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The use of a herbal supplement (PARANAT®) in the prevention of gastro-intestinal parasites in *Taurotragus oryx*

M. Sc. Diploma Thesis

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Statement

I declare that I worked out this M.Sc. diploma thesis titled " The use of a herbal supplement (PARANAT) in the prevention of gastro-intestinal parasites in *Taurotragus oryx*" alone and that I used only literature that is cited and mentioned in references. I agree with storing this thesis in the library of CULS Prague and enabling it for study use. In Prague: date

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Author’s Abstract

The effect of a commercially produced herbal mixture (Paranat) as a treatment against gastrointestinal parasites has been tested on the eland farm in Lany. The product was presented to 47 animals in total, divided in two groups (control and treatment by Paranat), each of them consisting of one adult male, breeding females and their calves. During the testing period faecal samples were taken on 4 occasions on weekly basis. This resulted in a total of 171 samples. These faecal samples were investigated and FEC was counted using concentration McMaster technique and later processed by use of SAS System V 9.2 (SAS Inst. Inc., Cary, NC) using a General Linear Mixed Model (GLMM).

The highest occurrence of parasite's eggs was proved in calves. In contrary to prediction the suppressive effect of Paranat on occurrence of gastrointestinal parasites was not proved in farmed *Taurotragus oryx,* due to both inconsistency and opposite effect between sampling days in particular group.

Keywords: *Ethnoveterinary medicine, Paranat, Herbal Veterinary Medicine*

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1. Introduction

Infections caused by gastrointestinal helminth parasites of livestock are among the most common and economically important diseases of grazing livestock (Perry et al, 2002). The major losses associated with nematode parasite infections are sub-clinical, and economic assessments show that financial costs of internal parasitism is enormous (Preston & Allonby, 1979; McLeod, 1995). The impact of animal disease on rural poor communities in the tropics have been graphically illustrated (Perry et al, 2002). Gastrointestinal parasitism in ruminant livestock emerged as having the highest global index within a wide range of disease constraints that affect the livelihood of the poor (Githiori et al, 2003). Control of these parasites is commonly achieved by use of synthetic anthelmintics in combination with grazing management. Unfortunately, misuse and poor formulation of the synthetic products have led to the development of anthelmintic resistance (Lans & Brown, 1998; Monteiro et al, 1998). Furthermore, the anthelmintic products may not be readily available to smallholder farmers, or to remote pastoralist communities (Githiori et al, 2003) and control by grazing management such as rotational grazing, is impractical for smallholder farmers and pastoralists due to complications in land ownership and restrictions in the size of individual farms (Githiori et al, 2003).

There is a need to consider a different approach that may lead us to a more sustainable control of parasites (Waller, 2003).
An integrated approach including biological control, reduced frequency of anthelmintic treatments, parasite vaccines, livestock breeds that are resistant to parasites and the use of plants with anti-parasitic properties as well as the use of traditional herbal remedies, or ethnoveterinary medicine should be perceived (Waller, 1999).

Ethnoveterinary medicine may present a cheaper, sustainable alternative if the compounds were demonstrated to work. This thesis provides an experimental evaluation of anthelmintic efficacy of a commercially produced herbal supplement, which bases its actions on the active compounds of medicinal plants (PARANAT). The evaluation was carried out in a ruminant host-parasite model using the Common Eland (*Taurotragus oryx)* herd bred at the University farm in Lány.

2. Literature overview

2.1 Herbal Veterinary Medicine

2.1.1 Introduction

In different fields, such as there are, botany, history, ethnomedicine and pharmacology, descriptions of medicinal plants can be discovered. Traditionally the knowledge and expertise on usage of these plants have been in hands of the plant scientist, the food scientist, pharmacologist, herbalist and farmer.

The interest in the use of these described medicinal plants in veterinary care, I believe, can form a partial solution to many contemporary livestock breeding problems.
Drawbacks to modern veterinary practice include questionable quality of allopathic drugs, development of chemo-resistance in livestock and user unfriendly effect such as high antibiotic and hormone residues in the milk and other animal products (Fielding, 1998;Monteiro et al., 1998, Mathias-Mundy, 2004)

For millennia plants have lived in a complex relationship with the rest of the planet's beings. It has been established that plants are complex combination of chemicals which can nourish, heal and kill their consumer. It is in the believes of the herbalist that by use of rational combinations in the practice of medicine, they can attain longer lasting, more profound improvements.
The active ingredients in the plant, which exists of constituents with complex actions, may benefit the patient through additive, antagonistic or synergistic effects and most active constituents may be unknown (Fougere, 2007).
An increasing number of recent studies indicates that nutrition could affect parasitism not only through quantitative variations of different diet components but also by the presence of some qualitative compounds in plants consumed by herbivores, and particularly secondary metabolites (Athanasiadou et al., 2003).

Observations of sick wild animals have been used around the world for traditional herbalists to find new medicine. Based on the folkloric believe that a sick animal will instinctively know how to medicate their ills, the herbalist follow their procedure to find the cure offered by herbs growing in the wild. To give an example, chimpanzees at Mahale Mountains National Park eat at least 26 plant species that are prescribed in traditional medicine for the treatment of internal parasites or gastrointestinal upset that they cause (Huffman, 1998). But although the majority of the evidence on antiparasitic activity of plants has been traditionally based on anecdotal observations, there is currently an increasing number of controlled experimental studies that aim to verify, validate and quantify in a scientific manner such plant activity (Athanasiadou, 2007).

In many parts of the world medicinal plants are still being used to combat parasitism, as they have been for centuries worldwide.

Drawn from the traditional practice, ethnoveterinary medicine describes a range of plant/s or plant extracts suitable for treating almost every parasitic disease of livestock (International Institute of Rural Reconstrucion, 1994; Athanasiadou,2007).

For example, seeds of garlic, onion and mint have been used to treat animals that suffer from gastro-intestinal parasitism, whereas extracts of the tobacco plant have been used to treat the skin of livestock afflicted with external parasites (Guarrera, 1999). Reports from around the world include exhaustive lists of plants that have been reported to have medicinal properties (Hammond et al, 1997; Akhtar et al, 2000; Waller et al, 2001; Nundkumar and Ojewole, 2002, Athanasiadou and Kyriazakis, 2004; Fajimi and Taiwo, 2005, Githiori et al, 2006).

Aspects of Ethnoveterinary Medicine covered in this review relate only to the use of herbal medicine, with an emphasis on plants with anthelmintic effects in ruminant livestock.

2.1.2 Ethnoveterinary Medicine

Ethno-veterinary medicine (EVM) is based on folk beliefs, traditional knowledge, skills, methods and practices to cure diseases and maintain health of animals (Mathias-Mundy &McCorckle, 1989; Tabuti et al, 2003). In other descriptions of EVM it is pointed out clearly how this study of local knowledge is associated with skills, practices, beliefs, practitioner

The health of the working or income-producing animal is of great importance, especially in small scale agriculture. It has been estimated by the WHO that 70% of the world population uses botanical medicine (Eisenberg, 1998) and up to 80% of Africa's population resort to traditional medicine for their health needs (Makhubu, 1998; Anonymous, 2002). Ethnoveterinary practice is used for the maintenance of good animal health in developing countries (Kudi,2003). In all developing nations livestock represents a major asset among resource-poor smallholder farmers, particularly pastoralist communities. In these pastoralist communities indigenous knowledge has been passed over orally and are rich in traditional knowledge of disease control (Lindsay, 1978;Otha, 1984; Wanyama 1997,2000;Ole-Marion,2003).

The traditional systems of veterinary services are useful where modern techniques are non-existent or, when available, are too expensive or difficult for the community to access (Wanyama, 1997, Dano and Bogh, 1999).

EVM describes the practical development applications within livestock production and livelihood systems. Its ultimate goal is to increase human well-being via increased benefits from stock raising (McCorkle, 1998), achieved by maintaining and promoting the health of the animals. Although EVM is widely used in many parts of East Africa, it is not generally accepted by scientists and veterinarians because it is believed that it is associated with superstition, without a place in reality and to be the domain of 'quacks' (Mesfina and Obsa, 1994, Dano and Bogh, 1999).

This statement brings me to some discussion points in EVM.

First I would like to point out that the base of knowledge founding the practices are passed down orally as described before, but I must add a specification to the community in which they practice EVM. In pastoralist communities it is common to share the knowledge and support the availability to all members of the community. However there are exceptions such as where opinions are sought from specialists who claim that they can predict outbreaks of disease and, also that they have a better knowledge of disease control (Fratkin, 1996; Wayama, 1997). A different situation is found in the case of sedentary smallholder farmers (SHF), where it is common that the EVM knowledge is generally preserve of a few people, and this knowledge consists of guarded secrets that are passed down the lineage line (Wanyama, 2000). A few records of animal husbandry methods used by SHF are available (Leakey, 1977;van der Merwe, 2000).

Second point of discussion applies to all communities practicing EVM as the seasonal availability of certain plants is inevitable. Their abundance or scarcity, the ineffectiveness of some treatments, and particularly their potential to induce toxicity in treated animals.

Superimposed on previous remarks is the problem of correct diagnosis of the causal factors of a disease (Schillhorn van Veen, 1997) and some potentially harmful practices in administering EVM (Mathias-Mundy and McCorkle, 1989: Martin et al, 2001). Generally we can find that the understanding of the causes of disease are poor. Because of this lack in understanding the disease process, the treatments and prevention methods may be inappropriate for the disease. In many cases, disease classification by pastoralists and SHF is based on observed signs and abnormalities in the animal, and treatment is usually offered to alleviate these symptoms (Ohta, 1984; Iles, 1993; Catley and Mohammed, 1996; Hammond et al, 1997; Wanyama, 1997, Namanda, 1998; Tamboura et al,2000; Nfi et al, 2001, Alawa et al, 2002, Ole-Marion, 2003).

There have been written many books on EVM and databases and website on the subject may be found (GIRAN, 2004; Ethnovetweb, 2003, PRELUDE, 2004; SEPASAL, 2004). Unfortunately most of them give only short descriptions of the plants used, and conditions that they treat. Often no validation of the effect against these conditions is provided. Most of the research on testing of EVM preparations has so far been carried out in Asia (Akhtar et al, 2000).

The renewed interest for traditional medicine, and in particular the traditional veterinary medicine, was the necessary impulse to revive and maintain the existing knowledge. Which can provide everybody worldwide with a local, sustainable treatment against the diseases threatening our income-producing livestock. Whereas ethnoveterinary practices (EVP) are disappearing because of rapid socioeconomic, environmental, technological changes and as a result of loss of cultural heritage losses under the guise of civilization (Mathias-Mundy & McCorkle, 1989; Nfi et al, 2001).

2.1.2.1 Role of EVM in stockraising (developed and developing world)

I would like to summarize some considerations to the potential EVM consists of in the present problems we face in stockraising. These considerations can be applied both to developing as developed nations.

First consideration focuses on the group of poor or remote stockraisers, who can neither afford nor may access expensive are distant conventional healthcare options. In these cases validated EVM techniques may be the most realistic choice. Furthermore we can even find that wealthier and better-situated stockraisers may benefit from such techniques where conventional services on offer do not respond to these producers particular veterinary needs.

Besides the financial power of stockraisers, we must take in count the production systems and market conditions where the value of the animals in question may not warrant the cost of professional veterinary care and inputs.

If we look at the growing agriculture in tropics and subtropics it is very common that the desired, imported commercial drugs may not be available, supplies may be expired, insufficient or even adulterated. Veterinary professionals to advise on the use of those drugs may be absent and stockraisers (especially illiterate in the language on the drugs labels and instructions) may be uncertain about their indications, dosages and even modes of administration. Dangers here include not only the obvious ones for patients but also the problem of escalating chemoresistance.

In emergencies or fast-spreading epidemics, there simply may not be time for anything other than local practitioners and treatments. Such treatments and help is often cheaper, which make for better return to stockraising and thus are more sustainable.

In the developed world we can see a rising trend in taking environmental pollution and food safety of powerful modern drugs and biocides. Ethnomedical alternatives may prove more benign.

Ofcourse the focus of stockraisers, veterinarians and animal care practitioners must be in the prevention of disease. In line with the philosophy of EVM it must be the long time knowledge on local ecology, livestock and wildlife ethology, natural resources and so forth which may result in preventing disease.

2.2 Regulation and Quality Control of Herbal Medicine

The regulatory status and quality control of herbs worldwide are as diverse as the countries they come from.
The first difference we can point out is the desired end product (tea, crude pills, standardized herbal medicines) and the technology needed to produce them.
It speaks for itself that here we can find large differences between developing and developed world.

We can find that the difference in legislation under which phytomedicine can be placed is depending of the decision whether or not it is considered medicine. In many countries we can find that the national legislation regards them as food or supplements and therapeutic claims are prohibited.

Even countries where traditional or folk medicine is practiced extensively, no statues may be in place to establish herbal medicines within a regulatory framework. However, many of these countries are developing regulations and guidelines with the help of the WHO.

An interesting topic is the legislative requirements regarding export of the medicinal plant products. With the growing economic incentive for many countries exporting on the herbal trade market implemented regulations have resulted in a higher standard for manufacturing practices. If we focus on the veterinary products on the herbal market, we find that the regulatory status is overall less developed. There is a need for a fair, responsible system to attest to proof of quality, to ensure safe and proper use (Fougere, 2007).

The Codex Alimentarius Commission, a joint project of the FAO and WHO, also adopted international guidelines on vitamin and mineral food supplements in the summer of 2005. The Draft Guidelines for Vitamin and Mineral Food Supplements include “other ingredients”, which may include herbs when included in a supplement. In addition, the World, Trade Organization has developed an international Agreement on the Application of Sanitary and Phytosanitary Measures, which defines standards for food safety, veterinary drug and pesticide residues, contaminants, methods of analysis and sampling, and codes and guidelines for hygienic practice (American Herbal Products Association, 2005).

The European Agency for the Evaluation of Medicinal Products has developed guidance documents for Human and Veterinary Medicinal Product Quality (European Agency for the Evaluation of Medicinal Products, 2001). PARANAT® is produced in France, according to the EU Directive on Food Supplements (2005).

Many herbal ingredients have been given GRAS status by the FDA and the AAFCO and are categorized as feed additives, with their intended use defined as “spices and other natural seasonings and flavorings”, or “essential oils, oleoresins, and natural extractives” (AAFCO, 2004). The quantity of the ingredient added to animal food should not exceed the amount reasonably required to accomplish its intended physical, nutritional, or other technical effect in the food (FDA 21 CFR Sec 582.1)

2.3 Anthelmintic herbal treatment

2.3.1 Introduction

Considerable research has shown some plants not only affect the nutrition of the animals, but also have antiparasitic effects (Waghorn & McNabb, 2003).
The usual mode of control of gastrointestinal parasites is based on the repeated use of anthelmintics (AHs), either in the prevention of infection or to cure the animals. However, the development of anthelmintic resistance in worm populations is now a worldwide phenomenon, in constant expansion (Jackson and Coop, 2000).
The constant increase in anthelminitic resistance has stimulated the search for alternative solutions, which is also supported by an enhanced public concern for more sustainable systems of production, less reliant on chemotherapy (Waller, 1999; Jackson, 2000). And we can also conclude on that for farmers it is desirable to minimize anthelmintic usage, to lower costs and to reduce the risk of the development of anthelmintic resistance and carcass chemical residues (Hoskin, et al, 1999).

With the contemporary production processes for meat and animal products, we are challenged with a higher parasitic load and to often the answers were sought in an aggressive chemical approach over the past few years. This kind of domestication of livestock has tipped the balance in favor of parasites. This is because domestication is almost always associated with restriction of livestock movement, increasing stocking rates, increasing the proportion of susceptible animals (young and breeding animals) and increasing productivity demands (Waller, 2002).

The problem of parasitic infection is not only an issue in the developed, industrial agriculture, but can have dramatic consequences even for the SHF. For example, nematode parasitic disease remains one of the greatest limiting factors in successful, sustainable ruminant livestock production worldwide (Perry and Randolph, 1999).

However the interest in different methods of prevention and cure of parasitic conditions has grown. Among these alternative methods, manipulation of host nutrition in order to improve the host resistance and/or resilience to parasitic infections seems to represent one promising short-term options. We can find an increasing number of recent studies that suggest some natural compounds of the diet, in particular plant secondary metabolites, could have direct or indirect effects on nematode populations (Athanasiadou et al, 2003, Min et al, 2003, Waghorn and McNabb, 2003).

2.3.2 Secondary metabolites, tannins and tanniferous plants

There have been done numerous studies on the interactions between secondary metabolites and nematode infections, mainly in sheep and in lesser quantity in goats have been described. A survey of individual profiles of egg excretion from 25 goats bred under rangeland conditions in two organic farms showed dramatic decreases in egg output in some individual goats, exceeding 95% reduction (Hoste et al, 2005). Since there was no AH treatment used, it seems postulated that such dramatic decrease in excretion could be related to the consumption of plants, whose components might affect some traits of life of the nematodes. In this case the precise nature of the compounds responsible for these changes remained undetermined. However, a study in Uganda were two groups of Anglo-Nubian goats, bred under rangeland environment, received or not an inhibitor of tannins (PEG) for 6 months. This distribution induced a significant rise in parasite egg excretion suggesting a possible repressive role of tannin on worm fertility (Kabasa et al, 2000). Moreover, the consumption of PEG was also associated with a better growth of the animals. Next to these previous observations from natural infections, results from *in vivo* studies performed in conditions of experimental infections tend to confirm the hypothesis of a possible impact of plant secondary metabolites on worms. Using quebracho, extracted from *Schinopsis* spp., as a source of tannins, Paoline et al (2003) showed that the presence of 5 to 6% of tannins in the diet resulted in significant variations in the biology of nematode populations. Min et al (2003) showed recently that a short term consumption of *Sericea lespedeza* (*Lespedeza cuneata*), a forage containing 46 kg of extractable tannins per kg DM, induced a rapid and significant reduction in trichostrongyle egg excretion compared to goats ingesting a control forage with only 6g of extractable tannins per kg DM. Last, the influence of two Acacia species, *A. karoo* and *A. nilotica* on goats experimentally infected with *H. concortus*, was examined (Kahiya et al, 2003). Significant effects on egg output and the number of worms were related exclusively to the consumption of *A. karoo* whose tannin content was much higher than *A. nilotica*.

Some *in vitro* studies, based on different bioassays, have been performed in order: (1) To identify the possible sources of tannin that can be used; (2) to examine the possible direct effects of plant components on the worms; (3) to underline and confirm the main implication of tannins in these effect by using of specific inhibitors. By this screening of bioacticvity, extracts of different species of legume forage (*Lotus pedonculatus, L. corniculatus, Hedysarum coronarium, Onobrychis viciifoliae*) have been shown to represent potential sources of secondary compounds affecting different stages of the parasites (Athanasiadou et al, 2001; Molan et al, 2000)

Described are two main hypotheses on the action of tanniferous plants and their effect on the parasitic nematodes. The first hypothesis suggests that the tannins might interfere directly with the biology of various nematode stages. On the other hand, indirectly, tannins could also improve the host nutrition by protecting the diet proteins from ruminal degradation and this could modulate worm biology. Results from the *in vitro* and *in vivo* studies indicate a rapid response after tannin consumption (Min at al, 2003; Paolini et al, 2003) which are in favor of the direct hypothesis, i.e. potential anthelmintic properties associated with some plant components. Nevertheless, the indirect option cannot be discarded (Hoste et al, 2005).

In the past decade several studies have been performed to investigate the potential use of tannin-rich plants as an alternative treatment against parasites. In particular, several legume forages like sulla (*Hedysarum coronarium*), big trefoil (*Lotus pedunculatus*), birdfoot, trefoil (*Lotus corniculatus*) or sainfoin (*Onobrychis viciifolia*) have been studied for their anthelmintic properties (Kahn and Diaz Hernandez, 2000; Min and Hart, 2002, Hoste et al, 2006). The role of tannin anthelmintic activity has been suspected (Molan et al, 200; Paolini et al, 2004). However, the mode of action of these polyphenolic compounds on nematodes remain obscure (Brunet et al, 2007).

Several results suggest that condensed tannins might be one class, amongst the various secondary metabolites present in forages, with potential anthelmintic properties (Bahuaud et al, 2005).

2.3.3 Paranat®

The product **Paranat®** is produced by a French company called Phytosynthese.
This company situated in RIOM Cedex, France, specialises in the production of plant derived additives to farm animal nutrition.

They focus on cattle, poultry and pig production in which they offer several supplements.
Paranat is promoted for the use in cattle and poultry production as a supplement used against parasitism.
Other products are divided under the topics of metabolism, digestion, respiration and stress.

Noyau Paranat B is sold in powder form and described by the company as a sensory additive made from plant extracts for ruminants.

Promoted as a sensory additive for ruminants to maintain their performance of growth and production.
Dose of 20g/day/100kg of body weight single animal as a minimum for five days is recommended for the as therapy in bovines, sheep and goat.
In case of existing parasitism there can be 5g/day/100 kg of body weight used continuously during periods of risk.

Research on the effect of Paranat in Mouflon, Red deer and Fallow deer has concluded that the normal dosage of paranat can be safely used for these animals. There has been found a lack of efficacy for respiratory parasites (Unpublished results of survey made for company VVS, Vermosice, Czech Republic).
It is stated that parasitism can be controlled with repeated treatments (2-3 per year).

According to the producing company Paranat should be efficacious in the prevention of the animals linked to

- nematodes: strongles
- trematodes:small and large flukes

The efficiency and strength of Paranat is linked to the combination of active constituents derived from plant extracts and was demonstrated *in vitro* and *in vivo*:

- santonine
- cineole

In a research made on grazing dairy heifers organised by PHYTOSYNTHESE to proof the effect of Paranat®, it describes thiosulfinates (2400ppm) and arecolin (50000ppm) as the active botanic compounds of the product (Medina, 2009).

The selected plants of Paranat (tansy, beetlenut, cassia and artemisia – the proportion of each plant in the mixture is confidential) are combined with garlic extracts known for their anti-parasitic characteristics.

2.3.3.1 Garlic (*Allium sativum*)

It is stated clearly, by the producing company, that garlic is used in the herbal mixture Paranat®. This plant belongs to the family of Liliaceae and genus *Allium*.
Allium is the name for the onion genus, close relatives of garlic include onion, shallot, leek, chive and rakkyo. *Allium sativum* is indigenous to Asia, but has been commercially cultivated in most countries. Very famous for seasoning dishes in the Mediterranean region, as well as in Asia, Africa and Europe. Ancient Egyptians have described the use of garlic for both culinary as medicinal purposes.

The selected constituents in garlic for medicinal use include alliin (10mg/g fresh weight or 30mg/g dry weight), the enzyme alliinase (10mg/g fresh-converts aliin to allicin), allicin (the main thiosulfinate in crushed garlic-typically 75%), other thiosulfinates, diallyl sulfide, and other allyl sulfides, ajoene, vinylthiins, S-allylcysteine, S-allyl-mercaptocysteine, and many other sulfur-containing compounds, amino acids, glycosides, vitamins, minerals and trace elements including calcium, selenium and germanium. High levels of fructans (approximately 65% of dry weight), fructose polymeres of 10 to 60U (fructants increase mineral absorption, support beneficial bacteria within the digestive tract and help to eliminate pathogenic gut bacteria and yeast). Fresh garlic has a sulfur content of approximately 3mg/g (Fougere, 2007).

The described clinical actions of garlic include anitplatelet, anicholesterolimic, antiseptic, mucolytic and vasodilator (Fougere, 2007).

Traditionally, garlic has been used in humans for abnormal growths, bronchitis, pneumonia, digestive problems, intestinal infection, tuberculosis, dysentery, earache and ear infection, vascular disorders of many kinds, and poor circulation. In World War I, the European soldiers applied it to their wounds and it was also known as Russian penicillin because of its ability to treat infection. In china, garlic is used to prevent influenza, relieve toxicities, and kill parasites such as roundworms and tapeworms (Bensky, 1993).

Garlic has been used throughout the centuries not only for human, but it has common traditional use in veterinary care. In Ireland, one of the most consistent uses of garlic was in the treatment of a cow that was unable to rise after calving. Tradition required that the “worm in the tail” should be removed. Typically, an incision was made over the base of the cow’s tail, and cloves of garlic were inserted under the skin, or a garlic poultice was applied to the site (*A treatise on Veterinary Medicine*, by James White, 1826).

In Indian ethnoveterinary use, the bulb is used for fungal infection and swelling of the tongue, oral blisters and wounds, rheumatism, contagious abortion, tetanus, milky diarrhea, abdominal pain, asthma, polyuria, sores, compound fracture, epilepsy and swelling of the kidney (Williamson, 2002).

It is clear in the above selection of the history of garlic as a medicinal herb, that its long reputation has been supported by medical science. Several studies have revealed a variety of pharmacologic activities of garlic, such as lowering of cholesterol, inhibition of platelet aggregation, immune enhancement, reduction of systolic and diastolic blood pressure and antithrombotic and antioxidant activities (Agarwal, 1996; Augusti, 1996; Kiesewetter, 1993).

Garlic has been used in the treatment of patients with roundworm (*Ascaris strongyloides*) and hookworm (*Ancylostoma caninum* and *Nectar americanus*). Allicin appears to be an anthelmintic constituent, and diallyl disyulphide was not effective (Kempski, 1967; Soh, 1960).
A study in carp reports successfully reducing parasitism by *Cappilaria* species when minced garlic is used (Peoa, 1988) and in the same form as a mosquito larvicide (Amonkar, 1970). A research by Abbels in 1999, on the effect of garlic on reducing fecal strongyle ova count in 12 donkeys (1 boiled bulb in 300ml water per donkey) was proven unsuccessful when compared with the control and fenbendazole treatment group. This failure may have been due to the method used to prepare the garlic, the

dose used, the method of evaluating the outcome, or the fact that it does not have anthelmintic activity in donkeys.

Garlic essential oil may be an effective preventive or curative treatment against several flagellated poultry parasites (Zenner, 2003).

2.3.3.2 Tansy (*Tanacetum vulgare*)

This perennial, herbaceous flowering plant of the aster family, native to temperate Europe and Asia, has been introduced to other parts of the world and in some areas has become invasive (Zohary and Heywood, 1997). Tansy has yellow, button-like flowers which are produced in terminal clusters from mid-to-late summer and produce a camphor-like smell. It is usually 50-150 cm tall, where the erect stem branches near the top (Lecain, et al, 2006).

Tansy has a long history of use and was first recorded as being cultivated for medicinal purposes by the ancient Greeks (Lecain, et al, 2006). Tansy was used to treat intestinal worms, rheumatism, digestive problems, fevers, sores and to treat measles (Lecain, et al , 2006; LeStrange, 1977; Mitich, 1992; Zimdahl, 1989; Durant, 1976). A bitter tea made by tansy flowers has been used for centuries as an anthelmintic to treat parasitic worm infestation (Sumner, 2004).

Other uses of tansy are described, such as the assumption that it would lighten and purify the skin and promoted as a face wash (LeStrange, 1977;Mitich,1992). Another remarkable history in the use of tansy is its use as an inducer of abortions (Mitich, 1992;Kingsburry, 1964), as well as it has been used by women to enhance fertility and prevent miscarriages (Lestrange,1997; Mitich,1992;Haughton, 1980). Today pregnant women are advised not to use the herb (Martin, 2000). Although most of the folkloric beliefs around the medicinal value of tansy has been discredited, it is still a component of some medicines and is listed as a treatment for fevers, feverish colds and jaundice (Lecain et al, 2006;Mittich,1992; Haughton, 1980).

Today's opportunities for tansy can be found in the rise of sustainable agriculture and the research of biological pest control for organic farming, where it can be used in companion planting. Planted alongside potatoes it repels the beetle attacking the vegetable. In one study tansy was found to reduce the Colorado potato beetle population by 60 to 100% (Mittich, 1992; Schearer, 1984;Lecain and Sheley; 2002).

The active ingredient in tansy that is found toxic for internal parasites is the volatile oil, which produces toxic metabolites as the oil is broken down in the liver and digestive tract. The active components of this volatile oil include 1,8-cineole, trans-thujone, camphor and myrtenol. The quantities and proportions of each varying seasonally and from plant to plant (Mittich, 1992; DePooter, et al, 1989; Jacobs, 2008; Judzentiene and Mockute, 2005; Keskitalo et al, 2001).

1,8-cineole is one of the ingredients of Paranat which promotes its anthelmintic use. The toxin is believed to defend the plant leaves against attacks by herbivores (Judzentiene and Mockute, 2005; Jacobs, 2008). Its many biological activities include allelopathy, anesthetic, antibacterial, carcinogenic, fungicide, herbicide, insectifuge, nematicide, sedative, testosterone hydroxylase inducer and others (Mittich, 1992; Depooter et al 1989; Jacobs 2008; Judzentiene and Mockute, 2005).

2.3.3.3 Beetlenut (*Areca catechu*)

Betelnut is the fruit of a tall asiatic betel palm, *Areca catechu*. The tree is slender, erect up to 30 meters tall with a smooth trunk and a crown of large pinnate leaves 1-2 cm long, orange coloured when ripe, with hard fibrous endocarp and a single seed, commonly called a nut (Moises, 2010). The betel nut is about 5 cm long and is mottled brown and gray in color. Encyclopedia Americana (1988) describes the chewing of the betel nut as a stimulant throughout southern Asia. Betel nuts contain the alkaloid arecoline, which is a mild stimulant that produces a feeling of well being. In some parts of the Orient, betel nuts are used to destroy intestinal worms. Elsewhere, they are used as a dewormer in veterinary medicine (Moises, 2010; Paraziale,2003). It is claimed to kill intestinal parasites, such as tapeworms, roundworms, pinworms and body flukes. Betel nut contains substantial tannic and gallic acids. Its astringent properties depend on these constituents. Tannins remain in the intestinal tract when ingested from plants such as catechu species. There they have an astringent effect in the duodenum opposite to what is produced by laxatives (Adzet and Camarasa, 1988). Both the extract of the seed and decoction show 100% cure for pork tapeworm and a 94,1% cure in 120 cases of cestodiasis (Moises, 2010). It is suitable as alternative dewormer for chicken using 2 grams per kilogram body weight (Betarmos, 2002). In goat and sheep the use of 30g/kilogram body weight is effective in expelling eggs and adults of roundworms (Moises, 2010).

2.3.3.4 Artemisia (*Artemisia spp.*)

The genus *Artemisia* consists over 200 to 400 species belonging to the daisy family Asteraceae. The hardy herbs and shrubs are known for their volatile oils. They grow in temperate climates of the Northern Hemisphere and Southern Hemisphere, usually in dry or semi-dry habitats (D'Andrea, et al, 2003). In most species we find strong aromas and bitter tastes from terpenoids and sesquiterpene lactones, which are assumed to be an adaptation to discourage herbivory (Valles and McArthur, 2001).

Wormseed (*Artimesia santonica*) has long been used as an anthelmintic. The anthelmintic activity of the plant is thought to be caused by lactones related to santoning, which is found in wormseed and other species of Artemisia. In addition, thujone can stun roundworms, which can then be expelled by normal intestinal peristaltis (Leung, 1980; Arnold,1989). Santonin is often employed as an anthelmintic for round worms, which it rapidly expels. It is less effective against thread-worms and has no effect on tape worm (Greenish, 1920).

As described by Khare in his book on Indian Herbal Remedies (2004) several different kinds of *Artemisia* have been traditionally used in Ayurvedic and Unani Medicine for the treatment of intestinal parasites. A strong decoction of *Artemisia absinthum* under medical supervision, *Artemisia maritima* as traditional medicine against ascarides and *Artemisia nilagirica* and *Artemisia vulgaris* used as anthelmintic in Ayurveda.

2.3.3.5 Cassia (*Cassia spp.*)

There is a great abundance of *Cassia* species which has led to the incomplete knowledge of all numbers. It is estimated that *Cassia* contains around 692 species (Frodin, 2005). Cassia tolerates a wide range of climates and temperatures, though it tends towards loving warmer conditions. The expressive flowers make them a desirable ornamental plant for parks and gardens.

"Cassia" is frequently encountered in texts on herbalism and alternative medicine. This is usually *Senna*. This is due to the confusion about which species belong into the *Cassia* family and in many cases it is hard or impossible to determine whether species from the present genus, or *Senna* or if *Cinnamomum* is meant. *Senna* is better-studied and its medical properties are thought to be more pronounced.

In Ayurvedic medicine the *C. fistula* is described as a "disease killer". Where it's quantities of anthraquinones are mainly useful against gastrointestinal conditions. Also *Cassia tora L.* (*Cassia obtusifolia L.*) leaves and seeds are according to Ayurveda laxative, antiperiodic, anthelmintic, ophthalmic, liver tonic, cardiotonic and expectorant (Deore et al, 2009). In The Ayurvedic Pharmacopia of India (part I, volume III, p153) the leaves and seeds are described to be useful in leprosy, ringworm, flatulence, colic, dyspepsia, constipation, cough, bronchitis, cardiac disorders.

In the research by Deore (2009) the alcoholic seed extract of *Cassia tora* demonstrated paralysis as well as death of worms (*Pheritima posthuma, Ascardia galli*) in a less time as compared to piperazine citrate, especially at higher concentration of 100mg/ml. Phytochemical analysis of the crude extracts revealed presence of flavonoids as one of the chemical constituent. The polyphenolic compounds in *Cassia* also show anthelmintic activity (Bate-Smith, 1962). Deore (2009) expects that the phenolic content in the extracts of *Cassia tora* interfere with energy generation in helminth parasites by uncoupling oxidative phosphorylation, similar to the action of some synthetic phenolic anthelmintics. But further studies are needed to establish the mechanisms of action.

2.4 Eland antelope (*Taurotragus oryx*)

Taxonomical classification:

- Kingdom: Animalia
- Phylum: Chordata
- Class: Mammalia
- Order: Artiodactyla
- Family: Bovidae
- Subfamily: Bovinae
- Genus: *Taurotragus*
- Species: *T.oryx*

It is said that the name ‘eland’ derived from the Dutch word for moose. It is claimed that the first Dutch settlers gave the name of the largest northern herbivore to the largest wild ruminant herbivore they encountered arriving in the Cape Province. It’s scientific name is made of three Greek words - *Tauros* (a bull), *tragos* (a he-goat) and *orux* (gazelle or antelope).
The eland antelope can be classified to 3 subspecies; the Cape eland - *T.o.oryx* (Pallas, 1766), the Livingstone’s eland - *T;o;livingstonii* (Sclater, 1864) and the East African eland - *T.o. pattersonianus* (Lydekker, 1906) as described in Kingdon (1997).

Also known as the Southern eland or eland antelope, the common eland originates in the savannah and plains found in East and Southern Africa throughout one-third of Africa, with a northern limit of its territory from the northeast through Angola and southern Zaire and then north to include Tanzania, Kenya and southern Somalia (Pappas, 2002).

The average weight of the male, up to 942kg, can be twice the weight of an adult female (Kingdon, 1982). Spiralled horns are found in both sexes, but tend to be thicker and shorter in males (Kingdon, 1982; Pappas, 2002). The dewlap in the male becomes very large and distinctive over time (Kingdon, 1997; Treus and Kravenchko, 1968).

Recognizing different kinds of animals is done easily by observing the transverse white stripes and dewlap tuft (Kingdon, 1982).

Because of its biological, morphological and physiological characteristics many scientific studies state that the eland antelope is closer to cattle than to other antelope species (Treus, 1983). In some cases eland antelope can be better utilized than cattle in the natural habitat of the animal. This has led to some Southern African farmers switching from cattle to eland.

As a herbivore, elands usually eat grass, branches and leaves. Many experiments showed that eland are able to readily adapt to changes in seasonal patterns. They prefer eating vegetation like cross berry (*Grewia occidentalis*) and scientists conclude that due to some kind of evolutionary adaptation the large antelope can survive on lower quality food in times of little rain. Elands also feed on foliage and a variety of seeds, seedpods, herbs and tubers.
The relation between the feeding behaviour and spread of parasitic diseases would be very important (Mayova, 2006). The Pappas (2002) reviewed literature on eland conclude that the eland has been classified as an intermediate feeder preferring forbs and foliage of shrubs and trees and also as a browser adapted to grazing. The eland is relatively resistant and can withstand a long period without water (Taylor, 1969). The minimum water consumption was 54,9 g per 1 kg of body weight (Treus, 1983).

Regarding the social structure of the eland herd we can conclude that the eland is a social animal and lives in large herds up to 500 animals (Estes, 1991). Pappas (2002) mentioned that females tend to stay together with juveniles, while adult males from smaller herds or wander about individually.
Gestation period is about 275 days (Treus and Lobanov, 1971) and usually females produce one calf (Treus, 1983).

Commercial breeding of captive elands focuses on the production of meat and milk (Pappas, 2002). In Askaniya-Nova in southern Ukraine breeding farms have been established since 1892 (Treus and Kravenchko, 1968). In these farms the main focus lays in milk production. Analysis of antelope milk revealed that it contains approximately twice as much protein and fat than milk from the domestic cattle. Some healing effect was also observed (Treus and Kravenchko, 1968).

2.4.1 Parasitism in eland antelope

Deer at low population densities on natural range are generally not affected by disease to any significant extent. However, they are susceptible to many diseases and hence disease becomes an important factor in the intensive management of deer.
The age and condition of a deer population affects the level of parasitism and disease. Generally speaking, as the number of animals held in captivity increases, so do losses due to diseases, if measures are not taken to prevent them.

Healthy deer normally do not have many worms. A low burden probably initiates immunity to the parasite and hence the host resists high burdens where grazing and reinfestation pressures are not too great.
There are widespread reports of gastrointestinal parasitism in the various species of deer. Generally gastrointestinal parasitism is most pronounced in younger animals up to 2 years of age.

Clinical signs of gastrointestinal parasitism include scouring, or diarrhea, loss of condition, decreased weight gains and failure to thrive.

According to a study of Moloo et al (1999) the eland is resistant to trypanosomiasis. But infected with the vector-borne (transmitted by the tick *Rhipicehalus appendiculatus*) disease caused by bacterium *Theileria taurotragi* can sometimes lead to fatalities (Grootenhuis et al, 1981). However the clinical signs observed in the eland are less pronounced in comparison with those found in cattle.

Most common parasites in eland include the following species; (1) *Strongyloides*, (2) *Trichuris* and (3) *Moniezia* spp. (4) *Nematodirus* spp. (Ezenwa, 2003; Goossens et al, 2005; Cordon et al, 2008).

3. Hypothesis

H1: The use of PARANAT will decrease the number of eggs of gastro-intestinal parasites present in the herd of *Taurotragus oryx* which are commercially bred on the University farm situated in Lany, Czech Republic.

H2: The repeated treatment with PARANAT will decrease or maintain of eggs of gastro-intestinal parasites present in the herd of *Taurotragus oryx* which are commercially bred on the University farm situated in Lany, Czech Republic.

H3: The treatment by PARANAT of control group at the end of experiment will decrease the number of eggs of gastro-intestinal parasites present in the herd of *Taurotragus oryx* which are commercially bred on the University farm situated in Lany, Czech Republic.

4. Material and methods

4.1 Study site

The experiment was carried out at eland farm in Lany, which belongs to the Czech University of Life Sciences in Prague, Czech Republic, supervised by Department of Animal Science and Food Processing in Tropics and Subtropics, Institute of Tropics and Subtropics14 km south west from the city of Kladno.

The animals has (except this study period and during winter housing) access to a paddock with total area of 2.5 ha fenced with wooden fencing (2.5m in height). The barn is divided in two pens by one axial feeding corridor.

4.2 Observed Species

47 animals in total were included in the research. From which 26 were female, 21 were male.

A total of 12 calves, 14 juveniles and 20 adults. All antelopes are marked by coloured ear tag with number to be recognized easily.

Table 1: Studied animals

sex	Calves	Juvenile	Adult
Male	Katar,Lesan,Victor, Gloran,Staplan, Ledjan,Loran,Elisak Cavalan	Vasut,Vorik,Kayin, Simba,Dajan,Ellien, Boris,Leon,Latif, Lumo	Lojza,Krul
Female	Viktorkaka,Torana, Dulua	Ghana,Volta, Gimbya,Lubumba	Lydia,Victorie, Lina, Katka, Staple,Viktorka,Sydney, Dulu,Dakarka, Eliska,Cavalia, Lejdym,Varna,Tora,Nassay, Toulavka,Lorie, Lesana

4.3 Method of data collection

4.3.1 sample collection

During the period from August till September 2011 faecal samples were collected. The faeces collection took place in the barn where the animals were closed inside. The samples have been collected immediately after defecation. The fresh samples were taken from the ground and collected in plastic bags marked with the date of collection and the number of the animal. In total 171 samples were collected in course of this study. The samples were stored in a refrigerator at 4℃ in a laboratory at the Institute of Tropics and Subtropics, University of Life Science in Prague until the examination.
Sampling took place 4 times: (I) before administration of Paranat – control and experimental group, (II) after 5-day treatment of Paranat to experimental group, (III) after another 5-day treatment of Paranat to experimental group (10-day treatment in total), (IX) after 5-day treatment of Paranat to the control group.

4.3.2 examination of the samples

The fecal eggs were counted using the **concentration McMaster technique**.
This technique is somewhat more complicated than the *Simple McMaster technique*, but the recovery of eggs is better and the sensitivity is higher (20 eggs per gram of faeces) (Vadlejch et.al, 2001). Therefore, this technique is recommended if a centrifuge is available. Furthermore, the procedure may be more flexible when many samples are handled simultaneously (Vadljech et al., 2001).

Equipment

- 2 plastic containers
- scale
- measuring cylinder
- stirring device

- sieve
- test tube with 4ml and 10ml marks
- test tube rack
- centrifuge
- flotation fluid (FAO): Saturated NaCl with 500g glucose per litre
- pasteur pipettes
- McMaster counting chamber
- microscope with 40-100 x magnification

Procedure

- weighing out 4.0 g of faeces and transfer it to container 1
- adding 56 ml tap water by means of the measuring cylinder (ratio should always be 14ml tap water to 1.0 g faeces. This ratio ensures that 15ml of the resulting faecal suspension correspond to 1.0 g of faeces)
- the faeces is mixed with the tap water thoroughly with a stirring device
- the faecal suspensions is strained through a sieve into container 2
- immediately after the filtering procedure, the faecal suspension is poured into a test tube to the 10ml mark.
- the test tube is centrifuged for 6 minutes at 1200 RPM (revolutions per minute)
- the supernatant is removed with a pipette. The sediment represents ⅔ g faeces
- flotation fluid is added to the 4ml mark
- the sediment is resuspended carefully, by sucking up and down in a Pasteur pipette several times.
- both sides of the McMaster counting chamber is filled with the faecal suspension, immediately after re suspension of the sediment.
- the filled McMaster chamber is left to rest for 3-5 minutes before counting
- the number of eggs are counted in both counting fields number of eggs per gram of faeces are calculated by multiplying the number of eggs by 20

4.3.4 Counting the McMaster chamber

Procedure

- different parasite eggs are counted within the engraved area of both sides of the chamber.
- 40 x 10 magnification is used
- general rules for counting is followed: all eggs inside the grid should be counted plus all eggs touching two sides of the grid, while excluding all eggs touching the two other sides of the grid

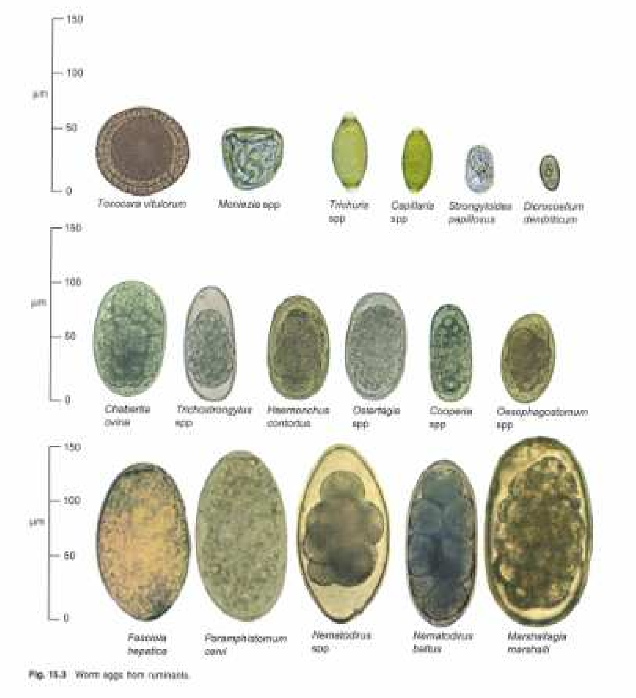
The distance between the upper and the lower glass of the McMaster chamber is 0.15 cm, and the two counting fields each measure 1 x 1 cm. Therefore, the faecal suspension under the two counting fields has a volume of 2 x 0.15 ml = 0.3 ml

In the *Concentration McMaster Technique*, 4 ml of the final faecal suspension in the test tube represent ⅔ g faeces, and therefore the counted volume of 0.3ml faecal suspension represents 1/20g faeces. The number of eggs per gram of faeces (EPG) can now be calculated by multiplying the total number of eggs in both sides of the chamber by 20

4.3.5 Identification of eggs

In this research I focus on the following parasite species Trichostrongylidae, *Coccidia* sp., *Capillaria* sp., *Eimeria* sp., *Nematodirus* sp., *Toxocara* sp. The eggs were identified according to size, shape and typical visible morphological traits analog to the identification pictures (Taylor et al., 2007).

Figure 1: Identification picture parasite eggs used in data collection (adopted from Taylor et al.,2007)



4.4 Data analysis

The faecal egg counts of Trichostrongylidae, *Coccidia* sp., *Capillaria* sp., *Eimeria* sp., *Nematodirus* sp., *Toxocara* sp. were analyzed in order to compare control and treatment means over time using the SAS System V 9.2 (SAS Inst. Inc., Cary, NC) using a General Linear Mixed Model (GLMM). All 171 samples were processed and evaluated for normality. The EPG of the certain parasite was included as a dependent variable. The independent fixed effect of 'group'- (experimental and control), 'Paranat_dosage'- (no Paranat, 5 days or 10 days treatment), 'sampling day'– (1st- 4th), 'age class'- (calf, juvenile, adult) and 'sex' and interaction of effects on parasite load were evaluated as explanatory variables. To account for the repeated measures on the same animals across different sampling day, the analysis was performed using the individual animal as a random factor, using PROC MIXED. The significance of effects in the GLMM was assessed using an F-test.

5. Results

Evaluation of the factors influencing the occurrence of parasite’s eggs.The effects of factors influencing the faecal egg counts output were statistically evaluated. The influence of all effects and interactions were calculated to find the best fitting model with the lowest value of Akaike information criterion starting with full model with all the factors and possible interactions. All the results of final models for each parasite are summarized in the Table 2.

The effect of factors remained in the best fitting model for each parasite. Every parasite was evaluated separately and the results are concluded in the Table 2. In these concluded results only the most significant variables were included, differing for the several types of parasites investigated. This means no comparison can be made on effect of Paranat between parasite types according to the data in Table 2.

Table 2: Results of evaluation of parasite's egg count

Parasite	Effect	F-ratio	Df	P_value
***Nematodirus* spp.**	**Age**	**2.68**	**2**	**0.0789**
	Sampling day*group	**0.47**	**7**	**0.8582**
Trichostrongylidae	**age**	**26.83**	**2**	**0.0001***
	Sampling day*group	**3.07**	**7**	**0.0052***
	Sampling day*Group*age	**2.04**	**14**	**0.0198***
***Coccidia* spp.**	**Paranat dosage*age**	**1.99**	**8**	**0.0518**
***Eimeria* spp.**	**Sampling day*Paranat dosage**	**3.85**	**6**	**0.0014***
***Toxocara* spp.**	**Age**	**0.68**	**2**	**0.5134**
	Sampling day*Paranat dosage	**0.72**	**6**	**0.6375**
***Capillaria* spp.**	**Sampling day**	**5.3**	**3**	**0.0017***
	Paranat dosage	**16.84**	**2**	**0.0001***
	Sampling day*group	**12.47**	**7**	**0.0001***
***Trichuris* spp.**	**Age**	**6.21**	**2**	**0.0045***
	Sampling day*Paranat dosage	**2.52**	**4**	**0.0444***

	Paranat dosage	**0.23**	**2**	**0.7964**

F - value of tested criteria - F-test, Df - degrees of freedom, P - achieved level of significance (P≤0.05) indicates “*”, Single effects and interaction of two and three effects.

What can be concluded in Table 2 is the not consistent effect of Paranat treatment on occurrence of parasites.

However, we can evaluate the most significant data of the FEC on these parasites. We can conclude that in case of *Nematodirus* spp.(Fig 2) and *Trichuris* spp.(Fig 3) the parasites were most abundant in calves.

Figure 2: Effect of the age on the occurrence of *Nematodirus* spp. eggs

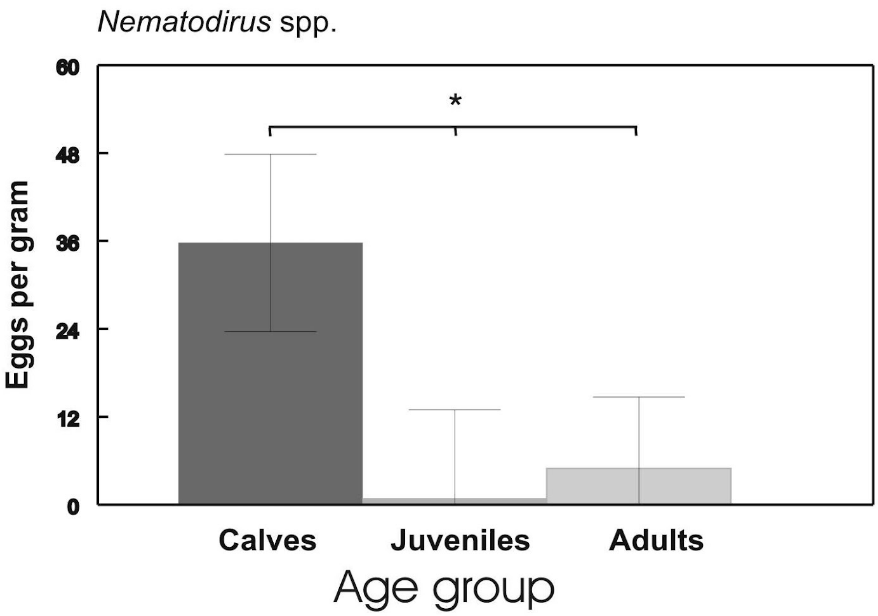
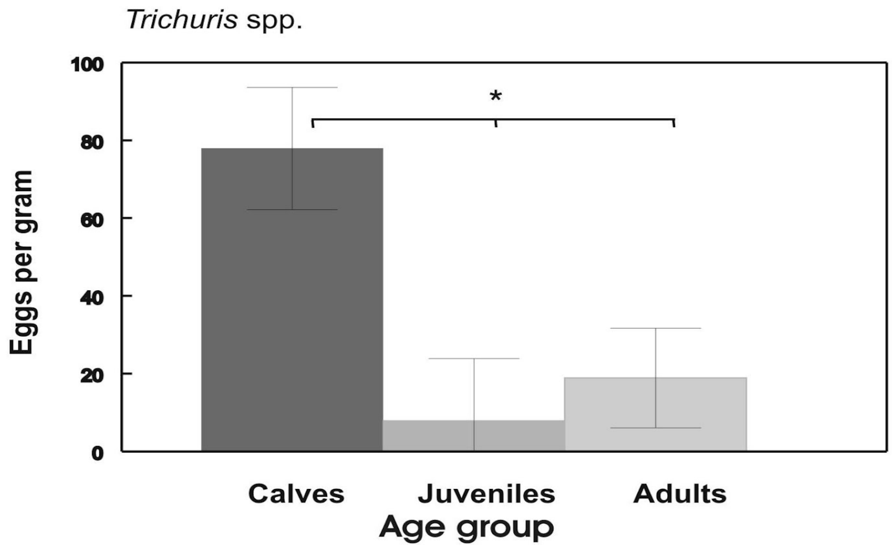
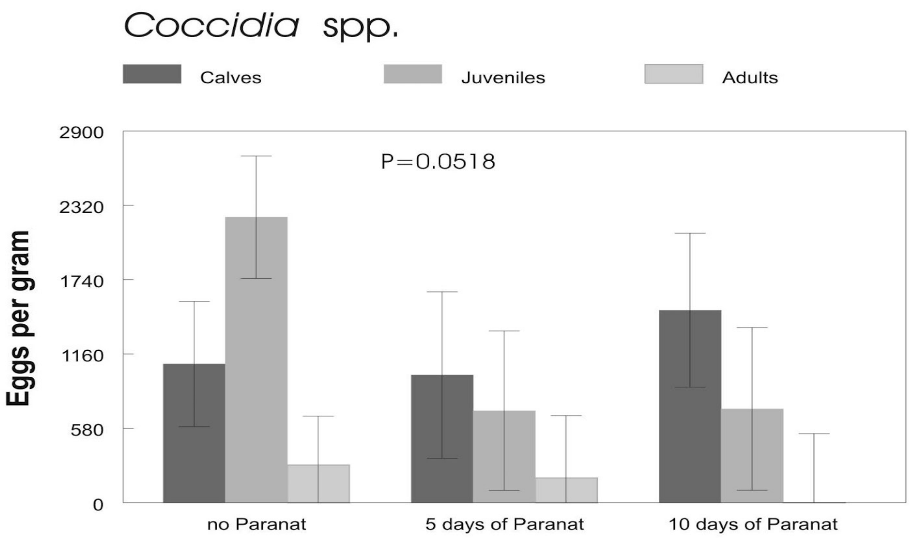


Figure 3: effect of age on occurrence of *Trichuris* spp. eggs



The highest occurrence of parasite's eggs, of *Coccidia* spp. was determined in calves (Fig 4).

Figure 4: Effect of length of Paranat treatment on occurrence of *Coccidia* spp eggs in each age class



The only significant positive results of the Paranat treatment can be found in the FEC of Trichostrongylidae.

The highest occurrence of parasite's eggs of Trichstrongyllidae was found in calves (Fig 5). Furthermore we found a decrease of Trichostrongylidae after use of Paranat dosage after 10 days of treatment. This trend was not consistent and could not be viewed as result of Paranat treatment (Fig 6).

Figure 5: Effect of age on occurrence of Trichostrongyllidae eggs

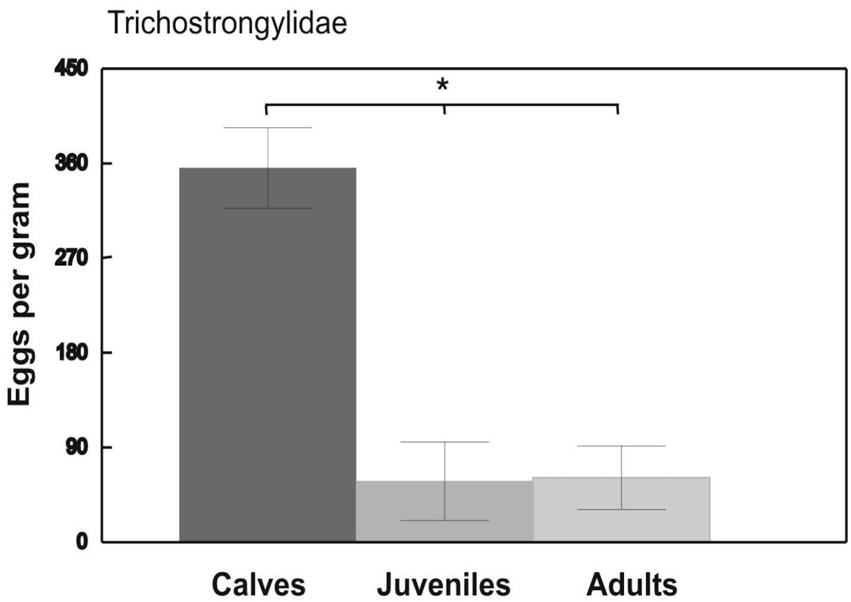
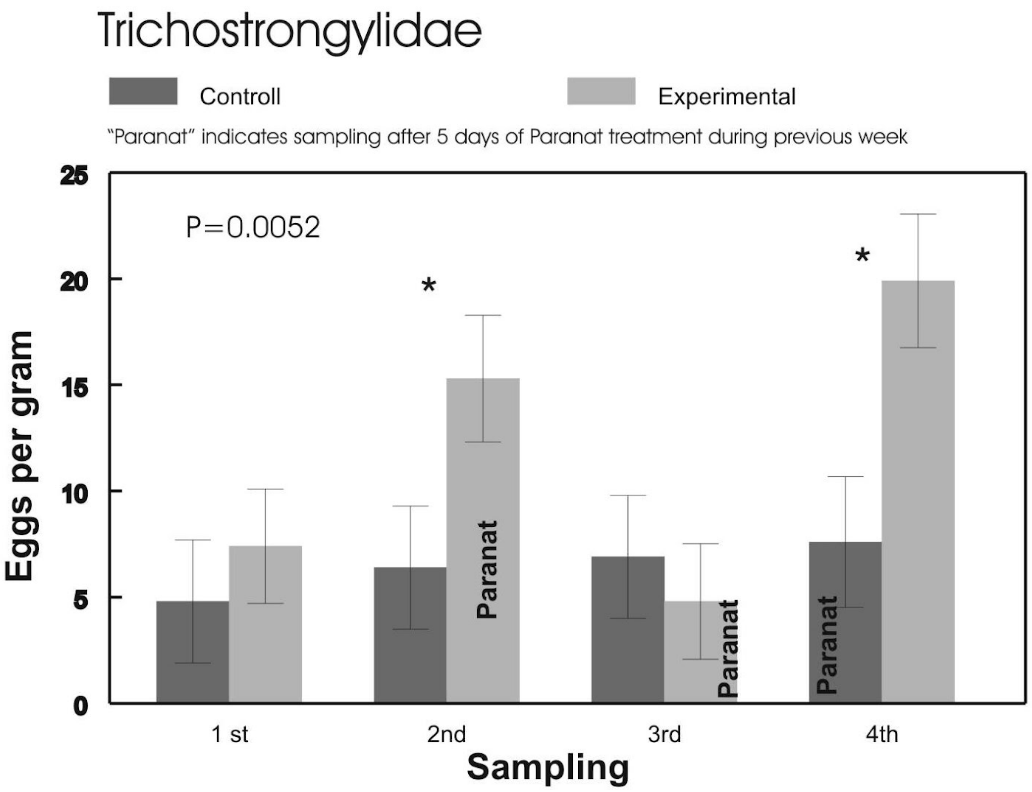


Figure 6: Effect of sampling day (sampling after Paranat is indicated) on Trichostrongyllidae parasite's eggs occurrence for each group



The effect of Paranat on *Capillaria* spp. can be seen as not consistent and seems to increase production of eggs by parasite. We find an increase of FEC after the 10-day treatment with Paranat (Fig 7) and yet an extreme difference in FEC between experimental and control group after 5 day treatment of the latest (Fig 8).

Figure 7: Effect of the length of Paranat treatment on occurrence of *Capillaria* spp eggs

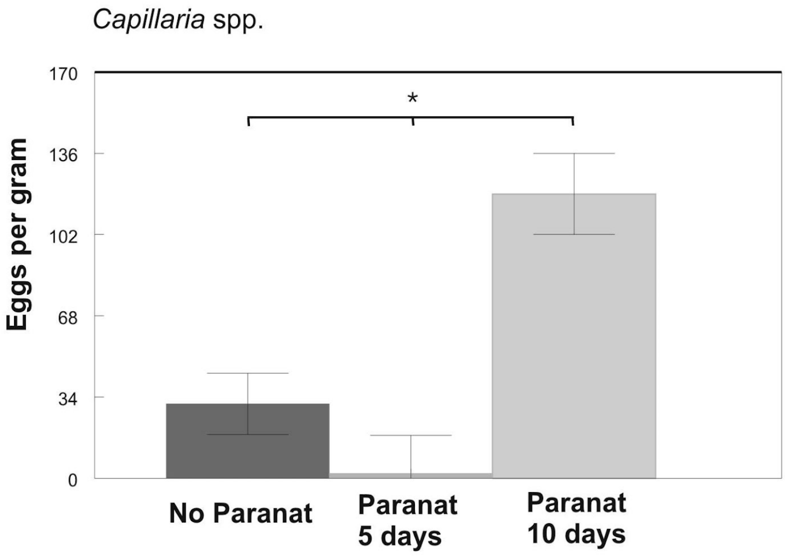
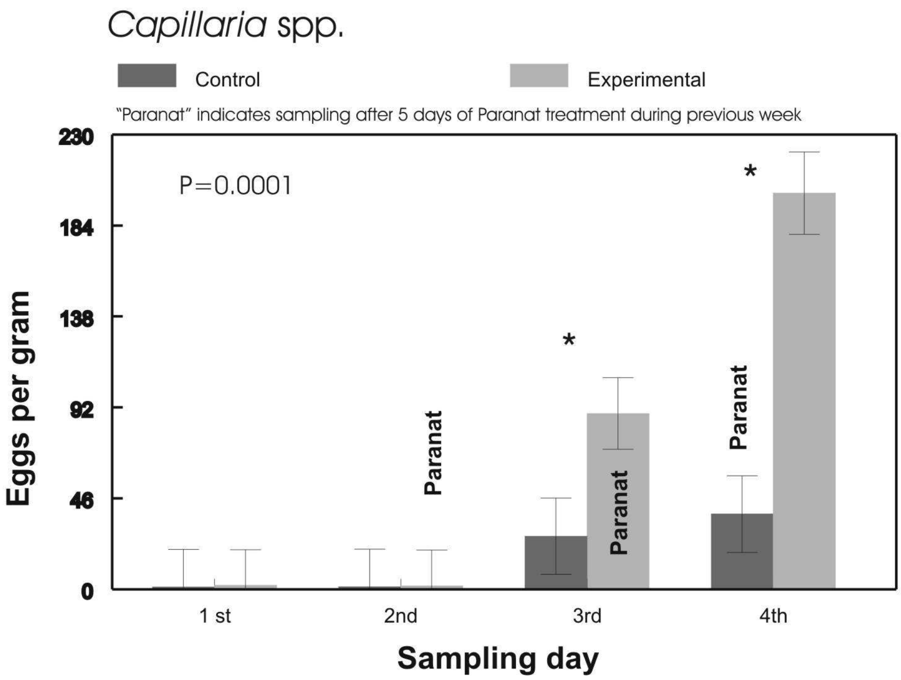


Figure 8: Effect of sampling day (sampling after treatment of Paranat is indicated) on occurrence of *Capillaria* spp eggs in both groups of elands



6. Discussions

The aim of this thesis was to evaluate the effect of a commercial herbal supplement on the gastrointestinal parasites present in farmed eland (*Taurotragus oryx*).
The usage of herbs were evaluated and literature review has given numerous scientific proof of their *in vitro* and *in vivo* suppressive effect on gastrointestinal parasites (Bensky, 1993; Kempski, 1967; Soh, 1960; Peoa, 1988; Zenner, 2003; Sumner, 2004; Moises, 2010; Paraziale,2003; Leung, 1980; Arnold,1989; Deore, 2009; Bate-Smith, 1962).

In vivo research model was found to be a useful tool for rapid screening of anthelmintic activity of plant preparations against nematode parasites. This parasite-host model has been used by the pharmaceutical industry in anthelmintic screening procedures for many years, because it is relatively easy to maintain (Wahid et al., 1989). However, to this no major suppressive effect of Paranat was revealed in present study (Table 2).
When we combine the available data and graphically process them, we can expect a lower action of the herbal mixture on the calves. The highest occurrence of parasites eggs was determined for *Trichuris* spp and Trychostrongyllidae (table 3,5) and seems to be of high influence. The calves experienced the least effect of Paranat, the intake of the product must be considered and since the intake of Paranat was not monitored on a daily basis, the assumption that the calves could have consumed considerable less amounts of the product can be supported by the influence of the rank of animals to feed intake and prevalence of GI parasites (Ruzickova, 2011). Another explanatory factor for these findings could be that the aromatic herbs present in the mixture may have led to decreased food intake even in our case the bulky feed mixture, which was Paranat mixed, was all eaten . As described in the research by Githiori et al (2003) lambs which were fed on traditional and double doses of *Artemisia* spp. showed reduced feed intake and significantly higher FECs were observed in lambs fed on this plant than in controls.

An even more unusual result was found in the processing of FEC data for *Capillaria* spp. Where the prevalence of the parasite was found increased in samples after 10-day treatment. As the first and second sampling of this research took place when the animals were still confined in the barn, as usual during winter times, it was during third and fourth sampling where the animals had access to the pasture (starting April) we found the increased prevalence of the parasite. There is a significant changing factor during the sampling days, taking in account the seasonality of GI parasite infections (Ruzickova, 2004) and the access to pasture. Research on the monthly fluctuation of GI parasite infection in these herds of animals have shown that there is a significant rise of *Capillaria* parasitism in the spring period. When the percentage of *Capillaria* spp. compared to prevalence of other parasites was still 15,8% in March, it had risen to 42,1% in May (Ruzickova, 2004). Taken this in consideration the rise in number of parasites during the third sampling can be seen as a natural occurrence due to changed environmental factors for the animals.

Furthermore we can see a large difference between the 5-day treated control group and untreated experimental group what was already after 10-day treatment at the fourth sampling, which could suggest that the treatment with Paranat was not effective on *Capillaria* spp. for the control group during grazing season.

This assumption can be supported by dependence of explanatory sampling day and group on the effect of Paranat on *Capillaria* spp (p=0.0001).

As a general discussion topic for this research there is to mention that in case of *in vivo* testing of plant material and its effect on parasites, there is a need for standardized tests, with adequate numbers of animals and characterization of the target parasite (Githiori, 2004). Although *in vivo* testing is constrained by expertise, costs, labour, time and facilities, it is necessary to obtain definitive proof of anthelmintic efficacy of any plant based preparation against GI parasites in ruminant livestock.

It is imperative that parasitic infections should be well characterized and methods of application of any plant needs to be ascertained (Githiori, 2004).

Furthermore we can take into account the procedure of evaluation of the effect, the FEC, which is a relatively insensitive test, influenced by factors such as feed intake (and faecal output), and host induced effects (age and immunity) (McKenna & Simpson,, 1987; Githiori,2004).

The use of different evaluation techniques in the research on the effect of Paranat can be seen also a future prospective for more extended results.

7. Conclusions

In conclusion the present study indicated an effect of the herbal mixture Paranat is not consistent and according to predictions and producer statement we found even increased evidence of parasite's eggs after Paranat treatment in elands. The highest occurrence of parasites was found in calves. In that age group, the highest occurrence of parasite's eggs was found for *Nematodirus* spp., *Trichuris* spp. And Trichostrongyllidae. Based on the study we could not recommend use of Paranat for preventive neither therapeutic treatment in captive elands.

I want to refer to my literature review on the use of herbal treatments against GI parasites to defend however the importance of scientific research for the evaluation of EVM treatments. In doing so, exploring the possibilities for the creation of a sustainable agriculture in the tropics and subtropics.
In comparison to the breeding farms present in the developed world, the SHF and pastoralists in the South encounter a larger scope of problems involving the control of parasitism in their livestock. The opportunities of a scientific based use of herbal treatments can enhance the production possibilities for these remote producers.
Further work with browsing trails in the tropical areas could help to avoid the destruction of the ecosystem by making sustainable use of plants in such land. By doing so, both biodiversity of plants and wildlife animals in such ecosystem will be preserved. In the future, these plants could provide resources not only for alternative anthelmintics (from plant secondary compounds) but also for other drugs with medical use (Hoste et al, 2005)

Management of pasture, such as pasture rotation, faeces removal from pasture and many other procedures can be seen as the simplest method of control (Barger,1997; Goosens et al., 2006, Thamsborg et al., 1999). The situation on the eland farm in Lany, were faeces are not cleaned from the grassy paddocks, there is a high concentration of animals per ha and the animals are housed on high straw litter creates during the winter an environment in favor of parasite survival and dispersion in the surrounding environment and animal reinfestation.

The synergistic effects of combining solutions are confirmed when compared to treatment or supplementation alone (Torres-Acosta et al, 2003; Varga-Magnana et al, 2003).

And due to two disparate factors: (I) the evolution of anthelmintic resistance, (II) consumer demand and organic farming movement we can conclude that the global threats imposed by nematode parasites on ruminant livestock production have never been so great as at the present time (Waller,2002).

Therefore, scientific proof of the beneficial effect of medicinal herbs on livestock production by treatment of GI parasites is needed.

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9. Appendices

Capillaria spp. egg

Photo- courtesy of Alena Ruzickova (2011)

Nematodirus spp egg

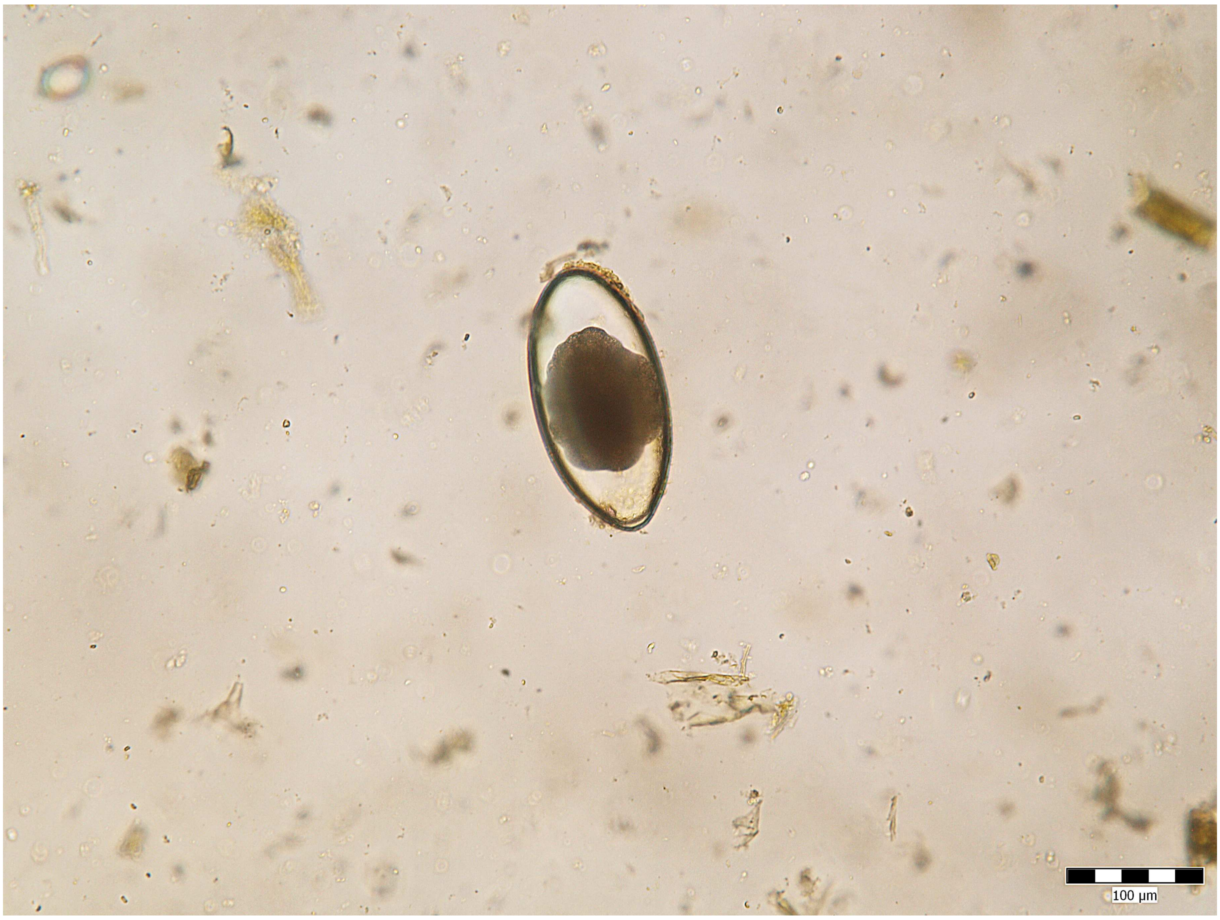

Photo- courtesy of Alena Ruzickova (2011)

Trichostrongyllidae egg

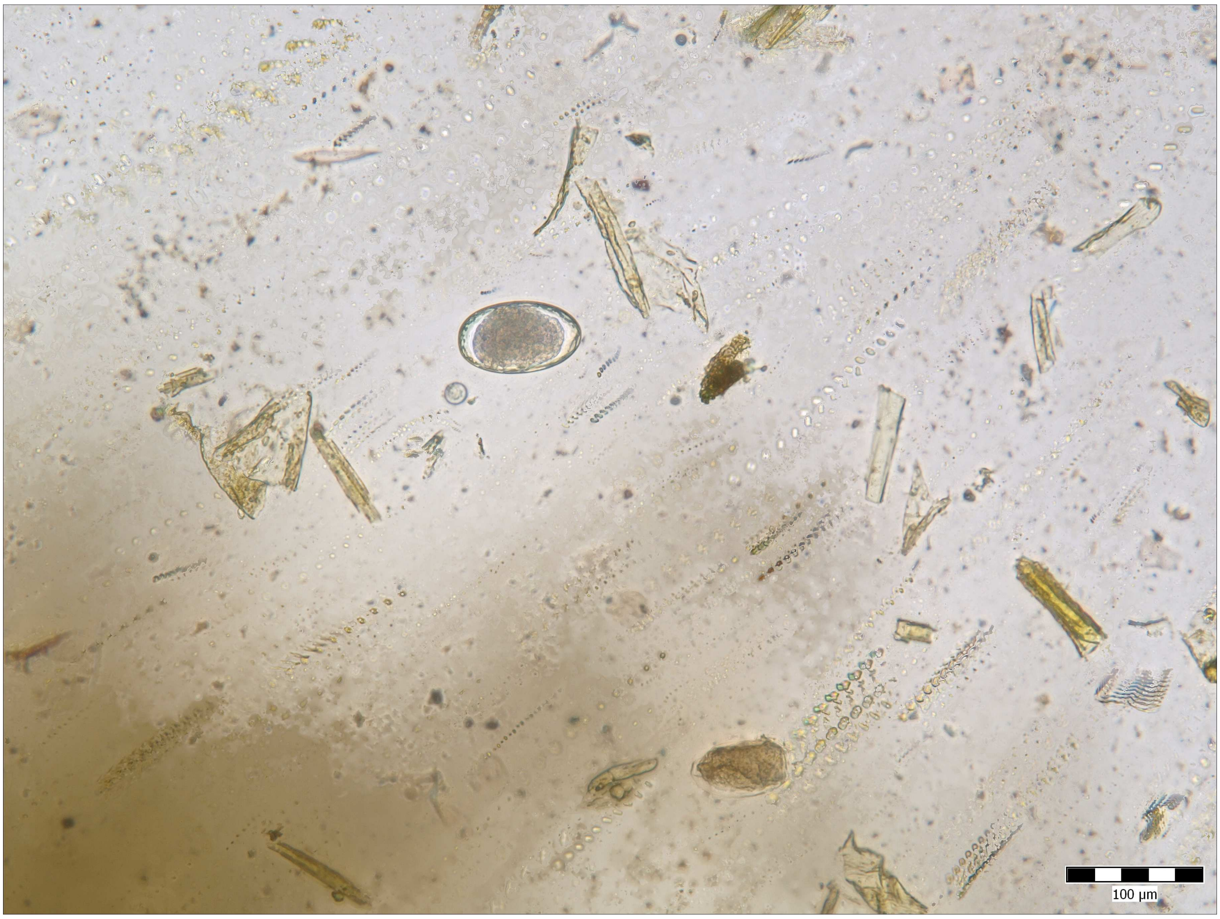

Photo- courtesy of Alena Ruzickova (2011)

Trichuris spp. egg

Photo- courtesy of Alena Ruzickova (2011)