservation of the diversity of the endangered species. The introduction of species increase the risk of diseases spreading into the wild or breed animals into the new areas (Leighton, 2002). Parasites and other diseases, which are one of a major threat to conservation, play a very important role (Lyles & Dobson, 1993; Pedersen, 2007) and wildlife disease management must be compatible with a strategy or program of conservation (Breed et al., 2009). So, it follows with the importance of study, documentation and monitoring the disease situation whether it is for any kind of moving with species (Daszak et al., 2000). Often overlooked issue should be clearly understood as an inherent step in the wildlife translocation and realizing that with the individual it should be transport any harmful pathogens, harbor by animals (Woodford, 2000). In the case of addition of new individuals for breeding or expansion of species spectrum of game etc., it is absolutely necessary quarantine, which is an integral part and should not be underestimated (Woodford, 2000).

As one of the possible consequences of exposure to the game (affecting not only the released animal but also other animals) to transports is the stress (Woodford, 2000). According to Stefanski (2001) stress together with parasites could very well contribute significantly to the outbreak of diseases and other health problems, by compromised of the immune system. In cases with high burdens of parasites or zoonosis, it is necessary to think about include anti-parasitic treatement, which is also a simple reason to prevent the introduction of other types of parasites to the current population (Woodford, 2000; Van Wyk & Boomker, 2011). It is important to be well acquainted with many factors (e.g. host-and parasite-related factors) which can influence success of plan, see below on this regard, in the following sections of this thesis.

2.2.1 Host-parasite specificity

Wild and domestic ruminants share a percentage of their nematode parasites (Allonby, 1975; Graber, 1980) and parasite species from wild ruminants are also presented in domestic hosts (Horak, 1979, 1981; Ruiz et al., 2001). In the study of Ezenwa (2003) 50% of strongyle-type nematode species parasitized more than one host species and higher prevalence was in correlation with host species on overlapped localities. According Horak (1980), ruminants have high possibility to host nematode species of the same family or genus and some helminth parasites of antelope are not highly host specific (Boomker et al., 1986). However, in the case of coccidian parasites prevalence is not correlated with bovid species richness, since unlike strongyles, coccidia are relatively more host-specific group (Pellérdy, 1974; Levine,1985; Levine & Ivens, 1986).

The first demonstration of experimental cross-infection was implement by Le Roux (1926), who first demonstrate of the experimental cross-infection of lambs with the larvae get from the faeces samples of a Hippotragus equinus, Connochaetes taurinus and also from the feces of Aepyceros melampus (Le Roux, 1930). Horak (1980) noted that some hosts are infected by accidental parasites, because of the low prevalence in the host population with more species of animals in the fenced small area. According to Ezenwa (2004), with more bovid species in these area, the percentge of strongyle-type nematodes and parasites diversity is higher, also added that territoriality may serve as protection of sharing parasites with other hosts. Question of sharing parasites across hosts in dependence of many factors (Figure 2.1-2) was studied by Negovetich et al., (2006). Boomker et al. (2000) explained the occurrence of the nematode Cooperia spp. as a direct consequence of sharing pastures of tree hosts of antelopes and also Ocaido et al. (2004) studied helminths that parasitize both wild animal and lifestock (cattle, goats) and found that twenty-two species of parasitic helminths affect this group living, grazing and browsing together. There is also pros for non-territorial wild bovids, because of less faecal contamination of the area (Ezenwa, 2004).

2.2.2 Other factors influencing the parasitofauna richness

Ezenwa (2003, 2004) studied the problem of small African reserves build for saving of endangered species (for instance, *Diceros bicornis* and *Ceratotherium simum*) and his results support studies of many previous autors such as Anderson (1983), Boomker et al. (2000), Horak (1980), and Pester & Laurence (1974) in that unprotected natural areas are less attacked by parasitic diseases. Creating of a small reservation may cause stress not only for wildlife, but may also destroy the balance between host and parasite. Although, it was argued that in natural conditions parasites do not regulate size of populations, mapping studies and is an integral part in whatever interference with natural conditions or planning such as breeding and restocking of game reserves (Tompkins et al., 2002). Before such movements, is an important factor if somehow differ area of import (e.g. precipitation), because the actual capture, transport and change of environment etc. are a lot of stress for animals that can produce outbreak of parasites in the host, they already harbor Penzhorn (2011).

A very important part of a reserve management is to prevent the group size and population density, which may to some extent support increasing the percentage prevalence in some group of parasites (McCallum et al., 2001), population control is an integral part of the game disease management (Lloyd-Smith et al., 2005; Cross et al., 2009), which will avoid similar problems in zoos or farms (Goossens et al., 2005).

There is interaction between nutrition of game animals and parasites, during dry season, nutritional stress supports a higher infestation of some types of hosts (Ezenwa, 2004) and high burden of parasite among adult sheeps and goats are also by reason of nutrition stress in West Africa (Fritsche et al., 1993).



Figure 2.1: "Concept of species sharing among different hosts (triangles) species in various habitats (dashed line). The hosts include browsers (=B), grazers (=G) and mixed feeders (=I), with the number indicating differences in host species. Arrow thickness represents the relative amount of overlap in reference to those parasites infecting both host species" (assumed from Negovetich et al., 2006).

Another factor influencing the transmission of intestinal parasites infectious stages is undoubtedly seasonality. Due to changes in temperature, precipitation, humidity the transfer is very influenced by it and thus the prevalence of parasites in host group species (Gillett, 1974). Many authors dealt with effort predict the annual attack of cattle, sheep and wildlife animals by gastro intestinal nematodes on the basis of seasonal changes in weather. Moisture decides on the possible survival and transmission stages of the life cycle of many groups of parasites to hosts a future in the dry season or rainy season there are many influencing factors (e.g. migration of larvae into the soil, eggs and infectious stages washed with droppings), long dry season may decrease the development and survival of parasite stages in the environment and prevalence with intensity of parasite group in animals (e.g. Stromberg, 1997; Turner & Getz, 2010) and during the dry season in Senegal which last 9 months, nutritional problems are increased by parasites (Ndao et al., 1995b).

2.2.3 Anti-parasitic behaviour

There are not only characteristics of reserves which inflict the parasite group diversity and prevalence. With the most known immunological defenses, animals also varied of adaptations, strategies and trade-off the risk of infection against many things such as nutrition. For many ruminant hosts it include feeding behaviour, social strategy, etc. Several authors have dealt with issue of feeding strategy vs. the prevalence of certain parasite group. For example *Tragelaphus scriptus*, browser feeding where is contamination with infective larva of nematods was very low (Apio, 2003) and generally browsers according Boomker et. al (1984) carry only few gastro intestinal nematodes. In the case of specific sites of defecation should be tactic to avoid re-infection, by *Tragelaphus scriptus* is itused for social function rather then protection of parasite (Apio, 2006).



Figure 2.2: "Diagram showing the determinants of the component community of a single host species. Both habitat preference and level of activity will determine the number of species that could be potentially encountered by the host. Infection requires that the parasite is present in the area and consumed by the host. The worm burden is controled by food preference and social organization. Factor related to the host are placed in ovals" (assumed from Negovetich et al., 2006).

Transmision from domestic to wild animals has been studied by many autors in Africa (e.g. Monnig, 1931; Horak, 1978, 1980; Allonby, 1980; Boomker, 1990; Phiri et al., 2011). According to Boomker (1990), the higher risk is for domestic animals rather then for wild ungulates, because of antelopes better tolerate helmints infection of cattle and sheep than viceversa. All depends on also if there is a contact between this group (Horak, 1980), according to Van Wyk & Boomker (2011) is a problem in intensive situations, especially with the spread of parasites with monoxenous cycles (those host specific parasite species), as is the case of Trichocephalus spp., known for their frequent presence in zoos (Boomker, 2007). Allonby (1980) estimated based on experimental framework, that area use by sheeps together with gazella attend to sharing helminths parasites, here gazelles may play the role of reservoar hosts, so anthelminthic treat in this case is functionless. In Central Africa Graber (1980) studied the impact of the helminth specificity on domestic and wildlife ruminants and found that impact is only rare or not at all in case of close contact of these two groups is consistent with that found by Allonby (1980). In conclusion, Ocaido et al. (2009), monitored the incidence of livestock diseases and found the same or relatively low as compared to other region without wildlife.

3 Aims of study

- I. To identify parasitofauna by coprological examination in selected species of wild ruminants in reserves Bandia and Fathala from the Republic of Senegal.
- II. If possible, to perform a parasitology examination of organs of post-mortal material to further host-specificity of ecto- and endoparasite from shot or dead animals during the expedition.
- III. Guidance to identify parasitofauna of domestic animals bred at close proximity to reserve with wild animals.
- IV. To compare the incidence of parasites in two different reserves in terms of breeding management and results of findings statistically evaluate.
- **V.** In the discussion and conclusion attempt to evaluate the possibility of mutual exchange among the various identified endoparasite species of wild ruminants and also alternatively their importance for domestic animals and vice versa.

4 Materials and methods

Parasite fauna examination of wild ruminant species (Table 4.1) at the two reserves and domestic cattle and goats in the neighbourhood of reserve was carried out during an expedition with the members of the conservation programme in 2011 between 13 February and 22 March 2011 (dry season).

Host species	Common	Reserve	
	name		
		Bandia	Fathala
		reserve	reserve
Aepyceros melampus Lichtenstein	Impala	+	+
Giraffa camelopardalis giraffa von Schreber	Giraffe	+	+
Hippotragus equinus koba Gray	Roan Antilope	+	+
Kobus ellipsyprimnus defassa Rüppell	Waterbuck	-	+
Kobus kob kob Erxleben	Kob	+	-
<i>Oryx gazella</i> Linnaeus	Gemsbok	+	-
Syncerus caffer brachyceros Gray	African buffalo	+	+
Taurotragus derbianus derbianus Gray	western Derby eland	+	+
Taurotragus oryx Pallas	Common eland	+	+
Tragelaphus scriptus Pallas	Bushbuck	-	+
Tragelaphus strepsiceros Pallas	Greater kudu	+	-

Table 4.1: Range of examined wild hosts from two reserves in Senegal (+ present; - absent).

4.1 Study area

Characterization of reserves are mentioned just a few information that may affect exogenous development of parasite stages (oocysts, cysts, eggs), but more detailed information is processed by many authors in the last years, and therefore no need to give them also, for example, there are cited some of these works (e.g. Antonínová, 2002; Nežerková et al., 2004; Bada, 2008; Hejčl, 2009; Hejcmanová et al., 2009, 2010, 2011; Koláčková et al., 2009, 2011).

Bandia Reserve (Figure 4.1-2) is located from the east of Dakar (14°35'N; 17°00'W) and it is the first breeding in Senegal. Pluvial precipitation in this area is about

484 mm. The area of the reserve is around 3500 ha and still extend (Rezk, 2011 personal communication). At the beginning was important to add several species of animals to reserve. To date, some species have been re-introduced since years 1991-1999, *T. derbianus derbianus* in 2000 and imported foreign species in year 1994 (Al Ogoumrabe, 2002). Domestic animals (cattle, goats) circulate freely around the reserve fence (Figure 4.3-5).



Figure 4.1: Overall view of the landscape in the Bandia Reserve.





Figure 4.2: A., B., C. Various examples of vegetation in Bandia Reserve.



Figure 4.3: In the Bandia Reserve wild animals are separated between domestic ruminant species by fences, these fences are also used to separate groups of Derby elands from other species within the reserve.



Figure 4.4: Domestic ruminants are freely occurring around reserve, but do not share pastures or watering with wild ruminants.



Figure 4.5: Domestic animals suffering especially from coccidiosis are looking for the remains of food during the dry season.

Fathala reserve (Figure 4.6) is situated in the south-western Senegal (13°39'N; 16°27'W) the main rainy season is from July to October with annual rainfall 839 mm. The area of the reserve is around 2000 ha and it is divided in two parts. The first is an area of around 1200 ha. Practically all animal species affect this part. Wildlife is native to Senegal (an example *T. scriptus*), but since 2001 local fauna has been enriched by a few species, such as *A. melampus, G. camelopardalis giraffa, H. equinus koba, K. ellipsyprimnus defassa, S. caffer brachyceros* and *T. oryx.* A smaller part of the reserve is used only for the group of *T. derbianus derbianus* antelopes, imported from the Bandia Reserve (Nežerková et al., 2004).



Figure 4.6: A., B., C. Examples of vegetation in Fathala Reserve.

4.2 Method of sampling faeces, internal organs, blood, and their storage until examination

4.2.1 Method of sampling faeces

Faecal samples of wild and domestic animals were collected immediately after defecation (in some cases soon as possible in Fathala, because of timidity of animals). The vast majority of hosts were seen during defecation. Samples from transported animals were removed directly from the rectum. A few individuals of cattle and goats next to the fence of the Bandia reservation were captured for sampling directly from the rectum, or the samples were collected after host defecation from several herds / groups, which were, freely moved or passed. Each sample was picked by a clean plastic bag or glove and immediately placed in a plastic resealable bag. The bag was always marked by waterproof marker indicating the host species name, number under which the information was recorded immediately after collection into a field book (the area of collecting, quality, etc.). In some cases, randomly selected or from young animals, a parallel samples in 2.5% (w/v) aqueous solution of potassium dichromate ($K_2Cr_2O_7$), 70% alcohol, or 10% formalin was made. Samples again cooled until examination at the laboratory of State Veterinary Institute Prague (SVI).

4.2.2 Sampling method of internal organs

Samples of whole or parts of internal organs were removed the same day immediately after evisceration. Samples were examined within a few hours by many native preparations and positive material was fixed using 70% alcohol, or 10% formalin.

4.2.3 Blood sampling method

Blood samples were obtained during the transports by the veterinarian Jiří Váhala (Dvůr Králové ZOO, Czech republic), who removed blood from the ear vein of anesthetised animals, a few drops of peripheral blood into the syringe and then blood smears were made.

4.2.4 Ectoparasites sampling method

Collection of ectoparasites was carried out during transports and the occasion postmortal examinations.

4.3 Parasitological examination methods

4.3.1 Native preparation method

Due to climatic conditions (high temperature) for fast processing of possible fresh material for the presence of parasitic stages, quick orientation and rapid assessment of the situation this method was used. The principle of this technique consists in the preparation of homogenate from about 1 or 3 g of faeces, mixed with and 5-10 millilitres of H_2O and sieved to remove large debris with tea strainer or water droplets on a glass prepared with sample spreads. Observation was carried out at 100–400 x magnification. With this method was also examined post-mortal material from 3 animal individuals. By gradual viewing of organ contents for the purpose of orientation for the presence of adult worms, or scrapings for the detection of parasitic stages.

4.3.2 Flotation-centrifugation coprological method

For the purpose of isolating some parasite eggs was used flotation-centrifugation coprological method according to Breza (1957). It usually consists in examining from 1 to 3 g of the excrement sample, precede through a tea strainer into a test tube and centrifuge for 7 minutes at speed 2500 rpm, pour off supernatant and add, so-called Breza solution to the sediment (3 volumes of saturated solution of $Na_2S_2O_3$, 3 volumes of saturated solution of MgSO₄ and 1 volume of H₂O resulting in specific gravity 1300) and again the same centrifugation. Finally, 3 drops were removed from surface membrane by fired crank with a diameter of 5 mm and transferred to a glass slide (Team of authors, 1989).

4.3.3 Qualitative coprological examination method

This method is intended for the quantitative determination of eggs or oocysts per gram faeces (e.p.g./o.p.g.). Therefore, the flotation-centrifugation coprological method was used according to Breza (1957), with the exact amount (5ml) "Breza" solution and the conversion of eggs (average of eggs/oocysts in three chambers, multiplied by 33x) in McMaster counting chamber.

4.3.4 Sedimentation technique

For detection of the fluke eggs presence in faeces was partly made in the field and subsequently in the laboratory of Prague according to Chyla, which is based on the principle of gradual sedimentation of parasitic stage. Several-fold washing of about 10 g excrements with H_2O , drain through the tea strainer and subsequent examination of sediment.

4.3.5 Larvoscopic technique

The L1 larvae larvoscopic method according to Vajda or modified method according to Baerman was used for the presence of lung nematodes in faeces (Team of authors, 1989).

For the Baermann modified method examination of non-formed sample, a sieve put into containers filled with conical bottom for net H_2O was used. The consistency of sparse faeces were taken into account and the test was carried out after a few hours (12 or more). This method was used primarily for testing faeces for the presence of lung and GIN larvae. This method was used in the field, but all samples were tested at the SVI laboratory.

In the diagnostic laboratory, the Vajdova method was used frequently. Sample (1-3 formed excrement (dropping), depending on the type of host) was placed on a watch glass and sprinkled with H_2O . Examination of the prepared sample was carried out after at least 1 hour. This method was used primarily for testing faeces for the presence of lung and gastro intestinal nematod (GIN) larvae. This method was used in the field, but all samples were tested at the SVI laboratory.

4.3.6 Faecal cultures - cultivation of L3 larvae

In the Prague laboratory was used positive sample for the presence of larval stages of gastrointestinal nematode (GIN) larvae for inclusion into the family group. The principle of this method is to obtain infective larvae from positive faecal samples for the presence of GIN (10–20g) at room temperature 23°C (\pm 2°C). Eggs obtained from a post-mortem examination were also used for cultivation of larvae. All larvae thus obtained from this method were immobilized with Lugol's iodine and observed under a microscope. For this purpose, a method based on star-shaped object was used (Figure 4.11). It was done according to Pavlásek & Nikitin (1991) who created special diagnostic utility for isolating from pure cultures of infective larvae. L-STAR filled with water and placed into the middle of the excrement mixed with sawdust in a Petri dish, which was regularly wetted and checked whether the larvae are released from the eggs and migrate into the water.



Figure 4.11: A. Special diagnostic equiptment (L-STAR) placed in faecal sample; B. detail of isolated invader larvae pure culture of Strongyle-type nematodes according to Pavlásek & Nikitin (1991).

4.3.7 Post-mortem examination

An incomplete helminthological autopsy was performed from animals killed due to inhalation (regurgitated material) or shot was inspected for the presence of ectoparasites and endoparasites. Organ samples (lungs, liver, gastrointestinal tract [stomachs, large and small intestine] and cardiac muscle) were taken immediately in the field, stored in the refrigerator and examinated within a few hours or immediately after evisceration. Visible ectoparasites were collected from the skin. Parts of organs or faeces were also fixed with 70% alcohol, 10% formalin and 2.5% (w/v) aqueous solution of potassium dichromate for later determination or examination using other diagnostic methods unfeasible in the field.



Figure 4.12: Example of positive digestive content of small intestine ex *A. melamphus* with visible reddish adult worms.

Blood samples

In order to determine the possible occurrence of blood parasites were collected blood samples from four host species (nine specimens of *A. melampus*, two *G. camelopardalis giraffa*, two *H. equinus koba*, nine *T. derbianus derbianus*). Blood smears were made, in field, from anesthetized animals designed for transport. Peripheral blood from the ear vein was taken mostly in the afternoon and immediately drop spreads out on the marked and slide glass. After thorough drying, the samples were stained with Giemsa. All blood smears were examined under 1000 x magnification.

4.3.8 Compression method for the presence of *Sarcocystis* spp.

Material was taken for the purpose of detecting the presence of *Sarcocystis* spp. (Apicomplexa: Sarcocystidae) in heart tissue, examined by many compression preparations.

4.3.9 Determination of parasites

The determination of adult nematodes from contens of post-mortal material were obtained from the gastrointestinal tract and performed with the help of available identification keys and other specialized literature (e.g. Skrjabin et al., 1952; Lapage, 1956; Boomker, 1977; Anderson, 2000; Anderson et al., 2009). With the aim of eventual qualification, unsporulated coccidian oocysts (genus *Eimeria*) in animals with a positive finding, an excrement sample was put in 2.5% (w/v) aqueous solution of potassium dichromate (K₂Cr₂O₇) in Petri dishes and let to sporulate at room temperature 23°C (\pm 2°C) according to descriptions and images from Levine & Ivens (1970), Pellérdy (1974) and Duszynski & Wilber (1997).

For parasitological examinations were used Leica DMLB microscope at the SVI laboratory or Intraco SM 2 microscope in Senegal.

4.4 Data analyses

A Statistica 9.1. software was used for statistical analysis results achieved, namely to determine the dependence of the burden of the parasite groups among host species or groups of hosts, feeding types and host ages. For this purpose, nonparametric the Mann-Whitney U test or the Kruskal-Wallis analysis were used.

5 **Results**

The results of examinations on the animals of the reserve Bandia, its surroundings and reserve Fathala are listed on the Figures 5.7-5.12 and all results of coprological methods are collectively shown in Table 5.1.

5.1 Blood parasites

Blood smears were examined for presence of blood parasites from four hosts (*T. derbianus derbianus, A. melampus, H. equinus koba, G. camelopardalis giraffa*) from Bandia Reserve. The result of this examination was negative in all twenty-two cases.

5.2 Results of using qualitative and quantitative coprological methods

Summary of faeces examination results of individual animals in in monitored localities (total 216 faecal samples) is shown in Table 5.1, Figure 5.16-17, below with relevant comment.
Host species	No. of	Endoparasite														
	samples	Protozoa				Cestoda			Nematoda							
		Eimeria spp.			Moniezia spp.			Strongyle-type			Trichostrongylus		Trichocephalus			
											spp.		spp.			
		NP	%	opg	PN	%	epg	NP	%	epg	NP	%	NP	%	epg	
Bandia reserve																
Aepyceros melampus	14	-	-	-	-	-	-	3	21.4	-	-	-	-	-	-	
Giraffa camelopardalis giraffa	7	-	-	-	-	-	-	2	28.6	-	-	-	7	100	-	
Hippotragus equinus koba	10	3	30	22	-	-	-	7	70	22	-	-	6	60	88	
Kobus kob kob	1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
Oryx gazella	1	-	-	-	-	-	-	-	-	-	-	-	1	100	88	
Syncerus caffer brachyceros	12	-	-	-	-	-	-	5	41.7	-	-	-	3	25	-	
Taurotragus derbianus derbianus	70	1	1.4	-	7	10	418	26	37.1	154	-	-	-	-	-	
Taurotragus oryx	12	2	16. 7	110	4	33.3	484	5	41.7	308	-	-	1	8.3	22	
Tragelaphus strepsiceros	3	-	-	-	-	-	-	2	66. 7	-	-	-	1	33.3	-	
Derbianus groups																
T. derbianus derbianus "Dering"	21	-	-	-	1	5	-	5	23.8	-	-	-	-	-	-	
T. derbianus derbianus "Niokolo"	33	1	3	-	2	6	-	11	33.3	-	-	-	-	-	-	
T. derbianus derbianus "Tubab"	16	-	-	-	4	25	-	10	62.5	-	-	-	-	-	-	
Domestic animal																
Cattle	13	10	76.9	4114	2	15.4	88	11	84.6	374	2	15.4	3	23.1	616	
Goat	7	5	7 1.4	8624	1	14.3	-	1	14.3	44	-	-	-	-	-	
Fathala reserve				_		_				_						
Giraffa camelopardalis giraffa	1	-	-	-	-	-	-	-	-	-	-	-	1	100	1089	
Hippotragus equinus koba	26	-	-	-	-	-	-	22	84.6	110	-	-	23	88.5	418	
Kobus ellipsyprimnus defassa	10	2	20	198	-	-	-	9	90	154	-	-	4	40	154	
Syncerus caffer brachyceros	8	-	-	-	-	-	-	4	50	484	-	-	1	12.5	-	
Taurotragus derbianus derbianus	12	-	-	-	-	-	-	9	75	11	-	-	-	-	-	
Taurotragus oryx	6	-	-	-	-	-	-	1	16.7	-	-	-	1	16.7	-	
Tragelaphus scriptus	3	-	-	-	-	-	-	1	33.3	-	-	-	-	-	-	
Derbianus groups																
T. derbianus derbianus "Carang"	8	-	-	-	-	-	-	5	62.5	-	-	-	-	-	-	
T. derbianus derbianus "wild males"	4	-	-	-	-	-	-	4	100	-	-	-	-	-	-	

Table 5.1: Summary results of the prevalence of faecal samples examined by flotation-centrifugation coprological method (NP no. of pozitiv samples; % prevalence of parasite group in host; opg/epg value of oocysts/eggs in one gram of faecal sample)

Reserve Bandia

A total of 130 faecal samples were examined from this reserve (Table 5.1, Figure 5.7) from 9 wild host species in which were found from 1 to 4 different parasite groups (Figure 5.8, 5.16) with overall prevalence rates ranged from 21.4% (*A. melampus*) to 100% (*G. camelopardalis giraffa* and *O. gazella*), and hosts infected with at least 1 parasite species (Figure 5.11).

To detect parasite fauna in domestic animals (Figure 5.10) outside reserve, 2 host species (No. 20) were examined and it was found animals infected with 3 and 5 different parasite group (Figure 5.13), total prevalence rates ranged from 71.4% (goats) to 100% (zebu cattle), hosts infected with at least 1 parasite species.

5.2.1.1 Eucoccidiida (Apicomplexa)

a) Wild animals

From all examined samples, three types of coccidia oocysts genus *Eimeria* were found. The highest percentage prevalence of coccidian was found in the host *H. equinus koba* (30%) with the same intensity in both samples (Figure 5.1 A). We done a quantitative method using the McMaster chamber in more positive samples in this parasite with the highest number o.p.g. in *T. oryx* (110 o.p.g.) and specific parasite identification (i.e. level of species) was not possible.



Figure 5.1: Example of coccidia genus *Eimeria* spp. detected in Bandia Reserve wildlife: A. ex *H. equinus*, B. ex *T. oryx*.

"Examination of faecal samples from *T. derbianus derbianus* revealed the presence of oocysts of the genus *Eimeria*, presumably, to represent a new species, *Eimeria derbiani* n. sp. The new species possesses nearly ellipsoidal oocysts (length/width ratio 1.3) with a bi-layered wall and an average size of $27.6 \times 21.5 \mu$ m. *E. derbiani* possesses a micropyle covered by a micropylar cap and ovoidal, single layered sporocysts with an average size of $14.9 \times 7.7 \mu$ m, each with a Stieda body. Sporozoites of *E. derbiani* possess a large refractile body and a nucleus. Sporulation lasted for 2 days at 23° C" (Máca, 2012 in press). In the case of antelope *T. derbianus derbianus* the prevalence and intensity was the lowest (1.4%) from all positive samples.

b) Domestic animals

Oocysts from the genus *Eimeria* found in both host species reached a prevalence of 71.4% (goats) and 76.9% (cattle). Cattle quantitative values 110–4114 o.p.g. and the most represented species of coccidia oocysts were similar to *E. auburnensis, E. bovis* and *E. ellipsoidalis* (most abundant) at least four cases, were also similar to the types of *E. zuernii* and *E. bukidnonensis* (Figure 5.2 B.). Goat hosts of 154–8624 o.p.g., which in some cases, after sporulation, resembled species of oocysts *E. christenseni* (Figure 5.2 A.), *E. ninakohlyakimovae, E. ovinoidalis, E. alijevi* and *E. arloingi*.



Figure 5.2: Example of sporulated oocyst of *Eimeria* found in domestic animals: A. *E. christenseni* ex goat; B. *E. bukidnonensis* ex cattle.

5.2.1.2 Digenea and Cestoda (Platyhelminthes)

a) Wild animals

The two host species were positive for the presence of the eggs of the tapeworm *Moniezia* (Figure 5.3) with prevalence 10% and 33.3% on *T. derbianus derbianus* (maximum 418 e.p.g.) and *T. oryx* (maximum 484 e.p.g.).

b) Domestic animals

The eggs of the tapeworm of *Moniezia* (Figure 5.3) were found in the cattle with prevalence of 15.4% and range of quantitative values 22–88 e.p.g. One goat faecal sample was positive with a prevalence of 14.3%.



Figure 5.3: Eggs of *Moniezia* spp. found from the Bandia Reserve: A. egg ex *T. derbianus derbianus*, B. egg ex cattle.

5.2.1.3 Nematoda (Nemathelmintha)a) Wild animals

The most common findings were eggs from this group of parasites with the lowest prevalence for Strongyle-type (Figure 5.4) from a total of 7 kinds of positive hosts, 21.4% *A. melampus* with lowest value of eggs (10 pieces). As shown in Figure 5.8, clearly, the highest percentage prevalence 70% in host species were *H. equinus koba* and the number of eggs in one sample up to 50 pieces (22 e.p.g.). *T. derbianus derbianus* showed to host more species of these parasites with 20 eggs in a single sample and a maximum of 1 to 25 eggs in a sample, the maximum value (154 e.p.g.) and *T. oryx* 308 e.p.g. for other species number of e.p.g. were not determined due to low intensity of eggs found.



Figure 5.4: Eggs of gastrointestinal nematodes isolated from wildlife animal faeces: A, C, D Stongyle-type eggs ex *S. caffer brachyceros* B. Stongyle-type egg ex *T. derbianus derbianus*.

In host species of *G. camelopardalis giraffa* and *O. gazella* prevalences of eggs of *Trichocephalus* sp. (Nematoda) (Figure 5.5) was 100% in contrast (8.3%) to that found in *T. oryx*. This parasite was found also in other four group of hosts (Table 5.1) with quantitative values of 22 e.p.g. in *H. equinus koba*, *T. oryx* and 88 e.p.g. in *O. gazella*, the highest value obtained for all 19 positive samples for the presence of parasites.



Figure 5.5: Isolated egg of Trichocephalus sp. ex G. camelopardalis giraffa.

b) Domestic animals

Examination of faecal samples of zebu cattle animals showed prevalence of 84.6%. including 5 group of parasites (Figure 5.10). The prevalence for Strongyle-type nematodes was much higher than that found (14.3%) in goat hosts with the eggs of several species of nematodes (Figure 5.6) with a maximum of 25 eggs in the sample (22–374 e.p.g.), which resembles in morphology to those of the nematode of *Haemonchus* and *Ostertagia* sp. Identified specimens of *Trichostrongylus* sp. in two samples of cattle were not detected in fecal samples originating from goats.



Figure 5.6: Eggs of gastrointestinal nematodes isolated from domestic animal faeces: A, C eggs of *Haemonchus* sp. ex cattle, B egg of *Oesophagostomum* sp. ex cattle.

Parasite eggs of *Trichocephalus* sp. were only found in cattle with 23.1 % prevalence, but with the highest quantitative value up to 616 e.p.g. from all examined samples in this area.



Figure 5.7: Relation between number of examined samples and positive samples from the Bandia Reserve.



Figure 5.8: Prevalence of parasites on their respective host groups living from the Bandia Reserve.



Figure 5.9: Relation between number of examined samples and positive samples of domestic animals next to the Bandia Reserve.





5.2.2 Reserve Fathala

A total of 66 faecal samples were examined from 7 wild host species from this reserve (Table 5.1, Figure 5.11, 5.16) in which from 1 to 3 different parasite groups (Figure 5.12) were found exhibiting over all prevalence rates ranged from 16.7% (*T. oryx*) to 100% (*G. camelopardalis giraffa*), in hosts infected with at least 1 parasite.

5.2.2.1 Eucoccidiida (Apicomplexa)

Only *K. ellipsyprimnus defassa* was infected with the oocyst of *Eimeria* with prevalence of 20% and quantitative value of 198 o.p.g. Specific parasite identification was not possible.

5.2.2.2 Nematoda (Nemathelmintha)

Most species of ruminants has been infected with Strongyle-type nematodes (6 host species in which forty-four samples were positive) with prevalences varying from 16.7% (*T. oryx*) to 90% (*K. ellipsyprimnus defassa*). Other values found from all examinated species were: the lowest quantitative value with 11 e.p.g. for *T. oryx* and the most high 484 e.p.g. for *S. caffer brachyceros*; values (i.e., number of e.p.g.) from other host species was not determined. In *H. equinus koba* and *K. ellipsyprimnus defassa* were found more species and number of eggs into one sample with up to 20 pieces in first case. *Trichocephalus* sp. were found from this order in 5 host species, in *G. camelopardalis giraffa* with 100% prevalence and lowest 12.5% in *S. caffer brachyceros*. This parasite was very common in *H. equinus koba* with prevalence of 88.5% and the number of eggs in one sample was of 30 pieces. Again, the quantitative method using the McMaster chamber allowed detect more

positive samples of this parasite with the highest number e.p.g. of *G. camelopardalis* giraffa (1089 e.p.g) to lowest in *K. ellipsyprimnus defassa* (154 e.p.g).



Figure 5.11: Relation between number of examined samples and positive samples from the Fathala Reserve.



Figure 5.12: Prevalence of parasites on their respective host groups living from the Fathala Reserve.

5.2.3 T. derbianus derbianus groups

In this case, groups separated from other individuals inhabiting together a reserve and one group of males freely released in Fathala Reserve. The results are summarized in the Table 5.1 and Figure 5.13, 5.16. Eighty-two fresh excrement samples were examined from this host, with an overall prevalence of all groups from 28.6% ("Dering" group) to 100% ("wild males" group).

a) T.derbianus derbianus groups Bandia

In the Bandia Reserve samples from 3 groups of this host were examined and it was found from 2 to 3 groups of parasites with an overall prevalence rates ranged from 28.6% ("Dering" group) to 62.5% ("Tubab" group), with hosts infected with at least 1 parasite. The least representatives were infected herds " Dering", where individuals from this group hosted Stongyle-type nematodes and tapeworm genus *Moniezia*. The group named "Tubab" had a greater percentage for both groups, the prevalence of parasites was determined by quantitative number Strongyle-type nematodes 154 e.p.g. (Didi) and *Moniezia* sp. from 66 to 132 e.p.g. (Dben) that 418 e.p.g. (Didi) for other eggs were found in very small numbers and therefore quantitative method was not used (Table 5.1). For a single group labelled, "Niokolo" were found oocysts of *Eimeria*.

b) T.derbianus derbianus groups Fathala

In Fathala Reserve samples from 2 groups of this host were examined and it was found that 1 group of parasites with an overall prevalence rates ranged from 62.5% ("Carang" group) to 100% ("wild males" group), with hosts infected with at least 1 parasite. Only the group of Strongyle-type nematodes eggs were found in this reserve.



Figure 5.13: Comparison of prevalences depending on breeding group of *T. derbianus derbianus* in both reserves.

- 5.2.4. Statistical analysis of achieved results
 - The comparison among *T. derbianus derbianus* groups

A test was made in order to evaluate weather there is any significant difference among groups of *T. derbianus derbianus* in burden of strongyle-type parasite group (Figure 5.14). Occurrence was significantly different among groups: Kruskal-Wallis test: H (4, N= 82) =13.30; p=0.0099. Also was tested whether there is a difference between the occurrence of parasites in this group fenced groups and wild group animals and it was also found to be significant: Kruskal-Wallis test: H (1, N= 82) =5.58; p = 0.018.



Figure 5.14: Prevalence in samples of five *T. derbianus derbianus* groups with occurence of Strongyle-type nematodes

• The influence of age in *T. derbianus derbianus*

It was compared how parasite burden differs with age of *T. derbianus derbianus* and it was found to be significantly different among host age groups: Kruskal-Wallis test: H (2, N=39) = 5.92; p = 0.052. Following multiple post-hoc test showed not significant difference between age groups, however trends is obvious as illustrated in the Figure 5.15.



Figure 5.15: No. of positive samples from of difference parasite burden with age of *T. derbianus derbianus*



Figure 5.16: Summary of all results from eamination by flotation-centrifugation coprological method.



- Figure 5.17: Summary of all results from examination by flotation-centrifugation coprological method.
 - The comparison among all ruminants

A test was made to explore weather there is any significant diference among all examined host species in burden of parasite group. Occurrence of *Eimeria* spp., Strongyle-type, *Trichostrongylus* spp. and *Trichocephalus* spp. were significantly different among hosts: Kruskal-Wallis test: H (12, N= 216) =101.47, 42.64, 31.38,127.92 ; p<0.001(*Trichostrongylus* spp. p=0.0017), but *Moniezia* sp. was not significantly different among hosts: Kruskal-Wallis test: H (12, N= 216) =16.91; p=0.15.

Comparison between wild hosts and domestic animal species showed that only *Eimeria* spp. and *Trichostrongylus* sp. were significantly different: Mann-Whitney U test, t < p, p < 0.001. All other parasites did not show differences, $p \ge 0.1$.

• The comparison among all ruminants between reserves

Also a test using parasite groups with hosts (*H. equinus koba, S. caffer brachyceros, T. derbianus derbianus, T. oryx*) present in both reserves was made. For *H. equinus koba* only *Eimeria* was significantly different: Mann-Whitney U test, p< 0.01 and all other parasites did not differ, p \geq 0.1. In the case of *S. caffer brachyceros* Strongyle-type and *Trichocephalus* spp. with not significant result: Mann-Whitney U test, p \geq 0.1; from *T. derbianus derbianus* Strongyle-type was significantely different: Mann-Whitney U test, p \geq 0.1; from *T. derbianus derbianus* Strongyle-type was significantely different: Mann-Whitney U test, p \geq 0.1.

• The influence of feeding strategy

A statistic test was use to compare how parasite burden differs with host species feed strategy (Figure 5.18). It was significantly different among hosts: Kruskal-Wallis test: H (2, N= 216) =26.92; p<0.001. Following multiple post-hoc test showed significant difference between 1 and 2 or 3 ($p\leq0.001$) but comparisons between 2 and 3 were not significant (p=0.43) see figure below.



Fig 5.18: Statistical analysis of different feeding trategy (GR = grass and roughage feeders, INT = intermediate feeders, CS = concentrate selectors) with occurence of Strongyle-type nematods.

5.3 Results of excrement cultivation from wild and domestic animals

For the larval culture was used the above-mentioned method. While samples from cattle were released larvae of the genus *Oesophagostomum* spp. and *Cooperia* spp. (Figure 5.19), those samples fixed in alcohol from *Impalaya* sp. (by extruding eggs from the female genital tract) were not obtained larvae. The larvae have been obtained only from samples *Haemonchus* spp. from *T. derbianus derbianus* (Figure 5.20) and *Cooperia* spp. from *S. caffer brachyceros* (Figure 5.21).



Figure 5.19: Larvae isolated from fresh faeces obtained 9. days after cultivation under laboratory conditions using an infected cattle: A. larva of *Oesophagostomum* spp. (a. detail of tail, b. detail of anterior region), B., C. *Cooperia* sp.



Figure 5.20: Larvae isolated from fresh faeces after cultivation under laboratory conditions ex *T. derbianus derbianus*: A, B larva genus *Haemonchus* spp., C detail of tail and last intestinal cell.



Figure 5.21: Larvae isolated from fresh faeces after cultivation under laboratory conditions ex *S. caffer brachyceros:* A. larva genus *Cooperia* B. detail of tail, C. detail of intestinal cells.

5.4 Ectoparasites collecting

When handling the antelope in the Bandia Reserve *T. derbianus derbianus* (name Teranga) a male tick of the Ixodidae family (genus *Hyalomma*) and on *H. equinus* a *Hyalomma* female individual were found (Figure 5.22).



Figure 5.22: Ticks of the Ixodidae family: A. genus *Hyalomma* ex *H. equinus*, B. ex *T. derbianus derbianus*.

5.5 Results of post-mortem examinations (Bandia reserve)

a) T. derbianus derbianus (female Mbalax)

The surface of the body, lungs, liver, gastrointestinal tract (stomach, large and small intestine) and cardiac muscle with a negative result.

b) A. melampus (two males)

The surface of the body was attacked by ticks of the genus *Amblyomma*, (3 female individuals were found) (Figure 5.23).



Figure 5.23: Ticks of the Ixodidae family: A., B. ticks of genus Amblyomma ex A. melampus.

Lungs, liver, stomach, large intestine and cardiac muscle with a negative result.

Small intestine contained nematodes of the genus *Cooperioides* (Figure 5.24), *Impalaya* (Figure 25-27) [(adult worms and with the highest probability of the male end (Figure 5.26) of the larvae sagittate formations after 3-4 molting (Anderson, 2000), because of the large amount of adults of this genus)] and *Cooperia* (Figure 5.28-29). In the case of genus *Cooperia* probably several species based on morphology of spicules are present.



Figure 5.24: *Cooperioides* sp. (male) ex *A. melampus*: A. anterior region of the male body; B. copulatory bursa of male; C., D. two views of spicules



Figure 5.25: *Impalaya* spp. (female) ex *A. melampus*: A. anterior region of the female body, B.Tail end of female, C. Vulva, releasing egg, D. Eggs within the uterus, E. eject egg



Figure 5.26: The L3-4 stage larvae isolated from abomasum and small intestine ex *A*. *melampus* A. larvae, B. anterior region, C. tail



Figure 5.27: *Impalaya* sp. (male) ex *A. melampus*: A. copulatory bursa of male, B. Dorsal ray, C. Spicules, D. proximal end of spicules, E. posterior end of spicules, gubernaculum



Figure 5.28: *Cooperia* spp. (female) ex *A. melampus*: A. anterior region of the female body, B. Tail end of female, C. Eggs within the uterus, D. eject eggs



Figure 5.29: *Cooperia* spp. (male) ex *A. melampus*: A. copulatory bursa of male, B. Dorsal ray, C. Spicules

5.6 Examination of faecal samples using sedimentation and larvoscopic technique

All collected samples were examined for the presence of lung L1 larvae and trematodes eggs. Results of all samples were initially testing in Africa and subsequently in SVI Prague from which all were negative.

6 Discusion

Department of the Institute of Tropic and Subtropics already have been involved in various research issues in Senegal, especially that of Niokolo Koba National Park, reserves of Bandia and Fathala. One of the long-term project is *ex situ* breeding Programme of semi-captive *T. derbianus derbianus* in these reserves (Koláčková et al., 2011) primarily in conjunction with the Society for the Protection of Environment and Fauna in Senegal, Directorate of National Parks in Senegal, Derbianus Czech Society for African Wildlife Czech University of Life Sciences in Prague headed by Dr Koláčková, and others. It also includes education, training and promotion of local residents, professionals and the general public in the world, as evidenced by the annual reports (e.g. Koláčková et al., 2012). Within these activities one part is about health monitoring in the game reserves and animals intended for transports, which are held regularly to maintain optimum composition of species and genetic variability especially in the case of *T. derbianus derbianus*.

In the past this issues was solved and as well as in Czech problem with parasite fauna of hoofed animals, there may play same role as these issues on conditions of Africa. Under this program, there was opportunity to be a member of the expedition and conducting an extraordinary task to examine the incidence of ectoparasites and endoparasites from the 9 species in the reserve of Bandia and 8 Fathala species by using basic parasitological methods, also from transported animals were taken blood for the detection of blood parasites and perform an incomplete helminthological autopsy to investigate the post-mortal material.

Develop of this research support that initiated by Antonínová (2002) and other efforts in order to continue of adding new data from Senegal authors (Schandevyl & Vercruysse (1982); Vercruysse, 1982, 1985; Kusiluka & Kambarage, 1996; etc.) during a short period of time of the expedition, as mentioned above, there were found parasite used coprological methods in both reserves and simultaneously and domestic animals, which occurs near the fences of Bandia Reserve.

From the group of protozoans was emerged the presence of oocysts genus *Eimeria* on a variety of species of wild ruminant (Pellérdy, 1974; Levine, 1985; Levine & Ivens,

1986). While Antonínová (2002) noted the occurrence of the most infected species of *K*. *ellipsyprimnus defassa*, in the pesent the coccidium genus *Eimeria* was the most common on *H. equinus*. When studying detailed morphometric parameters after sporulation of oocysts under laboratory conditions, examination of the material revealed another kind of species in contrast with that previously described in the genus *Taurotragus*. Based on the properties of isolated oocysts, it came to believe that this actually represents a new species, description and diagnosis was proposed and subsequently manuscript accepted for publication.

Also, in domestic animals oocysts occurring in the cattle and goats, which were left sporulate under laboratory conditions in order to be subsequently determined by the type morphological parametres, were highly positive. This results support work of Vercruysse (1982).

From the literature is known that in the study area can be found blood parasites (Gueye et al., 1993) and though was examined the blood smears of 22 animals from 4 species (*A. melampus, G. camelopardalis giraffa, H. equinus koba, T. derbianus derbianus*), but none parasite found.

The aspect of helmint disease were found parasites of cestoda and nematoda groups. The spectrum was lower in comparison with that of Antonínová (2002); present results could be caused by fact that in selected area different conditions and history of living.

The results of the examination clearly showed that nematode eggs in both reserves of ruminant species was ranked as the Strongyle-type eggs. This means of identification was choose, because of well-known reason for their difficult recognition at the genus and species.

Aware of the similarities of these eggs, was necessary to do a culture, an attempt to obtain the opportunity to the larvae of the genus using the diagnostic tool to determine infective larvae. Strongyle-type eggs were found in both reserves and hosts K. *ellipsyprimnus defassa* was the most infected host species in the monitored area.

Trichocephalus spp. widely occurred in both reserves which is a parasite related to the problem of intensive situations (e.g. zoo) and also because of their monoxenous cycle (Boomker, 2007; Van Wyk & Boomker, 2011).

The overall frequency of occurrence and intensity of infections by oocysts or eggs by using standard qualitative flotation-centrifugation coprological method according to Breza (1957) as well as a quantitative method using a McMaster counting chamber was very with low intensity, to which contributed to the dry season during the expedition.

Lung worms were not found in feces, as well as in post-mortal examinations of the lungs.

Dependence of the burden of the parasite groups was tested as well. When was tested influence of age and group of *T. derbianus derbianus* only effect of group was significant. Moreover, the occurence of studied parasites was tested among different ruminant species, only *Moniezia* sp. was not significant. Comparison of wild and domestic animals illustrated difference in genus *Eimeria* and *Trichostrongylus*. Feeding types showed significant difference berween grass feeders and intermediate feeders or concentrate selectors which consistent with e.g. Boomker et al. (1984), Apio (2003) and Ezenwa (2004).

During stay there was opportunity to investigate by incomplete helminthological autopsy two shot males of *A. melampus* and one dead (accident) *T. derbianus derbianus*. From the available literature identification and classification of nematode species was based on morphological parameters, primarily male copulatory organs, by identification keys of authors Skryabin et al. (1952), Lapage (1956), Boomker (1977), Anderson (2000) and Anderson et al. (2009). The obtained material is stored and fixed in SVI for the case of need for specialist or followers. Examination of other organs (liver, stomach, large intestine and cardiac muscle), were examined and they did not reveal occurence of any parasite species.

When handling with anesthetized individuals in Bandia Reserve was picked up a few representatives of the Ixodes ticks, genus *Amblyomma* and *Hyalomma*, whose presence

is characteristic for this area. Most likely these are from species of *A. variegatum* and *H. marginatum rufipes*, which authors describe in previous studies for instance Camitas et al. (1990), Walker et al. (2003), Mediannikov et al. (2010), but this is just an estimate based on these sources of literature. It was not possible to deal more deeply with this problem and is therefore not possible to assess the situation. In the area of Fathala Reserve is a large incidence of *Glossina* spp., but from this region none blood smears were examined for the presence of parasites, which we obtained only from the Bandia Reserve, but unlike the studies in domestic animals Gueye et al. (1993) in Senegal, during this study did not find parasites in wildlife animals.

Frequently, the discussed issue in these terms is that wild domestic animals and wildlife species pose a health risk caused by exchange of parasites. Answer these questions are not easy to clarify, it would require many long-term study, not only to detect spontaneous parasitic infections for each species we tested, but also experimental transmissions of selected species. As far as is know, and literature on coccidia genus *Eimeria* there are strict host specificity (Pellérdy, 1974; Levine, 1985; Levine & Ivens, 1986) from which we deduce that transmission between animals species we examined, are excluded. The situation is different for parasites, for example, when there are common intermediate hosts in trematodes and nematodes, as there is the possibility of mutual exchange Strongyle-type nematods (e.g. *Haemonchus* sp.) nematodes which is present in domestic animal in Senegal (e.g.Vercruysse, 1985) and is also parasite of wild species, confirm by Boomker et al. (2000) who explained the occurence of *Cooperia* spp. as a rusult of sharing pastures of wild and domestic animal in South Africa. It is possible and it succeeded.

In terms of a short period of time of the expedition we considered the chance of route of nematod transmission and tried to examine more detaile studying, observation of animals in the external environment. To avoid interference in Bandia Reserve Figure 4.4 demonstrate separation of wild with domestic animals. From figures it is clear that around the fence on both sides, can be found droppings and excluded from the data, that strongyle infective larvae in dependence on the external conditions may migrate vertically and

horizontally at different heights or to migrate in the substrate (e.g. Stromberg, 1997; Turner & Getz, 2010), without an evidence, just hypothetically, transference could happen, this would need to demonstrate by exploring the substrate and grassy areas around the fence.

For these reasons, it is very difficult to clearly determine the nature of epidemiological mutual transfer between the wild and domestic animals, determine what animal species is important in this transfer. Of course, it is necessary to take into account the climate, because these factors play a role not only during transmission but also influence own parasitological infection (heat, drought) because they erupt development of parasite into the infective stages.

At the end, can be say that the spectrum from the examination was various although intensity of coccidia oocysts and eggs was relatively low, we believe that that it can be a source of infection for animals especially younger groups, first of all in the period of favorable climatic conditions, where is ensured primarily moisture. Currently the material is processed.
7 Conclusion

In the one month period from February to March 2011 was studied parasitofauna of 11 wild ruminant species in the reserves Bandia and Fathala in Senegal. Endoparasites were detected in freely move domestic animals near fences of Bandia Reserve. Animals in both locations was often attacked by groups of nematodes from group of Strongylata and *Trichocephalus* spp. *Moniezia* spp. were diagnosed as parasitizing two species of antelope (*T. derbianus derbianus*, *T. oryx*), cattle and goats only from Bandia. Generally, the spectrum of parasites was relatively richer in Bandia Reserve. Like cattle and goats in European countries, the prevalence of coccidia genus *Eimeria* reached more than 70%.

Due to the very similar parasitofauna found in wild animals in both reserves, there is a real possibility of mutual exchange. For these reasons, mainly from epidemiological aspect recommend in order to the transport of animals from one reserve to another, will for some time, as far as practicable, observed in this region a sort of quarantine or isolation of animals between the original and newly accepted herd animals.

Based on the findings, particularly as regards the excreted intensity parasital stages, as well as because of results of negative tests think about whether it is appropriate to implement preventive anthelmintic treatment.

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