UNIVERSIDADE DE LISBOA

FACULDADE DE MEDICINA VETERINÁRIA

RETROSPECTIVE STUDY ON GASTROINTESTINAL PARASITES OF 28 FELIDAE FROM THE BARCELONA ZOO, FROM 1990 TO 2024

DANIELA PATRÍZIA GONÇALVES OCANDO

ORIENTADORA:
Dra. Raquel Meneses de Souza Silva
COORIENTADOR:
Doutor Luís Manuel Madeira de Carvalho
TUTORA:
Dra. Vanessa Almagro Delgado

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“God requires that we assist the animals, when they need our help.

Each being has the same right of protection.”

— San Francesco d’Assisi

ESTUDO RETROSPETIVO SOBRE OS PARASITAS GASTROINTESTINAIS DE 28 FELIDAE DO ZOO DE BARCELONA, DE 1990 A 2024

Resumo

Os jardins zoológicos tornaram-se valiosos na manutenção e criação de uma variedade de animais selvagens, entre eles espécies vulneráveis e criticamente ameaçadas, trabalhando assim para a sua conservação e prevenção de extinção. Esta grande responsabilidade de abrigar animais tão valiosos inclui a prevenção e a cura de doenças, visto que alojar animais longe dos seus habitats naturais envolve o controlo de diversas condições predisponentes que facilitam doenças e infeções, por vezes de agentes que não estão presentes no seu habitat natural, e contra os quais o animal não é capaz de se defender.

Infeções parasitárias já foram relatadas em vários jardins zoológicos e têm uma grande influência na saúde dos animais nestes estabelecimentos. A sua prevenção e gestão são essenciais para o bem-estar dos animais.

Quatro espécies de Felidae do Jardim Zoológico de Barcelona foram investigadas quanto aos seus parasitas gastrointestinais, de março de 1990 a fevereiro de 2024. O estudo consistiu na análise retrospetiva da base de dados do Jardim Zoológico sobre os parasitas de 12 leões Sul-Africanos (*Panthera leo melanochaita*), quatro jaguares (*Panthera onca*), sete leopardos do Sri Lanka (*Panthera pardus kotiya*) e cinco tigres de Sumatra (*Panthera tigris sumatrae*). Os parasitas presentes nas amostras de fezes (n=143) destes Felidae, no período determinado, foram *Toxascaris leonina* (16%), *Cystoisospora* spp. (2%) e outros parasitas não especificados: coccídeos (8%), nemátodes (3%), Toxocaridae (2%), oocistos esporulados (1%) e não esporulados (1%) e flagelados (1%). Além disso, no soro de alguns tigres de Sumatra testados, foi detetado *Toxoplasma gondii* (80%, n=5).

A presença parasitária foi comparada e discutida em cada espécie animal. Foi também comparada nas quatro diferentes estações do ano, bem como a sua evolução ao longo dos anos. A prevalência parasitária foi mais alta durante o verão para os leões Sul-Africanos, durante o outono para os jaguares e os leopardos do Sri Lanka, e durante o inverno para os tigres de Sumatra.

O nível de concordância entre a sensibilidade das técnicas de flutuação e de método direto, para detetar parasitas nos Felidae do Zoo, foi avaliado neste estudo, concluindo-se que as técnicas de flutuação foram mais sensíveis na deteção de amostras positivas, apresentando 84% de sensibilidade, enquanto os métodos diretos foram apenas 48% sensíveis.

Palavras-chave: Análise fecal, *Panthera* spp., Parasitas Gastrointestinais, Sazonalidade, Jardins Zoológicos

RETROSPECTIVE STUDY ON GASTROINTESTINAL PARASITES OF 28 FELIDAE FROM THE BARCELONA ZOO, FROM 1990 TO 2024

Abstract

Zoos have become valuable in the maintenance and breeding of a variety of wild animals, amongst them vulnerable and critically endangered species, thus working towards their conservation and prevention from extinction. This great responsibility of housing such valuable animals includes the prevention and curing of diseases, as housing animals away from their natural habitats involves the control of several predisposing conditions that facilitate disease and infections, sometimes from agents that are not present in their natural habitat, and against which the animal is not able to defend itself.

Parasitic infections have been reported in many different zoos and are a big influence in the health of animals in these establishments. Their prevention and management are essential for the animals' wellbeing.

Four species of Felidae from the Barcelona Zoo were investigated for gastrointestinal parasite load, from March 1990 to February 2024. The study consisted on the retrospective analysis of the Zoo's database regarding the parasites of twelve Southern African lions (*Panthera leo melanochaita*), four jaguars (*Panthera onca*), seven Sri Lankan leopards (*Panthera pardus kotiya*) and five Sumatran tigers (*Panthera tigris sumatrae*). The parasites present in the faecal samples (n=143) in these Felidae, in the given timespan, were *Toxascaris leonina* (16%), *Cystoisospora* spp. (2%) and other unspecified parasites: Coccidia (8%), nematodes (3%), Toxocaridae (2%), sporulated (1%) and non-sporulated oocysts (1%) and flagellates (1%). Also, in the serum of some tested Sumatran tigers, *Toxoplasma gondii* (80%, n=5) was detected.

The parasitic presence was compared and discussed for each animal species. It was also compared for the four different seasons of the year, as well as its evolution throughout the years. The parasitic prevalence was higher during summer for the Southern African lions, during autumn for the jaguars and the Sri Lankan leopards, and during winter for the Sumatran tigers.

The agreement level between flotation and direct method techniques' sensitivity, to detect parasites in the Zoo's Felidae, was evaluated in this study, with the conclusion that flotation techniques were more sensitive in detecting positive samples, showing 84% sensitivity, whereas direct methods were only 48% sensitive.

Keywords: Faecal analysis, *Panthera* spp., Gastrointestinal Parasites, Seasonality, Zoological Gardens

ESTUDO RETROSPETIVO SOBRE OS PARASITAS GASTROINTESTINAIS DE 28 FELIDAE DO ZOO DE BARCELONA, DE 1990 A 2024

Resumo alargado

Os zoológicos tornaram-se valiosos na reprodução e cuidado de uma variedade de animais selvagens, incluindo algumas espécies vulneráveis e criticamente em perigo de extinção, contribuindo assim para a sua conservação e prevenção de extinção. Esta grande responsabilidade de alojar animais tão valiosos inclui a prevenção e cura de doenças, entre elas infeções parasitárias, pois alojar animais fora dos seus habitats naturais implica o controlo de várias condições predisponentes, como o stress, que facilitam doenças e infeções, por vezes causadas por agentes que não estão presentes no seu habitat natural, e contra os quais o sistema imunitário do animal não se consegue defender. Estas doenças podem influenciar significativamente a saúde destes animais.

Infeções parasitárias já foram reportadas em vários jardins zoológicos e têm uma grande influência na saúde dos animais nesses estabelecimentos. A sua prevenção e gestão são essenciais para o bem-estar destes animais.

Este estudo investigou quatro espécies de Felídeos no Jardim Zoológico de Barcelona relativamente aos seus parasitas gastrointestinais, entre março de 1990 e fevereiro de 2024. O estudo consistiu na análise da base de dados do zoológico relativamente aos parasitas de 12 leões Sul-Africanos (*Panthera leo melanochaita*), quatro jaguares (*Panthera onca*), sete leopardos do Sri Lanka (*Panthera pardus kotiya*) e cinco tigres de Sumatra (*Panthera tigris sumatrae*). Muitos destes animais foram testados repetidas vezes ao longo dos anos, sendo considerada cada testagem como um caso diferente. Cada caso consistiu em um animal testado, a espécie do respetivo animal, a data na qual a amostra de fezes ou sangue foi colhida, o exame realizado para analisar a amostra em busca de parasitas e se a amostra era ou não positiva para parasitas. Foram então analisados 49 casos de leões Sul-Africanos, 23 casos de jaguares, 41 casos de leopardos do Sri Lanka e 33 casos de tigres de Sumatra, dando um total de 146 casos. Dos 146 casos, em 141 foram apenas analisadas amostras de fezes dos animais, em dois casos foram analisadas amostras de fezes e de soro, e em três casos foram analisadas apenas amostras de soro, sendo então 143 amostras de fezes e cinco amostras de soro.

Os parasitas presentes nas amostras de fezes (n=143) desses Felídeos, no período mencionado, foram *Toxascaris leonina* (16%), *Cystoisospora* spp. (2%) e outros parasitas dos quais não se identificou a espécie: Coccidia (8%), nematodes (3%), Toxocaridae (2%), oocistos esporulados (1%), oocistos não esporulados (1%) e flagelados (1%). Além disso, no soro de alguns tigres de Sumatra testados, foram detetados anticorpos de *Toxoplasma gondii* (80%, n=5). *Toxascaris leonina* foi o parasita que foi encontrado mais recorrentemente nestes

animais, este é um parasita difícil de erradicar, devido à sua resistência ambiental e facilidade de reinfeção dos animais por um ambiente contaminado.

A presença de parasitas foi comparada e discutida em cada espécie animal. Nos leões Sul-Africanos verificou-se uma prevalência parasitária de 53% (26/49), sendo que foram apenas afetados por *Toxascaris leonina* (35%), e por Coccidia (12%) e Toxocaridae (6%) não especificados; nos jaguares verificou-se uma prevalência de 26% (6/23), e mostraram uma maior variedade parasitária, sendo afetados por *Toxascaris leonina* (9%), *Cystoisospora* spp. (9%) e outros parasitas não especificados: Coccidia (13%), nematodes (9%), flagelados (4%), oocistos esporulados (4%) e oocistos não esporulados (4%); nos leopardos do Sri Lanka verificou-se uma prevalência parasitária de 15% (6/41), e infeção por *Toxascaris leonina* (5%), Coccidia (5%) e nematodes (5%); nos tigres de Sumatra verificou-se uma prevalência parasitária de apenas 10% (3/30), sendo que estavam infetados por *Cystoisospora* spp. (3%) e *Toxascaris leonina* (7%). Nos tigres de Sumatra verificou-se também a presença de anticorpos contra *Toxoplasma gondii* (80%) através da análise de amostras de soro (n=5). A análise para a identificação de anticorpos contra *Toxoplasma gondii* não foi realizada nas outras três espécies, e sendo um parasita difícil de detetar por meios coprológicos, não se pode descartar a hipótese de estes animais terem também contactado com o parasita.

Nesta dissertação avaliou-se a prevalência parasitária dos animais nas quatro estações do ano. A prevalência parasitária foi mais elevada no verão para os leões Sul-Africanos (76%), no outono para os jaguares (43%) e leopardos do Sri Lanka (29%), e no inverno para os tigres de Sumatra (23%), sendo também esta a estação em que ocorreram os únicos casos com parasitas em amostras de fezes, nos tigres de Sumatra. Foi realizado o teste Qui-quadrado para se avaliar se haveria relação entre os períodos em que os parasitas foram detetados e as estações do ano. O valor de p foi <0.05 apenas para a presença de Toxocaridae nos leões Sul-Africanos, e para a sazonalidade de todos os parasitas no mesmo animal, o que indica que houve uma relação entre a presença destes parasitas e a estação do ano em que ocorreram. Em relação a todos os outros parasitas, não se observou uma relação com a estação do ano.

Para além das estações do ano, realizou-se uma análise estatística na forma de gráficos de barras, dos parasitas presentes nas quatro espécies de Felidae ao longo dos diferentes intervalos de 5 anos, de 1990 até 2024. É importante ter em conta que antigamente o registo destes testes era feito em papel, sendo possível que, ao passar toda a informação para a base de dados em formato digital do Zoológico de Barcelona, alguma informação poderá ter sido perdida. A análise estatística aqui realizada foca-se nos resultados registados e não necessariamente nos possivelmente realizados. Nos intervalos de anos 2010-2014 e 2015-2019 não existe registo de análise de amostras de fezes nestes animais, possivelmente devido à razão mencionada neste parágrafo.

O leão Sul-Africano esteve infetado recorrentemente de 1990 a 2009, e depois outra vez no período de 2020-2024, sendo de notar que não há registo de testes realizados entre 2010 e 2019. O Zoológico tem registo de infeções do jaguar apenas nos períodos de 1990-1994 e de 2020-2024, sendo que em 1995-1999 apenas se realizaram duas análises de amostras, ambas negativas, e nos restantes anos não há registo de amostras testadas. Os registos do leopardo do Sri Lanka mostraram infeções em 1990-1994, 2005-2009 e 2020-2024, sendo que em 1995-1999 se testaram cinco amostras que resultaram negativas e nos restantes anos não há registos de amostras testadas. Os tigres de Sumatra tiveram infeções em 1995-1999 e 2020-2024, em 2005-2009 foram testadas seis amostras de fezes negativas para parasitas. Também em 2005-2009, nos tigres de Sumatra, foram testadas quatro amostras de soro para a presença de anticorpos de *Toxoplasma gondii*, que revelaram resultados positivos, e uma outra amostra foi testada em 2015-2019, que resultou negativa para anticorpos contra *Toxoplasma gondii*.

O nível de concordância entre a sensibilidade das técnicas de flutuação e dos métodos diretos para detetar parasitas nos Felídeos do zoológico foi avaliado neste estudo. O valor do Kappa de Cohen foi de 0.372, um valor que indica um nível de concordância mínimo (0,21-0,39) entre estes dois testes, sendo então dedutível que um é mais sensível que o outro. A significância estatística foi de <0.001, o que nos indica que estes resultados são muito provavelmente significativos e não obtidos por coincidência. Ao observar os resultados concluiu-se que as técnicas de flutuação foram mais sensíveis na deteção de amostras positivas, sendo que das 31 amostras positivas identificadas, a flutuação detetou 26 positivos (entre eles, 16 casos foram negativos em métodos diretos) demonstrando então uma sensibilidade de 84%, enquanto os métodos diretos detetaram 15 dos resultados positivos (dos quais apenas 5 foram negativos em flutuação) apresentando uma sensibilidade de 48%.

Palavras-chave: Análise fecal, *Panthera* spp., Parasitas Gastrointestinais, Sazonalidade, Jardins Zoológicos

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1. Introduction

Zoos have become valuable in the maintenance and breeding of a variety of wild animals, amongst them vulnerable and critically endangered species, thus working towards their conservation and prevention from extinction. This great responsibility of housing such valuable animals includes the prevention and treatment of diseases, as housing animals away from their natural habitats involves the control of predisposing conditions that facilitate disease and infections, sometimes from agents that are not present in their natural habitat, and against which the animal is not able to defend itself.

Parasitic infections have been reported in many different zoos and are a big influence in the health of animals in these establishments. The prevention and management of these infections are crucial for the well-being and survival of these animals (Panayotova-Pencheva 2013).

In this dissertation we will talk about parasitic screening and treatment in zoological establishments, and about parasites that occur in each of the four species of Felidae present in the Barcelona Zoo: *Panthera leo bleyenberghi/melanochaita* (read the Southern African lion chapter), *Panthera onca*, *Panthera pardus kotiya* and *Panthera tigris sumatrae*. In this study we prioritize the parasites found to have infected these animals, according to the database of the Barcelona Zoo, as well as the most frequent parasites in these animals, according to extensive research on various articles of reported parasites in these four Felidae species.

2. Internship reports

2.1 Report of the internship in Rescue Center Costa Rica

The trainee worked in the Rescue Center Costa Rica for nearly three months, from September 20th until December 13th, 2022, during which time she completed 652 hours of work.

This rescue center was founded during the pandemic, in 2020, by Vanessa Lizano, Founder and President, and has since been run by Vanessa and Laura Lizano, the COO (Chief Operating Officer). The veterinary team has had Dr. Raquel Meneses from the beginning and has since grown, with Dr. Alejandra Aranda working side by side with her. All of the rescue's staff, and many volunteers from all over the world, have worked relentlessly to grow and improve this institution's infrastructures to better accommodate and follow the mission to help as much wildlife as possible, either from illegal trading or illegal possession, rehabilitating injured animals or raising orphaned animals. This institution's goal is to rehabilitate and release all animals that arrive to the rescue, however, due to injuries, chronic diseases, or animals being accustomed to humans because of a previous life as pets, it is not always possible to release every animal, as many need human support for the rest of their lives and would not survive in the wild. Therefore, Rescue Center Costa Rica has spacious enclosures, with forms of enrichments, to accommodate the needs of the animals that require to remain in their care for life, as well as the needs of animals that temporarily stay there.

The rescue houses a variety of animals, amongst them mammals (sloths, racoons, coatis, opossums, ungulates, carnivores, primates and others), birds (toucans, many species of parrots, parakeets, small birds and birds of prey) and reptiles (turtles). These animals are usually taken care of by the coordinators and different teams of volunteers, with one team dedicated solely to sloths, as there's usually more of these animals in the rescue, and they require specialized care. Besides the housed animals, the rescue receives many animals for rehabilitation, handled and evaluated by the veterinary team, that also assists permanent animals' care.

During her internship, the student worked in the veterinary team with Dr. Raquel Meneses, Dr. Alejandra Aranda, Dr. Giovana Miranda, as well as with biologist Daniela Cordero, veterinarian nurses Erin Taylor and Camille Lasbleiz, and another intern, Giullia Ribeiro. The vet team was also aided by volunteers that would stay in the rescue from one week to several months. There, the student had many responsibilities and assisted in numerous procedures and caring for the animals, such as:

- Handling and restraining mammals, reptiles and birds.
- Administering daily oral medications directly with syringe or through food to various animals, including an ocelot (*Leopardus pardalis*), a margay (*Leopardus wiedii*), a blue-and-yellow macaw (*Ara ararauna*) and other mammals.
- Receiving new animals at the rescue, registering them, doing physical evaluation and cleaning if necessary, administering fluid therapy and, if neonates or juveniles, calculate milk or formula dosages and frequency of feeding.
- Daily syringe milk-feeding and monitoring of growth of various neonates such as sloths (two-toed sloths *Choloepus hoffmanni* and three-toed sloths *Bradypus variegatus*), squirrels (*Sciurus variegatoides*), racoons (*Procyon lotor*) and opossums (*Didelphis marsupialis*), as well as specialized feeding of neonate owls (Strigiformes) and parrots (Psittaciformes). The feeding and monitoring of neonates would occur from every 3h to periods of every 6h, many times including night feedings at 23h and 2h.
- Daily solid food feeding and monitoring of growth of various juveniles, and feeding of adult animals, such as parrots, racoons, opossums, owls, a porcupine (*Sphiggurus mexicanus*), among others.
- Daily monitoring and enclosure cleaning for various animals.
- Attending to emergencies, amongst them animals in need of urgent surgery or shock patients in need of fluids, oxygen and monitoring.

- Participating in surgeries and monitoring anaesthesia, such as orchiectomy surgeries in a racoon and rabbit (Lagomorpha), and mandibula reconstruction surgery in a Central American snapping turtle (*Chelydra rossignonii*).
- Aiding in imaging diagnosis, namely x-rays and ultrasounds.
- Observing and participating in the physical examination of the three carnivores in the rescue (ocelot, margay and coyote (*Canis latrans*)), which included dart anaesthesia and subsequent monitoring, general physical evaluation, blood, urine and fur sampling, cardiac and pulmonary auscultation, ultrasounds and animal monitoring after the administration of reversals.
- Participating in training classes for animal restraint, blood sampling, laser therapy and necropsies.
- Performing nebulization therapy.
- Performing euthanasia in suffering animals with no chance of recovery.
- Taking initiative in scheduling the carnivore feedings for the week.

The intern also participated in the training of volunteers and team management for the vet team. As part of the internship, the intern was requested to do two presentations, one of them on a project that would benefit the rescue. The intern did a presentation on a topic of her choosing, in this case, “Rodenticides in Wildlife”, because there was a suspicion of rodenticides being the cause of death of a couple of rescued orphaned juvenile squirrels, that would’ve had contact with the substance before arriving at the rescue. The intern also worked on a dietary book for all the animal species in the hospital, and for possible animals that could arrive, with attention to the age (juvenile/adult) and situation of the animals (pre-release situations).

2.2 Report of the internship in the Barcelona Zoo

The trainee worked in the Barcelona Zoo for three months, from February 20th until May 19th, 2023. This institution holds a collection of around 3500 animals of 231 different species, including mammals, birds, reptiles, amphibians, fishes and invertebrates. The medical and research area incorporated a surgery room, a pharmacy, an office, and a necropsy room.

The veterinary team was composed of two veterinarians, Dr. Hugo Fernández and Dr. Vanessa Almagro; two veterinary assistants, Aurora Pazos and Begoña Teruel; and a pathology intern, Laura Fuentes, as well as two trainees at a time, the student and Diana Faim, and for the second half of the internship, the student and Giulia Romani.

During the time spent in the institution, the trainee observed and assisted 419 cases, of which 340 were birds, most of which due to routine vaccination of all the birds in the zoo. Of the 419, 34 were clinical cases, 330 were management cases and 44 were necropsies. The remaining were cases registered solely for sample taking or for mortality. The trainee gained a lot of knowledge in many different areas by:

- Handling and restraining mammals, reptiles, birds and amphibians.
- Assisting on surgeries and minor procedures, namely a tooth and abscess removal on a red-necked wallaby (*Notamacropus rufogriseus*) and a hormonal implant replacement in a red-capped mangabey (*Cercocebus torquatus*).
- Discussing, calculating, and implementing sedation and anaesthesia protocols in small and in big animals, such as in Iberian ribbed newt (*Pleurodeles waltl*) and on an African lion (*Panthera leo*).
- Aiding in and/or discussing diagnostic imaging cases, done with x-rays and ultrasounds.
- Assisting in vaccination protocols in several avians.
- Administering drugs on collection animals, for example on a crested porcupine (*Hystrix cristata*).
- Withdrawing blood, for testing purposes, from a marginated tortoise (*Testudo marginata*) and from a mhorr gazelle (*Nanger dama*).
- Undertaking assisted feeding in a Northern caiman lizard (*Dracaena guinensis*).
- Observing and discussing chipping and animal sexing.
- Performing and thoroughly documenting necropsies, as well as taking tissue and fluid samples for routine testing and/or storage.
- Attending dart preparation, handling and cleaning training.
- Participating in discussions of clinical thinking and diagnosing of several different species.

The trainee was also always ready to aid in any other task, including the sanitizing of the surgery room and used materials.

The trainee undertook three presentations and discussions about the following articles with the veterinary team: “Standing sedation with xylazine and reversal with yohimbine in juvenile Asian elephants (*Elephas maximus*)” (Jansson et al. 2021), “Multi-sensor biologgers and innovative training allow data collection with high conservation and welfare value in zoos” (English et al. 2023) and “Hepatitis E virus and related viruses in wild, domestic and zoo animals: A review” (Spahr et al. 2017).

She also did a final oral presentation of a report case on a red panda (*Ailurus fulges*) with a squamous cell carcinoma. The presentation included a descriptive and statistical analysis of all the cases the trainee took part of during her time in the zoo establishment.

3. Literature review

3.1 Felidae

Felidae is a family within the Carnivora order that consists of 38 species.They are similar in some behavioural and morphological characteristics such as having rounded flat faces, facial whiskers and large eyes and ears (Kitchener et al. 2017; Lamberski 2015). They have the widest weight range of all carnivore families, weighing between 1 to 300 kg (Lamberski 2015). Felids are also naturally endemic to the whole world except for Australia as well as the poles (O'Brien and Johnson 2005). Felidae divides into two main subfamilies: Felinae and Pantherinae, with the exclusion of cheetahs, which belong to an early divergence from the felid line, in the subfamily Acinonychinae (Pocock 1917 as quoted by Kitchener et al. 2017). Pantherinae have a distinctive trait of a different larynx with long elastic vocal cords, that allows them to roar but not purr, unlike Felinae, that cannot roar, but can purr . Within Pantherinae are the genus *Neofelis* and *Panthera* (Kitchener et al. 2017).

This dissertation focuses on four big felines in the *Panthera* genus, namely *Panthera leo melanochaita*, *Panthera onca, Panthera pardus kotiya* and *Panthera tigris sumatrae,* from here on referred to as Felidae.

3.2 Felidae in Zoological establishments

Wide ranging carnivores are sensitive animals, easily affected by captivity, demonstrating stereotypical behaviours when placed in poor environments with insufficient enrichment (Clubb and Mason 2003).

Captive animals' requirements for wellbeing include personalized diets, appropriate spacious enclosures, enrichment, and specialized medical care. This care includes the vigilance of faeces and enclosure samples for routine parasitic screening, aiding the control of parasitosis.

3.3 Parasitology in Zoological establishments

Parasitological findings can come from routine checks or specific testing in sick animals that are suspected to be parasitized. Animals infected with gastrointestinal parasites may display nonspecific clinical signs such as looking weak and debilitated, losing weight, and excreting abnormal faeces (Abou-Madi 2013; Kassa et al. 2016).

3.3.1 Faecal examination protocols in Zoological establishments

Parasites and their ova or oocysts are most often difficult to observe with the naked eye, and the microscope is an essential tool. Some coprological methods involve the observation of a faecal sample directly smeared on the slide, others include faeces processing to obtain egg concentration and minimal slide contamination (Foreyt 2001; Abou-Madi 2013).

Direct techniques A common direct technique is the direct smear or wet mount, in which a sample of faeces is directly smeared in a slide and mixed with a drop of saline solution or water, or even Lugol's iodine (WCVM-US 2021), and covered with a coverslip, ready to be observed under the microscope (Abou-Madi 2013).

Flotation techniques Flotation techniques are very commonly used in the detection of nematode and cestode eggs and protozoan cysts. Trematode eggs are usually too heavy to be detected and require a more concentrated solution to be able to float (Abou-Madi 2013; MAFF (2006) as quoted by Kassa et al. 2016). The downside to increasing the solution's concentration is that protozoan and specific nematode larvae can be distorted and become too difficult to identify. To prevent this problem, it is recommended to perform two flotation techniques, one with zinc sulphate to detect *Giardia* sp. trophozoites/cysts, nematode larvae and protozoan (oo)cysts, and a saturated sugar flotation to detect heavier eggs (Abou-Madi 2013; Zajac and Conboy 2012 as quoted by Ballweber et al. 2014).

Common flotation techniques include the centrifugal technique and the passive flotation technique. The former being the centrifugation of the faecal material diluted in a salt solution, which better sediments faecal material that is not relevant for the observation and identification of eggs, separating it from the eggs that rise to the top of the solution. The latter being a technique that simply relies on gravitational force to separate the irrelevant faecal material from the parasites and parasitic eggs and cysts (Abou-Madi 2013; Kassa et al. 2016).

3.3.2 Prevention and deworming protocols in Zoological establishments

Some zoos have a protocol to administer deworming drugs every three months to prevent parasites (Aviruppola et al. 2016). This can lead to parasitic resistance to the drugs, and greater harm to fall upon the treated animal. Ideally, coprological analysis to determine whether an animal has parasites would be the best route to decide if treatment is needed (Abou-Madi 2013; Aviruppola et al. 2016). The European Association of Zoos and Aquaria (EAZA) recommends the faecal examination of jaguars to occur every 4 months with both parasitological and bacteriological testing (Biddle 2022). Pharmacological care is not always necessary. Even if the tested animal does have parasites, a medical evaluation and observation should determine if treatment is truly needed, as many animals have natural resistance to parasites, and don't fall into illness when affected (Mukarati et al. 2013; Aviruppola et al. 2016). So, antiparasitic drugs should only be administered to those suffering clinically from heavy parasitic infections. In cases of highly contaminated enclosures where animals are at risk of or continue to be re-infected, Biddle (2022) refers blind treatments at set intervals as a possible management protocol for jaguars.

Table 1: Drugs for the treatment of endoparasites in jaguars

Drug	Parasite class	Application	Dosage
Pyrantel	Nematodes	PO	3-5 mg/kg SID for 3-5 days.
Fenbendazole	Nematodes, cestodes	PO	10mg/kg SID for 3-5 days
	Giardia	PO	50mg/kg SID 10-14 days
Febantel*	*Giardia*	PO	56 mg/kg SID 5 days
Ivermectin	Nematodes	SC / PO	0,2 mg/kg single application 0,1-0,2 mg/kg monthly for *Dirofilaria* prophylaxis
Praziquantel	Cestodes	SC / PO	5,5-6,6 mg/kg single application
Milbemycine oxime	Nematodes	PO	2mg/kg single application 2mg/kg monthly for *Dirofilaria* prophylaxis
Moxidectine	Nematodes	Topical	1mg/kg monthly for *Dirofilaria* prophylaxis**
Sulfadimethoxine	Coccidia	PO	50mg/kg SID for 10-14 days***
Toltrazuril (oral formulations)	Coccidia	PO	30mg/kg SID: 3 days on, 4 days off, 3 days on****

* combo preparations to be used based on febantel contents
** combo preparations to be used based on moxidectine contents (Arther *et al*, 2005). Watch with canine products, moxidectine overall 1/4th of that in a feline product!
*** (Plumb, 2011)
**** off label use in domestic cats (Hirose *et al*, 2015)

Biddle R, editor. 2022. Jaguar (*Panthera Onca*) EAZA Best Practice Guidelines, pp. 57, table 4.

Besides the usage of antiparasitic drugs, some protocols are essential in the riddance and prevention of parasitosis (Abou-Madi 2013). These protocols include the removal of susceptible animals from contaminated areas and grouping them according to susceptibility to parasites, advanced hygiene protocols ("e.g. wearing gloves, washing hands, cleaning food and water bowls once or twice daily, repositioning the bowls so that the animal does not defecate in them, promptly removing manure and contaminated material" (Abou-Madi 2013, p.485), proper washing and disinfection of materials used in the cleaning, and usage of specific clothing and footbaths are also beneficial to prevent the propagation of parasites outside the enclosures), pest control to avoid parasite transmission through paratenic hosts, and habitat adaptation, like draining humid soils (Abou-Madi 2013; Bowman 2021).

3.3.2.1 Prevention and deworming protocols for Felidae in the Barcelona Zoo

In the Barcelona Zoo, the African lions are medicated every three months with a protocol that alternates between oral fenbendazole and ivermectin. The Sumatran tiger, Sri Lankan leopard and the jaguar are not medicated in a preventive way, as the parasitic load does not require it (personal communication, Dr. Vanessa Almagro, 2024).

3.4 Felidae species present in the Barcelona Zoo

3.4.1 Southern African Lion (*Panthera leo melanochaita*)

The Angolan lion (*Panthera leo bleyenberghi*) was an endangered subspecies of the African lion species (Steinmetz et al. 2006), distributed along Southwestern Africa in Namibia, Angola, Democratic Republic of Congo, Western Zambia, Western Zimbabwe, and Northern Botswana (Kitchener et al. 2017).

Panthera leo bleyenberghi was first reported by Lonnberg (1914), at the time referred to as *Felis leo bleyenberghi*. In 1939 the species was recognised as valid by Glover Allen (1939) in the Bulletin of the Museum of Comparative Zoology at Harvard College. More recently, as quoted by Kitchener et al. (2017), Wozencraft (2005) recognised 11 lion subspecies, amongst them the taxon of *Panthera leo bleyenberghi*.

Later, in 2017, Kitchener et al. (2017) analysed different molecular studies, concluding that only 2 subspecies of lion should be identified: *Panthera leo leo* and *Panthera leo melanochaita*. Revising the distribution of both subspecies in Kitchener et al. (2017), it was possible to conclude that *P. l. bleyenberghi* is now considered *P. l. melanochaita*.

In the Barcelona Zoo log, this species is registered as *Panthera leo bleyenberghi*, however, given the update of the subspecies denomination, it will be referred to as *Panthera leo melanochaita* during the materials, results, discussion and conclusion of this dissertation.

All parasites in lions discussed in this dissertation's literature review are from *P. l. melanochaita* or lions deduced as *P. l. melanochaita* from comparing the geographical location of the reported free-living animals, to the information given by Kitchener et al. (2017) regarding this subspecies' distribution.

The African lion (*Panthera leo*) is the largest felid species in the African continent, *P. l. melanochaita* have weight variations of 145-225 kg in males and 83-167 kg in females (Smuts et al. 1980) varying according to the feeding, genetics, and habitat (Bothma and Walker 1999). In 2016, *Panthera leo* was considered vulnerable by The IUCN Red List of Threatened Species (Bauer et al. 2016).

These are social animals, living in prides of various sizes, with member numbers ranging from two to 39 individuals. As the pride size varies, so does the size of their territory. These nocturnal hunters have a varied diet depending on the prey present within their habitat, majority of which is hunted, some scavenged (Bothma and Walker 1999).

Their prey sizes range from 20 kg to 800 kg or even more, depending on the size of the pride. They consume both flesh and internal viscera, especially intestines. When water availability is scarce, lions will also chew on plants such as *Sansevieria* sp., with the purpose of hydration from its roots (Bothma and Walker 1999). Given both the prey and carcass part preference variety, and adding to the equation soil and environmental exposure, these animals are subjected to a large parasitic diversity in the wild.

3.4.2 Sumatran Tiger (*Panthera tigris sumatrae*)

Panthera tigris sumatrae is a subspecies of the Indonesian tiger, first proposed by Pocock (1930). *Panthera tigris*' intraspecific taxonomy has been up for debate. Kitchener et al.

(2017) revised the taxonomy status, and based on molecular and morphological characteristics, they concluded that the nine existing subspecies (eight described by Wozencraft (2005) and one by Luo et al. (2004) as quoted by Kitchener et al. (2017)), should be reduced to two: *Panthera tigris tigris* and *Panthera tigris sondaica*. Liu et al. (2018) performed analysis using whole-genome sequencing and concluded that there are six subspecies of *P. tigris*, including *Panthera tigris sumatrae*.

The tiger is the largest feline to populate the Earth, however, the Sumatran tiger is one of the smallest in the species, with males weighting between 100 to 140 kg and females weighting between 75 to 110 kg (Mazák 1981).

This subspecies' distribution consists mainly of lowland and deep forest areas in the Indonesian island of Sumatra (Luskin et al. 2017). They populate areas with dense forest and prefer high elevations areas with suitable vegetation cover at ground level (Sunarto et al. 2012). Their habitat and population have been threatened the most by deforestation and plantations of palm oil and acacia, illegal trade and poaching of both tigers and their prey (Lynam 2010; Oswell 2010; Sunarto et al. 2012). In 2022, their classification in The IUCN Red List of Threatened Species was listed as endangered (Goodrich et al. 2022).

Tigers hunt a wide variety of prey, such as wild boars (*Sus scrofa*), mouse deer (*Tragulus* spp.), argus pheasants (*Argusianus argus*) and pigtail macaques (*Macaca namistrina*) (O'Brien et al. 2003).

3.4.3 Sri Lankan Leopard (*Panthera pardus kotiya*)

The Sri Lankan leopard was first described by Deraniyagala (1949), as quoted by Dowling (1951). It is naturally located solely in Sri Lanka. These animals are highly adaptable to their environment, preferring areas that are more elevated and further from humans, however roaming territory quite near highly populated areas (Kittle et al. 2018). If their territory is invaded by human settlements, they are likely to remain and hunt domestic animals, rather than relocating to find their natural prey (Shehzad et al. 2015; Kittle et al. 2018).

The Sri Lankan leopard is the biggest predator on the island, weighting between 29 to 77 kg, with females averaging 29 kg and males averaging 77 kg, displaying wide sexual dimorphism (Eisenberg and Lockhart 1972; Wijeyeratne 2016; Miththapala 2017). Some of their prey includes toque macaques (*Macaca sinica aurifrons*), barking deer (*Muntiakus muntjac*), wild boars (*Sus scrofa*) and porcupines (*Hystrix indica*) (Kittle et al. 2014).

This animal has been considered endangered by Kittle and Watson (2015), as quoted by Stein et al. (2020). Their main threats are habitat loss paired with reduction of prey and hunting caused by conflict or trophy hunting (Nowell and Jackson 1996; Packer et al. 2010; Jacobson et al. 2016).

3.4.4 Jaguar (*Panthera onca*)

In 2005 there were nine subspecies of jaguar, recognised by Wozencraft (2005), as quoted by Kitchener et al. (2017). However, Kitchener et al. (2017) have since proved that there are no significant genetic differences amongst jaguar subspecies, concluding that it is a monotypic species, meaning that there's only one subspecies.

It is distributed from southern Arizona and New Mexico in the United States of America, through Central America to northern Argentina (Carrillo 2007), most abundant in the presence of water, including the rainforest, seasonally flooded swamp areas and Pampas grassland, but also thorn scrub woodland and dry deciduous forest (Nowell and Jackson 1996).

This animal is considered, by The IUCN Red List, a near threatened species (Quigley et al. 2017). Its main threats are habitat loss and fragmentation, along with illegal hunting of the jaguar and the hunting of its prey (Ceballos et al. 2002), but also the retaliatory or protective hunting to prevent livestock losses (Zimmermann et al. 2005).

Jaguars are solitary and opportunistic hunters with a varied diet (Emmons 1987; Hayward et al. 2016). A 2016 study performed by Hayward et al. (2016), summarized diet preferences of jaguars from various previous studies, indicating a prey size range of 1 to 130 kg, with preferences for capybara (*Hydrochoerus hydrochaeris*), wild pig (*Sus scrofa*), common caiman (*Caiman crocodilus*), collared peccary (*Pecari tajacu*), nine-banded armadillo (*Dasypus novemcinctus*), giant anteater (*Myrmecophaga tridactyla*) and white-nosed coati (*Nasua narica*).

3.5 Most common parasites of Felidae

In this chapter we will elaborate on the most common parasites from the specific felid subspecies in this study, found in various articles, as well as the parasites described in the results from this dissertation.

In European zoos, the most common parasites are helminths, followed by protozoa, nematodes being the most common helminths, according to Panayotova-Pencheva (2013). This relates to the parasites' life cycles, as geohelminths (helminths that perform a specific stage of their life cycle on soil) have better conditions to re-infect caged animals than biohelminths (which include most cestodes (Deep et al. 2023) and all trematodes (Ayi 2007)), that need an intermediate host, not always present in captivity (Panayotova-Pencheva 2013).

3.5.1 Protozoans

3.5.1.1 *Giardia* spp.

Giardia is a protozoan genus from the phylum Fornicata, order Giardiida, and family Giardiidae (Taylor et al. 2016). This gastrointestinal parasite infects humans and various animals, amongst them mammals, birds and even amphibians (Bowman 2021).

This parasite has been reported in free-ranging *Panthera leo* (Bjork et al. 2000; Kruszka and Harris 2020), captive lions in Zimbabwe (Mukarati et al. 2013) and in zoos in China (Zou et al. 2022). It has also been reported to affect 30% of the captive *Panthera onca* in the Municipal Pomerode Zoo in Brazil (Müller et al. 2005).

Life cycle This parasite has two main life cycle stages: cyst and trophozoite. The cyst is the environmental stage, resistant to external factors and infectious to the host. The trophozoite stage occurs in the gastrointestinal tract of the host (Geurden and Claerebout 2010).

The cyst enters the host through oral ingestion and releases the trophozoite once it reaches the intestine. The trophozoite attaches itself to the epithelial cells of the duodenum and jejunum (jejunum and ileum in cats (Kirkpatrick 1986; Bowman 2021)) with its ventral adhesive disk, therefore, beginning the colonization of the intestinal mucosa (Geurden and Claerebout 2010). *Giardia* multiplies by binary fission in the lumen of the small intestine. The encystation of the trophozoites happens once they are exposed to biliary salts in the jejunum. Finally, the cysts, containing two potential trophozoites (Bowman 2021), are excreted in the faeces and are immediately infectious (Geurden and Claerebout 2010).

Clinical signs The main clinical sign is chronic diarrhoea caused by the parasite's interference in the intestinal absorption process (Kirkpatrick 1986). The diarrhoea will either be continuous or intermittent. Usual appearance of the faeces is mucoid, pale, and soft, as well as having a strong odour (Kirkpatrick 1986; Bowman 2021). A less common sign is the weight loss (Kirkpatrick 1986). Felidae may also experience no signs, or abdominal discomfort from slight abdominal pain to severe cramping (Tangtrongsup and Scorza 2010).

Diagnosis Although cysts are more often found in faeces, because trophozoites not only become cysts when exposed to biliary salts but are also more sensitive outside the hosts body and quickly die, trophozoites can still be found in direct smears of diarrhoeal faeces but are hardly found in formed stool (Bowman 2021). Zinc sulphate flotation technique (with specific gravity 1.18) can be used to find cysts. Other flotation methods such as sucrose distort the cysts and the results (Bowman 2021). Both the direct faecal smear and the zinc sulphate flotation can be stained with a drop of Lugol's solution of iodine to better identify the trophozoites and cysts (Bowman 2014).

The possibility of not finding cysts in an infected animal does exist, and other diagnose alternatives are *Giardia* Antigen detection kits and IDEXX SNAP for *Giardia* (Garcia and Shimizu 1997; Carlin et al. 2006).

Prevention and Control Guaranteed access to clean water and providing proper hygiene of the enclosure is a necessity (Tangtrongsup and Scorza 2010). This includes daily removal of faeces and steaming or disinfecting the infected surfaces with quaternary ammonium compounds (Tangtrongsup and Scorza 2010). Isolation, if possible, of animals with diarrhoea until treatment is completed and coprological exams demonstrate a successful treatment, will prevent propagation of the parasite.

Treatment Different studies performed in domestic cats have observed the efficacy of different drugs against giardiosis. Metronidazole has proven high efficacy rates, however, neurotoxicity is possible, even if not always registered, requiring animal monitoring during high dosage or chronic treatment (Scorza and Lappin 2004; Tangtrongsup and Scorza 2010; Gruffydd-Jones et al. 2013). It has also been successfully used in a European wild cat (*Felis silvestris*) (Peisert et al. 1983).

The combination of febantel, pyrantel and praziquantel (Scorza et al. 2006; Tangtrongsup and Scorza 2010) has shown positive results in giardiosis therapy. Both this combination or fenbendazole are good choices if the animal is co-infected with nematodes or cestodes (Tangtrongsup and Scorza 2010; Gruffydd-Jones et al. 2013).

A study from Da Silva et al. (2011) shows 100% efficacy with a single dose treatment of secnidazole. Minor side effects were reported, showing high salivation in the first few minutes *post* drug administration, and apathy and anorexia for 48h in more sensitive animals. Secnidazole has shown limited usage for not being commercially available in veterinary medicine.

Additional treatment options include quinacrine and furazolidone, reported by Tangtrongsup and Scorza (2010).

3.5.1.2 *Toxoplasma* sp.

Toxoplasma is a Coccidia (subclass) from the phylum Apicomplexa, order Eucoccidiorida and family Sarcocystiidae (Taylor et al. 2016). *Toxoplasma gondii* is the only identified species in the genus and its only identified definitive hosts are members of the Felidae family (Bowman 2021). This parasite can, however, infect almost all warm-blooded animals, having a wide range of paratenic hosts (Bowman 2021).

It has been reported in *Panthera tigris sumatrae* and *Panthera onca* in Czech, Slovak, and Mexico City zoos (Sedlák and Bártová 2006; Alvarado-Esquivel et al. 2013; Bártová et al. 2018), in captive *P. t. sumatrae* in Sidney, Australia (Hill et al. 2008), in free-ranging *P. onca* in Brazil (Onuma et al. 2014) and French Guiana (Demar et al. 2008), and in captive *P. onca* in Brazil (Silva et al. 2001; André et al. 2010) and the United States of America (Spencer et al. 2003).

It has also been reported in free-ranging lions from southern Africa (Kruszka and Harris 2020), from South Africa, Namibia, Botswana and Zimbabwe (Cheadle et al. 1999; Penzhorn et al. 2002; Hove and Mukaratirwa 2005; Seltmann et al. 2020), and from the Serengeti (Riemann et al. 1975).

Life cycle Sporulated oocysts have two sporocysts, each with four sporozoites. They are ingested and ruptured in the intestine, releasing the sporozoites. The sporozoites invade intestinal and associated lymph node cells to multiply and evolve to tachyzoites (a rapidly multiplying form) which will then spread to all other tissues of the body and continue the multiplication process (Bowman 2021). Tachyzoites may also be transmitted "transplacentally" (Dubey 2010, p.5) from a mother to her foetus (Dubey 2010).

In the brain, striated muscles, and liver, bradyzoites (slowly multiplying forms) start forming cysts in which they remain protected and viable for the rest of the host's life. This form is infective to fundamentally all warm-blooded animals by ingestion. Both tachyzoites and bradyzoites multiply by endodyogeny (Bowman 2021).

When Felidae ingest bradyzoites, these have a similar activity to sporocysts and invade intestinal cells where either they form tachyzoites, following the asexual cycle, or they are differentiated into gametes which will in turn form oocysts that will be expelled in the faeces (Bowman 2021).

In Felidae, it has been hypothesized that cysts containing bradyzoites may rupture and the bradyzoites travel to the small intestine enterocytes to restart the enteroepithelial cycle (Dubey 2010).

Oocysts are more infective in paratenic hosts than in Felidae, as they multiply more efficiently in these animals, whereas bradyzoites multiply into millions when infecting Felidae, being the most efficient form of infection for this family (Dubey 2010).

Clinical signs Most animals don't show clinical signs, however there have been cases of anorexia, vomiting, diarrhoea, and fever (Dubey 2010; Spycher et al. 2011; Bowman 2021). In pregnant animals, there is also the risk for malformation or even abortion in mothers infected with *Toxoplasma gondii* (Bowman 2021).

Diagnosis As the clinical signs are nonspecific and many times non-existent, they are not enough to diagnose the infection (Dubey 2010). Therefore, diagnosis options are available, such as isolation and inoculation on laboratory animals, tissue culture and finding the parasite in tissues removed by biopsy or necropsy following microscopic evaluation. Another option is impression smears visualized under the microscope with Giemsa staining (Dubey 2010). Visualising tachyzoites indicates active infection, however the visualization of bradyzoite cysts may indicate a latent infection and it's not enough for a diagnosis (Dubey 2010).

Diagnosis can also be done by the observation of oocysts in Felidae faeces, through any conventional flotation technique. Salt solutions with over 1.18 specific gravity should be avoided as the oocysts may appear distorted (Dubey 2010). Since cats shed *Toxoplasma* for short periods of time post infection, and it's difficult to correctly identify the oocysts, the detection of the parasite in faeces is not a reliable method of diagnosis (CUCVM 2018).

Serologic techniques may also be used for the detection of antibodies (Modified Agglutination Test - MAT, Indirect Fluorescent Antibody Test - IFA, ELISA, among others) (Dubey 2010). Another option is the detection of DNA through PCR (Dubey 2010).

Prevention and Control To prevent captive Felidae from being infected, the establishment should avoid feeding the animal uncooked meat or freeze the meats before the animal's consumption (Dubey 2010). The choice of certain meat types can also be beneficial as beef is less likely to be infected than horse, pork, or sheep meat (Dubey 2010).

Proper hygiene of the enclosure including daily removal of faeces is also recommended to avoid the sporulation of oocysts, as well as proper hygiene from the carer, wearing protective clothing, masks, and gloves. The caretaker should also use instruments when cleaning faeces and subsequently thoroughly washing and sterilizing said instruments, through autoclaving, or heating them up to 70°C for at least 10 minutes. Hand washing should be done as often as necessary. Cases of zoonosis can and should be avoided with these practices (Dubey 2010).

Treatment The two main drugs utilized are sulfonamides and pyrimethamine, which are usually well tolerated, however with the possibility of causing megaloblastic anaemia, thrombocytopenia and/or leukopenia (Dubey 2010; Bowman 2021). To target these effects, it is beneficial to give the patients folinic acid and yeast. Sulfadiazine, sulfamethazine, and sulfamerazine are good sulfonamide options for the therapy (Dubey 2010). This therapy works best on the acute stage of the infection, and it has limited effects on subclinical disease, but it has been demonstrated that sulfonamides are able to restrain tissue growth in cysts in mice, in an article by Beverley, J. K. A. (1958), as quoted by (Dubey 2010).

Other drugs used in the treatment for toxoplasmosis include spiramycin, piritrexim, roxithromycin, clindamycin hydrochloride, clindamycin phosphate (Dubey 2010; Bowman 2021), cyclosporin A, atovaquone (Dubey 2010), a novel triazine (Mui et al. 2008) and ponazuril (Mitchell et al. 2004; Mitchell et al. 2006) that, according to the success in its usage in treatment in rats (Mitchell et al. 2004), it may reduce shedding of oocysts in cats. Atovaquone has been reported to kill tissue cysts, putting it in an advantage point. Although there are other options for the therapy of *Toxoplasma gondii* infection, none have been qualified enough to fully replace sulfonamides and pyrimethamine as the mainly used drugs (Dubey 2010).

3.5.1.3 *Sarcocystis* spp.

Sarcocystis is a Coccidia (subclass) from the phylum Apicomplexa, order Eucoccidiorida and family Sarcocystiidae (Taylor et al. 2016). This parasite has a wide array of hosts, as carnivores and humans are the definitive hosts, and numerous herbivores and omnivores are intermediate hosts (Bowman 2021).

This parasite has been reported in free-ranging lions from East Africa (Kruszka and Harris 2020) and Tanzania (Bjork et al. 2000). *Sarcocystis neurona* has been reported in free-living jaguars in the Brazilian Pantanal (Onuma et al. 2014)

Life cycle Fully sporulated oocysts, containing two sporocysts, each with four sporozoites, are ingested by the intermediate host (Bowman 2021). The zoites invade, firstly, mostly the mesenteric endothelial cells and multiply asexually through schizogony. The first generation merozoites will then multiply in the endothelium of various capillaries, other generations of merozoites may be formed in circulating mononuclear cells (Bowman 2021). The merozoites will travel and enter striated muscle cells, or even nervous cells, in which the resulting bradyzoites will multiply and form sarcocysts (Bowman 2021).

The carnivorous definitive host ingests the sarcocysts in the prey's flesh and the zoites it contains invade their enterocytes. In the intestinal cells, gametogony occurs and, in the resulting fusion of gametes, oocysts are formed. The sporulation occurs inside the definitive host and the oocyst is discharged with the faeces, fully sporulated and immediately infective (Bowman 2021).

Domestic cats and free-living jaguars have been identified as an intermediate host for *Sarcocystis neurona* (Dubey et al. 2000; De Meneses et al. 2014; Onuma et al. 2014), for which, the definitive host is the opossum (*Didelphis* spp.) (Bowman 2021).

Clinical signs Carnivores don't usually show clinical signs when infected with *Sarcocystis*. Dubey and Fayer (1983) allude to cases of carnivores being experimentally fed heavily infected meat and most animals were not affected. Some demonstrated punctual vomiting and inappetence, but it was not proven if the signs related to the diet or to the parasite.

Intermediate hosts have demonstrated many clinical signs, sometimes leading up to death (Dubey and Fayer 1983; Bowman 2021). Cats acting as an intermediate host for *Sarcocystis neurona* may also demonstrate neurological clinical signs (Dubey et al. 2001; Dubey et al. 2003; Bisby et al. 2010).

Diagnosis In the Felidae, the diagnosis through clinical signs is not viable, as they are very rare. The diagnosis can be performed upon coprological tests and observation of oocysts through a microscope.

In Felidae acting as an intermediate host, PCR or Western blot of cerebrospinal fluid may also assist diagnosis. However, diagnosis depends mostly on the histopathological observation of lesions, zoites and sarcocysts in the central nervous system (Bowman 2021).

Prevention and Control To break the cycle, carnivores should not be fed uncooked meat, as a high percentage of adult cattle and sheep worldwide have found to be infected with

Sarcocystis spp., and their faeces should be adequately disposed of, to prevent contaminating intermediate hosts (Dubey and Fayer 1983).

Adequate hygiene in the enclosure, and especially from the animal handlers is essential, as humans are also definitive hosts.

Treatment Some routine treatments include sulfonamides with pyrimethamine, diclazuril (Bowman 2021) and ponazuril (Lindsay et al. 2000; Franklin et al. 2003; Bisby et al. 2010; Bowman 2021). Yet another option would be the combination of trimethoprim with sulfonamides (Bisby et al. 2010; Bowman 2021).

Bisby et al. (2010) reports a cat affected with encephalomyelitis caused by *Sarcocystis*, demonstrating various neurological clinical signs, that was successfully treated with several compounds, amongst them clindamycin with a palliative buprenorphine and fluid therapy, then ponazuril, and later, folic acid, and trimethoprim sulfonamide with pyrimethamine.

3.5.1.4 *Cystoisospora* spp.

Cystoisospora is a Coccidia (subclass) from the phylum Apicomplexa, order Eucoccidiorida and family Sarcocystiidae (Lindsay 2004). It used to be included in the *Isospora* genus in the Eimeriidae family, but based on molecular evidence and life cycles, the *Isospora* affecting mammals was changed to *Cystoisospora* (Lindsay 2004; Barta et al. 2005). However, to this day, some sources still use the term *Isospora* when referring to the genus affecting mammals. This parasite affects mostly carnivores and swine (Bowman 2021). Common *Cystoisospora* in Felidae include *C. felis* and *C. rivolta* (Bowman et al. 2002; Bowman 2021).

This parasite has been reported in captive lions in Zimbabwe (Mukarati et al. 2013), and free-ranging lions from Tanzania (Bjork et al. 2000) and from east, southern and west Africa (Kruszka and Harris 2020). It has also been reported in free-ranging jaguars in Belize (Patton et al. 1986).

Life Cycle The life cycle of the *Cystoisospora* is monoxenous, but paratenic hosts may also contribute to the exposure of the carnivorous definitive host to the parasite (Bowman 2021).

The parasite enters the definitive host's body orally, in the form of a sporulated oocyst, containing two sporocysts with four sporozoites each. From there the zoites invade enterocytes from the small intestine and multiply through schizogony. Third generation merozoites evolve and originate gametes. From the fusion of said gametes the oocyst is originated and expelled, not yet sporulated, from the body with the faeces (Bowman et al. 2002; Bowman 2021).

Clinical signs Adult, immunocompetent felids don't demonstrate clinical signs. Kittens, however, have shown clinical signs such as enteritis, mild diarrhoea, emaciation and even death (Bowman et al. 2002; Bowman 2021). Dubey (2018) however shows this parasite to have little consequences in both adults and kittens.

Diagnosis Oocysts can be observed under the microscope from faeces subjected to sugar flotation (Bowman 2021).

Prevention and Control Similar to the prevention of other Coccidia. Adequate hygiene measures in the enclosure and in the carers' personal conduct is essential.

Treatment As it was for other coccidiosis, sulfonamides are useful in the treatment of *Cystoisospora*, such as sulfadimethoxine alone or with ormetoprim, sulfadiazine in combination with trimethoprim, or sulfaguanidine alone (Bowman et al. 2002; Bowman 2021). Amprolium is also useful alone or in combination with sulfadimethoxine (Bowman et al. 2002). Other drugs that can be used are toltrazuril, ponazuril, quinacrine or furazolidone (Bowman et al. 2002; Bowman 2021).

3.5.2 Helminths

Animal's susceptibility to helminths increases if the nutritional needs are not met, therefore, adequate and good quality of food is essential to prevent disease, among other complications (Rimfa et al. 2019).

The most common helminths reported, in Zoo animals in Europe, are from the phylum Nematoda (Panayotova-Pencheva 2013). Panayotova-Pencheva (2013) also concludes in her study that the most common nematodes found were of the Ascaridida order, followed by Strongylida, Enoplida, Oxyurida and Rhabditida.

3.5.2.1 Ancylostomatidae

This Nematoda family belongs to the order Strongylida and contains seven different genera, amongst them *Ancylostoma* and *Uncinaria* (Taylor et al. 2016), the only two affecting felids (Bowman et al. 2002). Although it has been demonstrated in Felidae, *Uncinaria stenocephala* is not very common in this family (Bowman et al. 2002).

Parasites of this family have been reported in a variety of cases in Felidae. Different subspecies of *Ancylostoma* have been reported in free-ranging lions from Tanzania (Bjork et al. 2000) and from east and southern Africa (Kruszka and Harris 2020), as well as captive lions in Zimbabwe (Mukarati et al. 2013). Biocca (1951) also mentions the study of this parasite in lions in Zambia (Northern Rhodesia at the time). *Ancylostoma* has also been reported in 9/12 (75%) free-ranging Sumatran tigers examined in Sumatra (Amarilis et al. 2023) and in captive Sumatran tigers in a zoo in Indonesia (Arrayyansyah et al. 2014).

The jaguar is also a species that has shown to be infected by *Ancylostoma*, reported in captive jaguars in 30 AZA (Association of Zoos and Aquariums, at the time American Zoo and

Aquarium Association) North American Zoos (Hope and Deem 2006), and specifically in a zoo in New York (McClure 1931) and in a zoo in Mexico (Rendón-Franco et al. 2013). Patton et al. (1986) quotes Thatcher (1971), Canavan (1929) and McClure (1933) to report *A. pleuridentatum*, and quotes Thatcher (1971) to report *A. tubaeforme*, both in *P. onca* (jaguar). Vieira et al. (2008) also reports *Ancylostoma* sp. in jaguars in Brazil.

Uncinaria stenocephala has been reported in lions in southern Africa (Kruszka and Harris 2020).

Life cycle The worm, in the form of third-stage larvae, infects the host both orally and through skin penetration (Bowman et al. 2002).

The larvae may enter the definitive host orally by consuming a paratenic host, such as rodents. In the paratenic host, the parasite persists in the musculature of the animal (Bowman et al. 2002). When the felid feeds on the paratenic host, the larvae enter the gastrointestinal tract and penetrate the stomach and proximal intestinal wall, where they develop into the adult form. Once developed, they return to the intestinal lumen (Bowman et al. 2002).

When entering through skin penetration, the larvae migrate to the lungs via the bloodstream, move up the trachea and enter the gastrointestinal tract through the oesophagus. They develop into the adult form inside the wall of the gastrointestinal tract, from where they will re-enter the lumen (Bowman et al. 2002). In the lumen, the hookworm reproduces sexually, and eggs are shed in the faeces. The egg develops into the larval stages outside the host (Bowman 2021).

Clinical signs Depending on the genus, different intestinal blood losses are expected. *Ancylostoma tubaeforme* causes significant blood losses (Bowman et al. 2002), whereas *Uncinaria stenocephala*'s impact on blood loss is very limited (Bowman 2021).

Clinical signs are not commonly demonstrated but depending on the intensity of the infection and on the parasite, strong clinical signs may be expected, such as weight loss, regenerative anaemia coupled with melena, and possible death (Bowman et al. 2002; Bowman 2021).

Amarilis et al. (2023) report cases of ten Sumatran tigers infected with *Ancylostoma* sp., five of them co-infected with other helminths (four without clinical signs), and five infected only with *Ancylostoma*. Of the ten, six demonstrated anaemia and two showed signs of dehydration. All the animals infected only with *Ancylostoma* sp. were anaemic.

Diagnosis Clinical signs may aid the diagnosis, but the confirmation is made through the evaluation of eggs in faecal flotation (Foreyt 2001).

Prevention and Control The enclosure must be cleaned daily with faeces removal, to prevent larvae from developing (Bowman 2021). They develop best in shaded areas, without excess water, therefore these areas should be monitored in cases of infection (Bowman 2021). Hookworm larvae may also be eliminated from the enclosure with sodium borate. It works well in "gravel- or loam-surfaced runs" (Bowman 2021, p.199), yet it should not be used in vegetation as it destroys it (Bowman 2021).

Paved surfaces should be washed properly, and then sprayed or mopped with 1% sodium hypochlorite solution (Clorox®) (Bowman 2021).

Treatment According to Bowman (1992), as quoted by Bowman et al. (2002), *Ancylostoma* has been treated in cats with febantel, febantel-praziquantel, dichlorvos, n-butyl chloride and disophenol sodium. Selamectin is also recommended to treat *A. tubaerforme* (Bowman et al. 2002), toluene to treat *A. braziliense*, and ivermectin is recommended to treat both *A. braziliense* and *A. tubaerforme* (Nolan et al. 1992).

3.5.2.2 Ascaridoidea

This is a superfamily from the order Ascaridida. From this order, *Toxascaris leonina* and *Toxocara cati* are the most common species affecting cats (Bowman et al. 2002), belonging to the families Ascarididae and Toxocaridae, respectively (Li et al. 2018).

Toxocara cati and non-specified species of *Toxocara* have been reported in all four of the animal subspecies studied in this dissertation. Free-ranging lions in Tanzania (Bjork et al. 2000) and east and southern Africa (Kruszka and Harris 2020) have shown to be infected, as well as captive lions in Zimbabwe (Mukarati et al. 2013).

Toxocara has also been reported in free-ranging Sumatran tigers in Sumatra (Amarilis et al. 2023) as well as in captivity in a zoo in Malaysia (Lim et al. 2008) and in a zoo in Indonesia (Arrayyansyah et al. 2014). It has also been reported in both free-ranging (Sepalage and Rajakaruna 2020) and captive (Aviruppola et al. 2016; Kobbekaduwa et al. 2017) Sri Lankan leopards in Sri Lanka.

Lastly, *Toxocara* sp. has been reported in free-ranging jaguars in various countries in Central and South America (Patton et al. 1986; Noronha et al. 2002; Srbek-Araujo et al. 2014; Uribe et al. 2021) and in captivity jaguars in New York (McClure 1931) and Peru (Aranda et al. 2013).

Toxascaris leonina has been reported in all four species as well. Reported in free-ranging lions in East Africa (Kruszka and Harris 2020) and in captivity in Zimbabwe (Mukarati et al. 2013). Seneviratne (1955), as quoted by Perera et al. (2013), reports *T. leonina* in Sri Lankan leopards in Sri Lanka. Arrayyansyah et al. (2014) report this parasite in the Sumatran tiger, in a zoo in Indonesia. It has also been reported in free-ranging jaguars in Belize (Patton et al. 1986) and in jaguars in Peruvian zoos (Aranda et al. 2013). Vicente et al. (1997) mentions the existence of *T. leonina* in jaguars in Brazil.

Life Cycle Once laid and shed in the faeces, the eggs of *Toxascaris leonina* take one week to reach the infective stage, whereas eggs of *Toxocara* take four weeks to reach said state (Bowman 2021).

Toxascaris leonina The eggs are ingested directly by the definitive host (canids and felids), in which the larvae hatch, or are ingested by a paratenic host, in which the hatched larvae remain encysted in different tissues until the animal is preyed on by the carnivorous host (Bowman 2021). Once in the definitive host, the larvae migrate through the gastrointestinal tract and invade the mucosa of the small intestine to moult. The larvae then return to the lumen to mature and lay eggs (Bowman 2021).

Toxocara cati Eggs containing infective larvae are ingested either by the definitive felid host, or by a paratenic host, hatching in both (Bowman 2021). If ingested by the definitive host, the larvae will invade the intestinal wall and, either migrate through blood vessels to the lungs, travel up the trachea and return to the gastrointestinal tract through the oesophagus, where they will develop into adults in the intestine, or continue through the blood vessels, past the lungs, and encyst into different organs and tissues, in which case, the larvae may be reactivated and travel to the stomach wall, where they moult into adults, or travel to the mammary glands in acutely infected lactating mother felids (cats), and are ingested by the offspring (Bowman 2021). If paratenic hosts ingest the eggs, the larvae carry out somatic migration, from the intestines entering the bloodstream to travel to the liver, to the lungs and finally to tissues, where they remain encysted until the definitive host ingests the infected tissue, and the larvae moult into their final form in its stomach wall (Bowman 2021). Adults in the definitive host's small intestine lay eggs to be shed in the faeces (Bowman 2021). In the environment, the singular cell in the egg goes through two moults to become an infective larva of the 3rd stage. Such occurs in a four-week period (Bowman et al. 2002; Bowman 2021).

Clinical signs In both *Toxocara cati* and *Toxascaris leonina*, clinical signs are usually not apparent, except in severe infections (Bowman 2021).

Diagnosis *Toxocara* and *Toxascaris* may be diagnosed with microscopical observation of preparations obtained from faecal flotation techniques (Foreyt 2001). Diagnosis can also be performed post-mortem with observation of the roundworm in the intestine or other affected organs (Sprent 1956).

Prevention and Control Eggs of *T. leonina* moult to their infective stage in one week, whereas *Toxocara* eggs take four weeks to become infective, which is a possible explanation for why *Toxascaris* infections are harder to eradicate (Bowman 2021).

Pest control is very important, because according to Okulewicz (2008), as quoted by Okulewicz et al. (2012), *Toxocara* and *T. leonina*'s main infection route to caged zoo animals is the paratenic hosts the felids may consume, such as rodents.

Toxascaris and *Toxocara* eggs are very resistant to various environmental variables and endure such conditions for years, while persisting infective (Bowman 2021). As it is virtually impossible to eradicate the eggs from soil, possible solutions would be to replace at least the top thirty centimetres of soil, or to cover it with a concrete or asphalt layer (Bowman 2021).

If the contaminated area consists of manmade floors, such as concrete, high-pressure washers are advised, with posterior spraying or mopping with 1% sodium hypochlorite (Bowman 2021). These processes only loosen the eggs from the surface, to kill them, 60°C temperature for five minutes is necessary (Bowman 2021), for example with heated water. Wires and wooden surfaces are incredibly hard to clean (Bowman 2021).

Treatment Both *Toxocara* and *Toxascaris* can be treated with dichlorvos, pyrantel, piperazine, febantel and mebendazole (Bowman et al. 2002; Rimfa et al. 2019).

Additionally, *Toxocara cati* can be treated with ivermectin, milbemycin, fenbendazole, nitroscanate and selamectin or praziquantel formulated with febantel or pyrantel (Bowman et al. 2002; Rimfa et al. 2019). *Toxascaris* can also be treated with toluene (Bowman et al. 2002).

Okulewicz et al. (2012) reports an Angolan lion (*P. l. bleyenberghi*) treated successfully for a *T. leonina* infection with Valbazen® (albendazole and cobalt sulphate) and Advocate™ (imidacloprid and moxidectin). The animal however, demonstrated the parasite again after two months. It is unclear whether the infection persisted or there was a re-infection.

Hase et al. (2007) reports a captive lion with a *Toxocara* sp. and *Balantidium coli* infection, successfully treated with metronidazole, fenbendazole, ampicillin-cloxacillin, as well as antihistamine and vitamins with fluid therapy.

4. Materials and Methods

The information utilized was collected from a database used by the Barcelona Zoo to register and archive all the information on previous and current inhabitants as well as animals formerly and presently owned by the Zoo. The database used is registered on an online software called ZIMS (Species360 Zoological Information Management System, Version 1.7 Updated: 27 Jan. 2014), a program used worldwide in more than 1300 zoos, aquariums and wildlife institutions as a tool for organizing records of the animals in their care.

The information searched is from March 1990 until February 2024. In the "Medical Records", it was filtered by selecting "Tests & Results", selecting the sample type "Feces" and the taxonomies "*Panthera leo bleyenberghi*" (given the updated denomination of this subspecies, mentioned in the literature review, it will be referred to as *Panthera leo melanochaita* in this dissertation), "*Panthera pardus kotiya*", "*Panthera onca*" and "*Panthera tigris sumatrae*", the four members of Felidae present in the Barcelona Zoo. Additionally, these four taxonomies were also cross searched with cases that wrote the parasite name "*Toxoplasma*" in the notes, in any sample type, as it is a gastrointestinal parasite difficult to find in the faeces, and its infection is also possible to be detected in serological testing.

The enclosures of the Felidae are cleaned daily, with removal of faeces and food remains, the analysed faeces were therefore sampled not too long after excretion from the animal. It was possible to identify the faeces of each animal for different reasons: the Sri Lankan leopards, jaguars and Sumatran tigers live in isolation, as solitary animals; the lions share an enclosure, but at times sleep separately, therefore allowing the identification of faeces in the isolated enclosures; or if needed, the employment of different coloring to the lions' diet to mark the faeces of each animal, would make the differentiation easier.

The database contained 25 variables: species, identification number of the animal, date of faeces collection, 10 types of tests performed to identify the parasites in the faeces ($NaNO_3$ Flotation, $ZnSO_4$ Flotation, Sugar Flotation, NaCl Flotation, Non-specified Flotation, Direct Exam, Sedimentation, Stained MIF (Merthiolate Iodine Formalin), Acid-Fast Finding and Baermann Technique), serum analysis utilised to find *Toxoplasma* positivity, the season of the year, the year span in which the tests were performed and nine types of parasite categories: Coccidia, *Isospora* spp. (given the updated changes to the genus, mentioned in the literature review, it will be referred to as *Cystoisospora* spp. in this dissertation), nematodes, *Toxascaris leonina*, Toxocaridae, *Toxoplasma gondii*, flagellates, non-sporulated oocysts and sporulated oocysts. As informed by the Zoo, the analysis performed on the serum samples of *Panthera tigris sumatrae* in 2007 and 2008, although not specified, were most likely done with the latex agglutination test, a test performed on a different occasion to attempt to detect *Toxoplasma* sp. antibodies on the zoo's tigers, in 2018.

With this data, we analysed the parasitic prevalence in the four climatic seasons of the year: spring (March, April, May), summer (June, July, August), autumn (September, October, November) and winter (December, January, February). Chi-square tests were performed and interpreted using a confidence level of 95%, and the results were considered statistically relevant when the significance level was $p<0,05$.

We also analysed the parasitic prevalence in each of the four Felidae species during the studied time, as well as the evolution of the parasitic prevalence in each species and in all Felidae throughout 1990-2024, in the Barcelona Zoo.

To show the level of agreement between testing samples using flotation methods and using direct methods, Cohen's kappa value was evaluated for these two parameters in the 103 cases, in which these two types of techniques were performed.

The statistical analysis and the table and graph design were done utilising the programs IBM SPSS Statistics Data Editor (Version 29.0.0.0 (241), 1989, 2022) and Microsoft Excel (Version 2405 Build 16. 0. 17628. 20006., 2018).

5. Results

The database contained a total of 146 cases of animal testing among the four species. Of the 146 cases, there are 141 cases where only faecal samples were tested, three cases where only serological samples were tested, and two cases where both types os samples were tested. Therefore having a total of 143 cases with faecal samples and five cases with serological samples.

Since the database dates to 1990 and, at the time, the registering of medical procedures and tests was done on paper, it is possible that data may have been lost throughout the years. Therefore, in this study, the examined data is based on the currently registered tests that were performed.

In the time period searched, there are the following tested felids: 12 African lions, seven Sri Lankan leopards, four jaguars and five Sumatran tigers. Many were tested repeatedly throughout the years, being considered as different cases. There are 49 cases of tested lions, 41 of leopards, 23 of jaguars and 33 of tigers. Each case consisted of one tested animal, the species of the animal, the date in which the faeces or blood were collected, the analysis performed to search for parasites, and whether it was positive for each of the various parasites in the database.

In some cases, the specific genus and species of the parasite were not identified and were therefore registered as what was possible to identify, as is the case for Coccidia, nematodes, Toxocaridae, flagellates, non-sporulated oocysts and sporulated oocysts.

In Tables 2 and 3 it is possible to see the parasitic prevalence in each animal in the four seasons of the year. In Table 2 we see the results of gastrointestinal parasites found in coprology, and in Table 3 we see the results of gastrointestinal parasitic infections found in serology.

Unlike every other parasite found in this study, *Toxoplasma gondii*'s presence was detected in the form of antibodies in non-routine serum analysis performed on the Sumatran tigers, as it is demonstrated on Table 3. Its prevalence percentages are calculated according to the amount of serum samples analysed (n=5).

The Southern African lion shows higher Coccidia prevalence during summer (29%), higher Toxocaridae prevalence during autumn (30%) and higher *Toxascaris leonina* prevalence during spring (100%) and summer (47%), however, since only one registered testing occurred during the spring, the 100% prevalence is a statistical bias. *Toxascaris leonina* is the only detected parasite that showed positive results on all of the four seasons.

The jaguar shows higher parasitic prevalences during spring for Coccidia (21%), *Cystoisospora* spp. (14%) and *Toxascaris leonina* (14%), and higher parasitic prevalences during autumn for nematodes (29%), flagellates (14%), and sporulated (14%) and non-sporulated oocysts (14%). There are no registered tests performed during summer on this animal species.

The Sri Lankan leopard shows higher Coccidia prevalence (9%) during spring, and higher prevalences for nematodes (14%) and *Toxascaris leonina* (14%) during autumn.

The Sumatran tiger only shows positive results for parasites during winter, for *Cystoisospora* spp. (8%) and *Toxascaris leonina* (15%).

Also on table 2 we can observe the total prevalence of parasitic infection in each of the four species: the Southern African lion had a prevalence of 100% (1/1) during spring, and during summer 76% (13/17). The lion also has a 50% (5/10) parasitic positivity prevalence during autumn and a 33% (7/21) prevalence during winter.

The jaguar shows 21% (3/14) of parasitic positivity in the spring, 43% (3/7) during autumn, and 0% (0/2) during winter. The jaguar didn't have any registered testing performed during the summer. The Sri Lankan Leopard shows 9% (2/22) parasitic prevalence during spring, 0% (0/1) during summer, 29% (4/14), during autumn and 0% (0/4) during winter.

Lastly, the Sumatran tiger only had positive results during winter, with a prevalence of 23% (3/13).

The Sumatran tiger shows positive parasitic results from coprology only during winter, however, this animal also shows *Toxoplasma gondii* infected serum samples during summer and autumn, and one negative sample in the spring. In autumn, three samples were analysed, all positive, and in the summer, the only tested sample was positive for *Toxoplasma gondii* antibodies as well, showing 100% prevalence in both summer and autumn, and 0% in spring. The Chi-square testing done on the seasonality of the parasitic prevalence shows values that are mostly higher than 0.05, except for the 0.006 value shown in the Toxocaridae prevalence in *Panthera leo melanochaita*, and the 0.047 value in this same animal's total prevalence's seasonality.

Table 2: Parasitic prevalence findings, from coprology, in the Barcelona Zoo's Felidae in the four seasons of the year

		Prevalence (%) (# positives/ # performed tests)				
Species	Parasite	Spring	Summer	Autumn	Winter	p-value
Southern African lion *Panthera leo melanochaita*						
	Coccidia	0 (0/1)	29 (5/17)	0 (0/10)	5 (1/21)	0.063
	Toxascaris leonina	100 (1/1)	47 (8/17)	20 (2/10)	29 (6/21)	0.228
	Toxocaridae	0 (0/1)	0 (0/17)	30 (3/10)	0 (0/21)	0.006
Jaguar *Panthera onca*						
	Coccidia	21 (3/14)	NA	0 (0/7)	0 (0/2)	0.330
	Cystoisospora spp.	14 (2/14)	NA	0 (0/7)	0 (0/2)	0.495
	Nematodes	0 (0/14)	NA	29 (2/7)	0 (0/2)	0.082
	Toxascaris leonina	14 (2/14)	NA	0 (0/7)	0 (0/2)	0.495
	Flagellates	0 (0/14)	NA	14 (1/7)	0 (0/2)	0.303
	Non-sporulated oocysts	0 (0/14)	NA	14 (1/7)	0 (0/2)	0.303
	Sporulated oocysts	0 (0/14)	NA	14 (1/7)	0 (0/2)	0.303
Sri Lankan leopard *Panthera pardus kotiya*						
	Coccidia	9 (2/22)	0 (0/1)	0 (0/14)	0 (0/4)	0.611
	Nematodes	0 (0/22)	0 (0/1)	14 (2/14)	0 (0/4)	0.256
	Toxascaris leonina	0 (0/22)	0 (0/1)	14 (2/14)	0 (0/4)	0.256
Sumatran tiger *Panthera tigris sumatrae*						
	Cystoisospora spp.	0 (0/2)	0 (0/9)	0 (0/6)	8 (1/13)	0.717
	Toxascaris leonina	0 (0/2)	0 (0/9)	0 (0/6)	15 (2/13)	0.423

	Total Prevalence (%) (# positives/ # performed tests)				
Species	Spring	Summer	Autumn	Winter	p-value
Southern African Lion *Panthera leo melanochaita*	100 (1/1)	76 (13/17)	50 (5/10)	33 (7/21)	0.047
Jaguar *Panthera onca*	21 (3/14)	NA	43 (3/7)	0 (0/2)	0.390
Sri Lankan leopard *Panthera pardus kotiya*	9 (2/22)	0 (0/1)	29 (4/14)	0 (0/4)	0.311
Sumatran tiger *Panthera tigris sumatrae*	0 (0/2)	0 (0/9)	0 (0/6)	23 (3/13)	0.225

- Number of
NA - Not Applicable, no test was performed in said season

Table 3: Parasitic prevalence findings, from serology, in the Barcelona Zoo's Felidae in the four seasons of the year

Species	Parasite	Prevalence (%) (# positives/ # performed tests) Spring	Summer	Autumn	Winter	p-value
Sumatran tiger *Panthera tigris sumatrae*						
	Toxoplasma gondii	0 (0/1)	100 (1/1)	100 (3/3)	NA	0,082

- Number of
NA - Not Applicable, no test was performed in said season.

Regarding prevalence, the African lion (*P. l. melanochaita*) shows the highest parasitic prevalence of the four species, as it is shown in Table 4, with 53% prevalence, followed by the jaguar (*P. onca*) with 26% prevalence. The jaguar, however, surpasses the lion with higher parasitic variety reported. The Sri Lankan leopard (*P. p. kotiya*) has the third highest parasitic prevalence, with only six positives in 41 tested samples (15%). The Sumatran tiger (*P. t. sumatrae*) shows the lowest parasitic prevalence, only with three positive faecal samples out of 30 tested samples, with a prevalence of 10%. The tiger, however, did show additional positive cases for *Toxoplasma gondii,* a parasite that was not found in coprological tests and was only reported in non-routine serum tests. This parasite was found in 80% (4/5) of the evaluated samples.

Toxascaris leonina is the most prevalent parasite in these animals, with 23 positives in 143 tested samples, equating a 16% prevalence, and is also the only parasite species present in all four host species. Coccidia has also shown to be present in all the four host species, given that *Cystoisospora* spp. is a genus from the Coccidia subclass.

The second most prevalent parasites were the non-identified Coccidia with 8% prevalence (11/143).

Non-identified nematodes showed to be present in both the jaguars and Sri Lankan leopards, however not in many samples, with a 3% prevalence (4/143).

Toxocaridae show a 2% (3/143) prevalence and flagellates and sporulated and non-sporulated oocysts show the lowest prevalences, all with 1% (1/143).

Nematodes in general show a prevalence of 21% (30/143), and include *Toxascaris leonina* and Toxocaridae. Protozoans show a prevalence of 9% (13/143), comprising Coccidia, *Cystoisospora*, flagellates and sporulated and non-sporulated oocysts.

Table 4: Parasitic prevalence findings, from coprology, in the Barcelona Zoo's Felidae from 1990 to 2024

Parasite	Prevalence (%) (# positives/# performed tests)				
	P. leo melanochaita	*P. onca*	*P. pardus kotiya*	*P. tigris sumatrae*	Total
Coccidia	12 (6/49)	13 (3/23)	5 (2/41)	-	8 (11/143)
Cystoisospora spp.	-	9 (2/23)	-	3 (1/30)	2 (3/143)
Nematodes	-	9 (2/23)	5 (2/41)	-	3 (4/143)
Toxascaris leonina	35 (17/49)	9 (2/23)	5 (2/41)	7 (2/30)	16 (23/143)
Toxocaridae	6 (3/49)	-	-	-	2 (3/143)
Flagellates	-	4 (1/23)	-	-	1 (1/143)
Non-sporulated oocysts	-	4 (1/23)	-	-	1 (1/143)
Sporulated oocysts	-	4 (1/23)	-	-	1 (1/143)
	Prevalence (%) (# positives/# performed tests)				
	P. leo melanochaita	*P. onca*	*P. pardus kotiya*	*P. tigris sumatrae*	Total
Total Nematodes	41 (20/49)	17 (4/23)	10 (4/41)	7 (2/30)	21 (30/143)
Protozoans	12 (6/49)	17 (4/23)	5 (2/41)	3 (1/30)	9 (13/143)
	Prevalence (%) (# positives/# performed tests)				
	P. leo melanochaita	*P. onca*	*P. pardus kotiya*	*P. tigris sumatrae*	Total
Total of Infected Samples	53 (26/49)	26 (6/23)	15 (6/41)	10 (3/30)	29 (41/143)

- Number of

On Table 5 we can see the results of the test performed to analyse the agreement level between flotation techniques and direct method techniques, used to evaluate the faecal samples of the Felidae in the Barcelona Zoo. There is a total of 103 cases in which both tests were performed, of the 103 cases, flotation detected 26 samples positive for parasites, of which 16 were negative when tested with direct methods, and direct method detected 15 positive samples, of which only five were negative on flotation testing. Ten samples were detected as positive from both techniques, giving us a total of 31 positive samples (16 detected in flotation + 5 detected in direct method + 10 detected in both techiques).

Table 5: Agreement level between Flotation and Direct method techniques to find positive parasitic infections in the Zoo's Felidae

		Direct method		
		Negative	Positive	Total
Flotation	Negative	72	5	77
	Positive	16	10	26
	Total	88	15	103

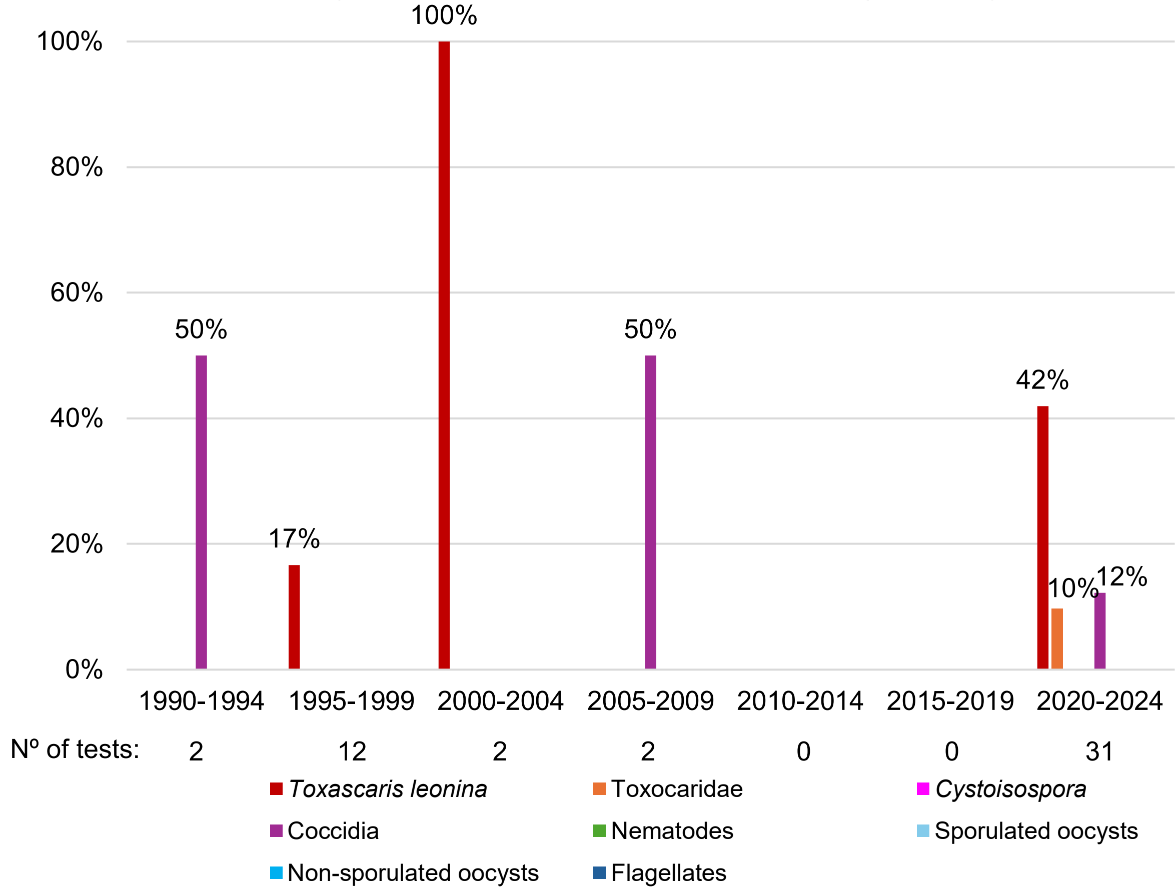
Cohen's kappa value = 0.372

Statistical significance < 0.001

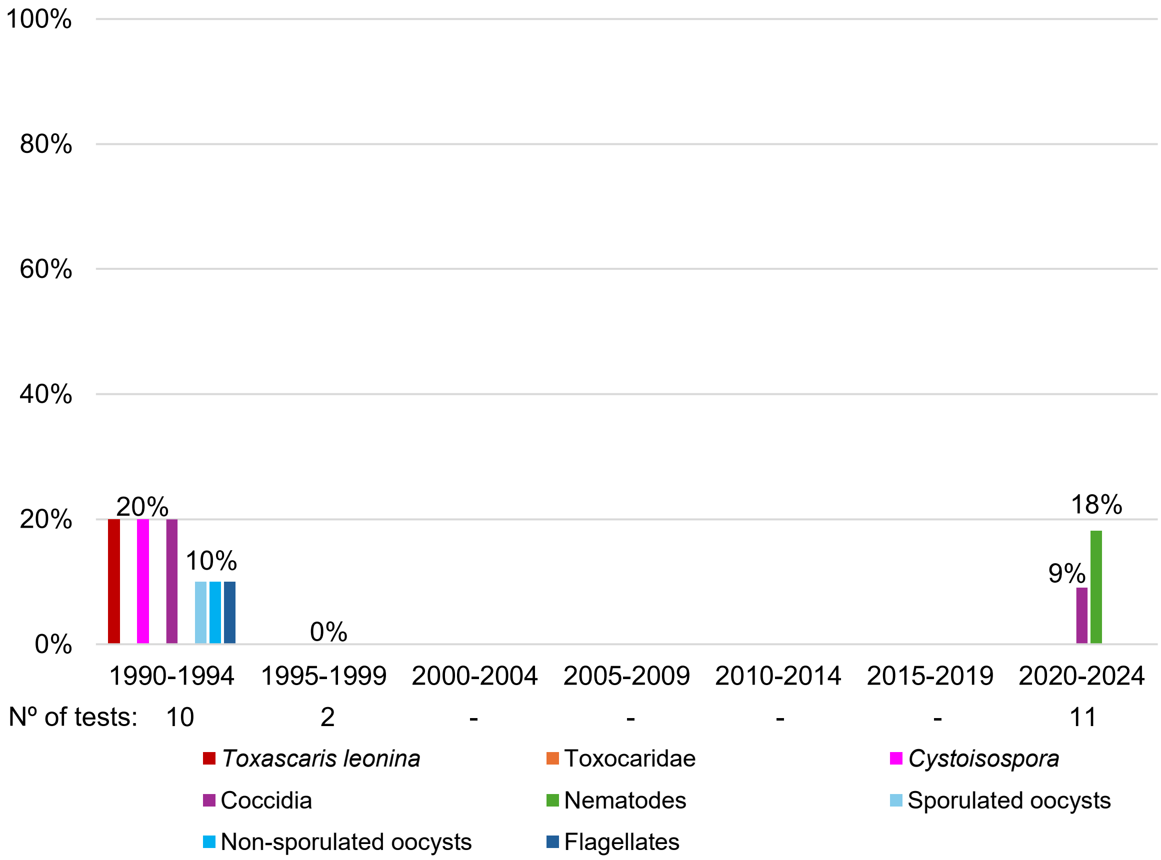
Another relevant study in this dissertation is the evolution of the parasitic prevalence of all the parasites in the studied animals throughout the 35 years of sample analysis. The following Graphs (1-4) show the obtained results in each of the four species. On Graph 5 we see the overall prevalence evolution in all the Felidae combined, throughout the years, from 1990 to 2024.

Graph 1 shows that *Panthera leo melanochaita* has been recurrently infected with *Toxascaris leonina* and Coccidia. In the years 1990-1994, 2000-2004 and 2005-2009, they demonstrated higher infection rates, but it is relevant to note that in those years very few tests were performed. From 2010 until 2019 there are no registered tests. In 2020-2024 many samples were analysed, showing relatively high infection rates overall as well. Some of those samples showed Toxocaridae infections without certainty of the genus of the parasites found.

Graph 1: Parasitic positivity rate in *Panthera leo melanochaita* throughout the years



Graph 2: Parasitic positivity rate in *Panthera onca* throughout the years



Graph 2 shows that all the registered parasitic findings in *Panthera onca* occurred in 1990-1994 and 2020-2024, mostly from the lack of registered test results in the other time periods. In 1995-1999, samples were taken to be tested, and they were both negative for parasites. In 1990-1994 the jaguars were infected with a variety of parasites, in many of which it was not possible to identify the genus, as is the case for Coccidia, sporulated oocysts, non-sporulated oocysts and flagellates. In those years it was possible to also find *Toxascaris leonina* and *Cystoisospora*. In 2020-2024 it was not possible to identify the genus of the positive samples categorized as nematodes and Coccidia.

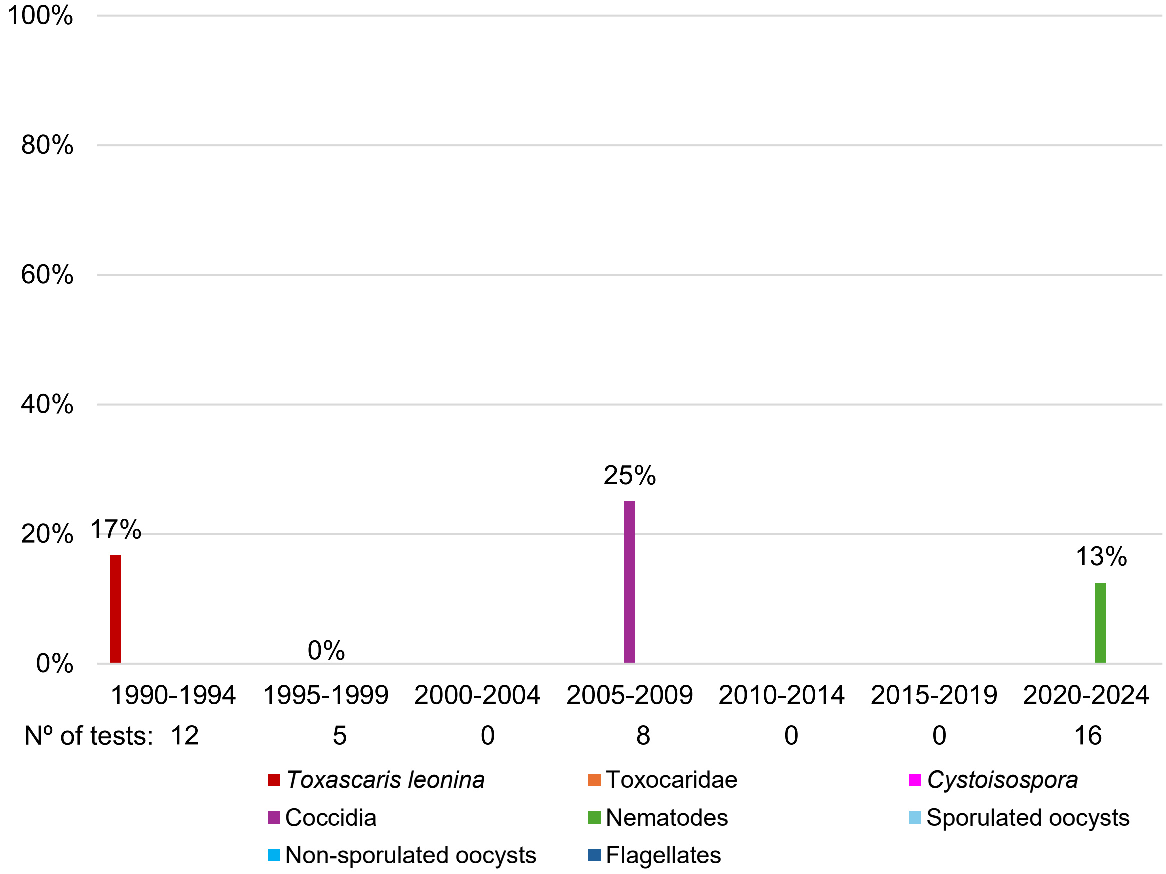
Graph 3 demonstrates infections in *Panthera pardus kotiya* in 1990-1994, 2005-2009 and 2020-2024, all from different parasites, *Toxascaris leonina*, Coccidia and nematodes, showing no re-infections from the same agents in different five-year periods. In 1995-1999, five samples of faeces were submitted for coprology and all the tests resulted negative for parasites. In 2000-2004 and from 2010 to 2019 there are no registered tests. The Sri Lankan leopard shows low parasitic prevalence, overall, throughout the years.

Graph 4 shows that *Panthera tigris sumatrae*, like *Panthera onca*, doesn't have many registered faeces analysed for parasites. It does show positive results in 1995-1999 for *Toxascaris leonina* and in 2020-2024 for *Cystoisospora*. In 2020-2024 the parasitic prevalence is very low, at only 5% in the 20 tested samples, showing that the tigers habiting the Zoo were not very prone to be infected by parasites. In 2005-2009, six samples were tested, and all were negative. In 2005-2009, the Sumatran tiger also had serum samples tested for *Toxoplasma gondii* antibodies, and of the four tested samples, 100% resulted positive for exposure to the parasite. In 2015-2019 the tigers were again tested for *Toxoplasma gondii* antibodies, with negative results this time (0/1). These results are not represented in the graph, as they are not from coprology.

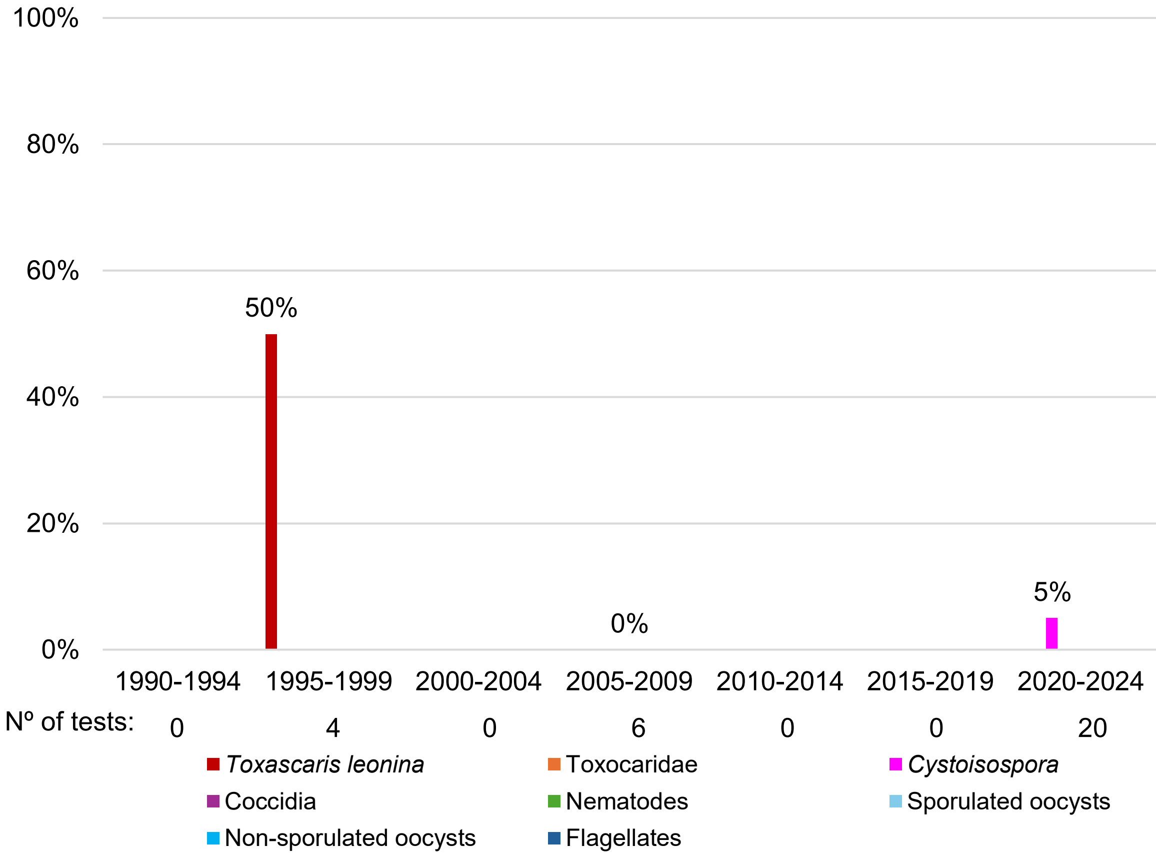
Graph 5 represents the junction of the previous four graphs, demonstrating the evolution of the parasitic positivity rate in all Felidae at the Barcelona Zoo. It is possible to visualize that there are no registered test results from 2010 to 2019. The only registered tests in 2000-2004 were the ones done on faecal samples from the lions and both samples tested positive for *Toxascaris leonina*. The 2020-2024 period has, by far, the most registered coprological tests performed.

In 1990-1994, the Sri Lankan leopards and jaguars were infected with *Toxascaris leonina*, the same with the Sumatran tigers and the African lions in 1995-1999, both infected with *Toxascaris leonina*. Also, in 1990-1994, the African lions and the jaguars were infected with Coccidia. Later, in 2005-2009, the Sri Lankan leopards and African lions were both infected with Coccidia.

Graph 3: Parasitic positivity rate in *Panthera pardus kotiya* throughout the years



Graph 4: Parasitic positivity rate in *Panthera tigris sumatrae* throughout the years



More recently in 2020-2024, the Sri Lankan leopards were infected with nematodes, same as the jaguars, and the jaguars were infected with Coccidia, same as the African lions. Coincidentally, both parasitic infections demonstrating 17% prevalence in Felidae.

Graph 5: Parasitic positivity rate in all Felidae throughout the years

Graphs 1-5: the "Nº of tests" indicates the number of testing procedures performed for each species in all the year spans; the positivity rate was calculated as number of positive results / number of tests performed x 100.

6. Discussion

This study shows some interesting results. The lions showed, overall, 53% prevalence, with spring and summer as their most prevalent seasons; the jaguars showed 26% prevalence, with higher parasitic prevalence during autumn; the leopards had 15% prevalence, peaking during autumn; and the tigers showed 10% prevalence, with the only positive results, in coprology, occurring during winter. The higher parasitic prevalency that occurs during spring on the lions is a statistical bias, as the 100% result is due to the fact that a single tested sample resulted positive in that season. Regarding the parasites, *Toxascaris leonina* showed the highest prevalence, 16%, followed by the non-identified Coccidia with 8% prevalence, non-identified nematodes with 3%, *Cystoisospora* spp. and Toxocaridae with 2% each, and flagellates, sporulated oocysts and non-sporulated oocysts with 1% each.

Overall, the lion shows a higher consistent parasitic prevalence throughout the year when compared to the other species. The jaguar is the second most affected species of Felidae in the Barcelona Zoo, and the higher parasitic prevalence showed overall by the lions and jaguars could be related to different factors, such as the type of enclosure, it's hygiene and how much contact the animals could easily have with old faeces and/or paratenic or intermediate hosts (Bowman 2021). In the Barcelona Zoo, all of these animal's enclosures are cleaned daily, removing food remains and faeces and washing the interior areas with detergent. The enclosures are also thoroughly disinfected once a month (personal communication, Dr. Vanessa Almagro, 2025).

A company has been hired for pest control, operating with traps and poison. The poison has not been a problem for the health of the Felidae, as the controlled pests do not live in the same area as these predators (personal communication, Dr. Vanessa Almagro, 2025).

Another important factor would be how vulnerable the species is to infection by certain parasites and to develop clinical signs (Lim et al. 2008) as these lead to the coprological testing for parasites. The lions in the zoo have shown to be consistently infected with parasites, hence the chronic preventive treatment and more regular faecal examinations (personal communication, Dr. Vanessa Almagro, 2024).

The Sri Lankan leopards and the Sumatran tigers show a very low quantity of positive cases, even though they were tested more regularly than the jaguars. This could be caused by a number of factors, such as enclosure hygiene, feeding, or even the animals' own immune response and parasite-host interactions to be different than the affected species (Aviruppola et al. 2016; Parsani et al. 2001).

The lions have been the most tested of the four Felidae subspecies. The reasoning behind this is that, aside from the more regular testing due to active parasitic control, it should be noted that there have also been more lions present in the Barcelona Zoo, from 1990 to 2024, than any of the other three Felidae species, as there are records of 12 tested lions, seven different tested Sri Lankan leopards, five tested Sumatran tigers and four tested jaguars. This shows a statistical bias regarding the results in this species, comparing to the other three.

Throughout the seasons, the lions showed higher prevalence during summer, being affected by *T. leonina* and Coccidia in this season. These parasites can infect Felidae through paratenic hosts (Dubey and Fayer 1983; Dubey 2010; Bowman 2021) and one of the factors that could possibly increase parasitic prevalence in the summer, is the breeding season of some rodents peaking during spring (Feng and Himsworth 2014), or, regarding Coccidia, the possibility of infection cycles having more impact in certain seasons, in cattle and other animals whose meats are provided raw (Dubey and Fayer 1983; Dubey 2010; Bowman 2021) as the feeding for lions. However, such conclusions would require further investigation. Furthermore, the Barcelona Zoo provides their Felidae with raw meat that has been previously frozen, therefore working towards avoiding coccidiosis and other parasitic infections (personal communication, Dr. Vanessa Almagro, 2025).

Toxascaris leonina being the most found parasite in these species, with 16% prevalence, shows agreement with the information collected from the literature review. This parasite is incredibly hard to eradicate as it survives extreme conditions and latches to the surface of the floor with great strength (Bowman 2021). It is nearly impossible to remove eggs from enclosures with natural landscapes and floors (for example dirt, grass and trees), which are the type of landscape in the Zoo's Felidae enclosures, and since the eggs need only one week to reach the infective stage, animals can be quickly reinfected (Bowman 2021). Okulewicz (2008), as quoted by Okulewicz et al. (2012) states that the main infection route *T. leonina* possesses to infect caged zoo animals is through paratenic hosts, and it is a possibility that rodents would be, therefore, responsible for the transmission to these animals. The Zoo manages these pests with the help of a company dedicated to such, as previously mentioned.

The ability *Toxascaris leonina* has to resist various environmental conditions (Bowman 2021) is also shown in this study through its presence in the four seasons of the year: during spring in the jaguars with 14% (2/14) prevalence, and in the African lions (1/1); in the summer in the lions with 47% (8/17) prevalence, the most prevalent season for this parasite in the lions (excluding spring); in autumn in the Sri Lankan leopards with 14% (2/14) prevalence and in the lions with 20% (2/10) prevalence; and in winter in the Sumatran tigers with 15% (2/13) prevalence and in the lions with 29% (6/21) prevalence. It doesn't show a pattern in these animals regarding seasons, except that in the lions, it was most prevalent during summer and occurred in all the four seasons.

The second most common parasites were Coccidia with 8% prevalence (11/143).

A study performed by Panayotova-Pencheva (2013) concluded that the most prevalent parasites in European zoos are usually nematodes, followed by protozoans. This statement coincides with the results from this dissertation as the nematodes show a prevalence of 21% and protozoans show a prevalence of 9%. The most common, and only identified, nematodes were from the order Ascaridida, also coinciding with the results from Panayotova-Pencheva (2013), that observed that the most reoccurring nematodes in European zoos were from the Ascaridida order.

The Chi-square testing done on the seasonality of the parasitic prevalence shows values that are mostly higher than 0.05, except the 0.006 value shown in the Toxocaridae prevalence in *Panthera leo melanochaita*, and the 0.047 value in the *P. l. melanochaita*'s seasonality. The 0.047 p-value indicates that the lions may have a seasonality in their parasitic infections, with summer, autumn and perhaps spring (would need further testing) as the seasons during which they are most susceptible to infection.

The 0.006 p-value, and the fact that Toxocaridae was only detected during autumn in the lion, leads to the conclusion of a possible correlation of the parasitic occurrence to autumn. Such is not observed in the rest of the cases, which show a p-value higher than 0.05, indicating that the occurrence of the other parasitic infections may not be related to the season of the year (McHugh 2013).

Regarding the Table 5 measurement of the agreement level between flotation and direct method techniques, a Cohen's kappa value of 0.372 shows a minimal level of agreement (0.21-0.39) (McHugh 2012) between these two techniques, meaning that one is more likely to detect positive parasitic samples, in other words, one is more sensitive. Upon observation of the results in the table, it's safe to say that flotation has higher sensitivity. There are 31 detected positive samples: 16 positive only in flotation, 5 positive only in direct methods, and 10 positive in both methods. Out of the 31 positive samples, flotation detected 26 (16 of which were negative in the direct methods), showing a sensitivity of 84%, whereas the direct methods detected 15 positive samples (of which, only 5 were negative in the flotation examinations), showing a sensitivity of 48%.

The statistical significance, <0.001, being under 0.05 shows that these results are most likely not due to coincidence. It is possible to establish that in this study, flotation was a more sensitive technique than the direct method. This result is as expected, because flotation techniques concentrate the eggs, cysts, larvae, or other parasitic forms for visualization and are usually more sensitive in detecting parasites (Parameshwarappa et al. 2012; Mahmood et al. 2014).

Regarding the Graphs 1-4, they demonstrate the parasitic positivity rate of the four Felidae species at the zoo from 1990 to 2024. As stated before, it's a big possibility that the lack of data, in the years 2010-2014 and 2015-2019, and the scarce registered results from other years, is likely due to lost information in the process of transitioning from paper registry to the online registry on ZIMS.

In Graph 1 it is possible to see that even though the lions have showed higher parasitic prevalence in the past, such numbers occurred in years where very few tests were performed, showing a statistical bias. The years 2020-2024 have the highest registered number of performed tests in the species. One of the reasons would be the fact that the lions have shown repeatedly infected faeces, leading to persistent parasitological control, not only with chronic treatment but also more frequent faecal sampling.

Toxascaris leonina has been a frequent parasite throughout the years in the Southern African lions at the Barcelona Zoo. As mentioned above, it is a difficult parasite to eradicate (Bowman 2021). Coccidia has also been a recurrent parasitic infection in these animals, probably due to ingestion of raw contaminated meat (Dubey and Fayer 1983; Dubey 2010; Bowman 2021).

In Graph 2 we see that jaguars have a low number of registered tests, and a lack of tests from 2000 to 2019. There have been very few tested jaguars, only four, and besides the possibly lost registry, it is a relevant factor to mention when comparing the number of tests in jaguars to the other three Felidae species. In 1990-1994 we observe that the jaguar samples showed a big variety of registered parasites, many of which Coccidia, as well as flagellates and *T. leonina*. The higher Coccidia prevalence could indicate that contaminated raw meat was fed to these animals (Dubey and Fayer 1983; Dubey 2010; Bowman 2021). In 2020-2024 the positivity rates for parasites are lower, showing possible improved parasitic control in the species.

In *Panthera pardus kotiya* parasitic prevalence evolution throughout the years, demonstrated in Graph 3, we can see that this species has been tested many times throughout the years and the parasitic prevalence hasn't been high, nor has there been a persistence of the same parasites in different five-year periods, with infections by *Toxascaris leonina*, Coccidia and nematodes.

The *Panthera tigris sumatrae*, on Graph 4 shows a very disperse parasitic prevalence throughout the decades, with a relatively high prevalence in the low number of tests performed in the 1995-1999 period. The Sumatran tigers' lack of parasitic findings leads us to believe that, at the Barcelona Zoo, they could not be very susceptible to the existing pool of parasites or different enclosures may have an effect in these findings (different access to paratenic hosts) (Bowman 2021).

The tigers were the only ones to show positive results for *Toxoplasma gondii*, but the parasite did not appear in coprology because it is a difficult parasite to find in the faeces, as it is shed for small periods of time post-infection (CUCVM 2018). Since the other three species of Felidae weren't serologically tested for *Toxoplasma gondii* antibodies, there is a possibility that they have been infected as well, because this parasite can easily go unnoticed, as it doesn't typically lead to noticeable clinical signs (Dubey 2010), and is shed in faeces for a short period of time (CUCVM 2018), being hard to detect in faecal exams.

The Sumatran tigers, like the lions, show *T. leonina* infection in the 1995-1999 period, however the lions had positive samples in 1996, and the tigers were positive for this parasite in 1995. With the lack of registered tests on the Sumatran tiger in 1996, it is hard to say if they were infected at the same time as the lions, and if the infection was passed on to the lions from contaminated materials or clothing.

In Graph 5 we can analyse all the previous graphs combined to identify the general prevalence in each parasite throughout the years, with *Toxascaris leonina*, as previously mentioned, showing the highest prevalence, always 17% or higher, when present. Coccidia has also shown to be present in all the four species and they have recurrently reappeared in coprological testing. Murnik et al. (2024), performed a study in which Carnivora's most commonly found parasites were also from the Ascaridida order, followed by Coccidia.

The 2020-2024 period shows the highest number of performed tests in all the year spans which shows growth and improvement in the parasitic screening of these species. This could also be influenced, as previously mentioned, by the fact that all the new tests are immediately registered on the online ZIMS database, therefore being less likely to misplace the information over time.

The common infections amongst different species in the same time periods could be purely coincidental, as many are not occurring in the same year. Nevertheless, these findings could still have happened due to cross contamination from carers' clothing pieces or contaminated materials. It could also be due to contact with the same paratenic hosts or for sharing meat supplies (Bowman 2021).

The differing parasitic prevalence between the four species of Felidae analysed, could be due to different variables in the day to day of these animals. One of the variables is different enclosures, and whether the animal has easy access to pests, such as rodents, or even other disease transmitting animals, that could weaken the Felidae's immune system. Another variable is the hygiene measures carried out by handlers, regarding cleaning, discarding of faeces, usage of individual protection equipment, practices of personal hygiene to prevent transmission of zoonoses, different meat and meat sources for feeding and if they are clear from parasites and/or cooked, and access to clean water. Another relevant aspect is the general wellbeing of the animals, as a more stressed animal may develop a weakened immune system and be therefore more vulnerable to parasitic diseases (Geraghty et al. 1981; Pérez Cordón et al. 2008; Edwards et al. 2019).

7. Conclusion

In conclusion, from this study, we can see that *Toxascaris leonina* is a reoccurring and difficult parasite to eradicate from the Felidae at the Barcelona Zoo, particularly from the African lions, and is the most common parasite amongst their Felidae. *Toxascaris leonina* is followed by Coccidia as the second most prevalent parasites, and records showing it to be a complicated parasite to completely eradicate from the Zoo. The most prevalent parasites in this study were nematodes (*T. leonina*, nematodes, Toxocaridae) followed by protozoans (Coccidia, *Cystoisospora* spp., flagellates, non-sporulated oocysts and sporulated oocysts). Also, the most common, and only identified, nematodes found were from the Ascaridida order.

The Sri Lankan leopards and Sumatran tigers show the lowest infection rates out of all Felidae at the Barcelona Zoo and are probably the most resistant to infection, unlike the Southern African lions and the jaguars, that showed to be the most susceptible to parasitic infection, with higher prevalence rates.

The different species showed different seasons in which their parasitic prevalence was higher: summer for the African lions, autumn for the jaguars and Sri Lankan leopards, and winter for Sumatran tigers.

Toxoplasma gondii is not always easy to be detected, and if an animal is not tested serologically for its antibodies, this parasite can easily go unnoticed. In this dissertation, its antibodies were searched for and detected solely on Sumatran tigers.

Regarding diagnostic methods, flotation techniques were more sensitive than direct methods in the detection of parasites in the faeces of Felidae at the Barcelona Zoo.

In this study we can also see how hard and rigorously the veterinary team and handlers from the Barcelona Zoo have worked in the last five years, to be up to date regarding the parasitic load in Felidae and to maintain a plan for repeated parasitic control of these animals.

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Appendices

Table 1: Database of all the gastrointestinal parasites registered in Felidae from the Barcelona Zoo from 1990 to 2024

#- Case number; **Date-** dd.mm.yy; **Tests:** 0- not performed, 1- performed; **Parasites:** 0- not present, 1- present; **Flot.-** Flotation; **Sporul.-** Sporulated

				Tests											Parasites										
#	**Species**	**Animal**	**Date**	**$NaNO_3$ Flot.**	**$ZnSO_4$ Flot.**	**Sugar Flot.**	**NaCl Flot.**	**Non-Specified Flot.**	**Direct Exam**	**Sedimentation**	**Stain MIF**	**Acid Fast**	**Baermann**	**Serum Analysis**	***Cystoisospora***	**Coccidia**	**Flagellates**	**Nematodes**	***T. leonina***	**Toxocaridae**	***T. gondii***	**Sporul. Oocysts**	**Non-Sporul. Oocysts**	**Season**	**Year Span**
1	*P. l. melanochaita*	1	29.01.24	1	0	0	0	0	1	0	0	0	1	0	0	0	0	0	1	0	0	0	0	Winter	2020-2024
2	*P. l. melanochaita*	1	28.10.23	1	0	0	0	0	1	0	0	0	1	0	0	0	0	0	0	1	0	0	0	Autumn	2020-2024
3	*P. l. melanochaita*	2	28.10.23	1	0	0	0	0	1	0	0	0	1	0	0	0	0	0	0	1	0	0	0	Autumn	2020-2024
4	*P. l. melanochaita*	3	28.10.23	1	0	0	0	0	1	0	0	0	1	0	0	0	0	0	0	1	0	0	0	Autumn	2020-2024
5	*P. l. melanochaita*	1	17.07.23	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	1	0	0	0	0	Summer	2020-2024
6	*P. l. melanochaita*	2	17.07.23	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	1	0	0	0	0	Summer	2020-2024
7	*P. l. melanochaita*	3	17.07.23	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	1	0	0	0	0	Summer	2020-2024
8	*P. l. melanochaita*	1	08.05.23	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	1	0	0	0	0	Spring	2020-2024
9	*P. l. melanochaita*	1	11.02.23	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	Winter	2020-2024
10	*P. l. melanochaita*	1	20.06.22	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	1	0	0	0	0	Summer	2020-2024
11	*P. l. melanochaita*	2	20.06.22	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	1	0	0	0	0	Summer	2020-2024
12	*P. l. melanochaita*	3	20.06.22	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	1	0	0	0	0	Summer	2020-2024
13	*P. l. melanochaita*	1	11.08.20	1	0	0	0	0	1	0	0	0	0	0	0	1	0	0	0	0	0	0	0	Summer	2020-2024
14	*P. l. melanochaita*	4	11.08.20	1	0	0	0	0	1	0	0	0	0	0	0	1	0	0	0	0	0	0	0	Summer	2020-2024
15	*P. l. melanochaita*	5	11.08.20	1	0	0	0	0	1	0	0	0	0	0	0	1	0	0	0	0	0	0	0	Summer	2020-2024
16	*P. l. melanochaita*	6	11.08.20	1	0	0	0	0	1	0	0	0	0	0	0	1	0	0	0	0	0	0	0	Summer	2020-2024
17	*P. l. melanochaita*	1	03.02.20	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	1	0	0	0	0	Winter	2020-2024
18	*P. l. melanochaita*	4	03.02.20	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	1	0	0	0	0	Winter	2020-2024
19	*P. l. melanochaita*	5	03.02.20	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	1	0	0	0	0	Winter	2020-2024
20	*P. l. melanochaita*	6	03.02.20	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	1	0	0	0	0	Winter	2020-2024

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				Tests											Parasites										
#	**Species**	**Animal**	**Date**	**$NaNO_3$ Flot.**	**$ZnSO_4$ Flot.**	**Sugar Flot.**	**NaCl Flot.**	**Non-Specified Flot.**	**Direct Exam**	**Sedimentation**	**Stain MIF**	**Acid Fast**	**Baermann**	**Serum Analysis**	***Cystoisospora***	**Coccidia**	**Flagellates**	**Nematodes**	***T. leonina***	**Toxocaridae**	***T. gondii***	**Sporul. Oocysts**	**Non-Sporul. Oocysts**	**Season**	**Year Span**
21	*P. l. melanochaita*	7	13.07.06	1	0	0	0	0	1	0	0	0	0	0	0	1	0	0	0	0	0	0	0	Summer	2005-2009
22	*P. l. melanochaita*	5	10.11.04	0	1	0	0	0	1	0	0	0	0	0	0	0	0	0	1	0	0	0	0	Autumn	2000-2004
23	*P. l. melanochaita*	6	10.11.04	0	1	0	0	0	1	0	0	0	0	0	0	0	0	0	1	0	0	0	0	Autumn	2000-2004
24	*P. l. melanochaita*	8	30.07.96	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	Summer	1995-1999
25	*P. l. melanochaita*	9	30.07.96	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	Summer	1995-1999
26	*P. l. melanochaita*	10	15.12.93	0	0	1	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	Winter	1990-1994
27	*P. l. melanochaita*	3	29.02.24	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	2020-2024
28	*P. l. melanochaita*	2	29.02.24	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	2020-2024
29	*P. l. melanochaita*	1	29.02.24	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	2020-2024
30	*P. l. melanochaita*	1	14.11.22	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Autumn	2020-2024
31	*P. l. melanochaita*	1	03.07.21	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Summer	2020-2024
32	*P. l. melanochaita*	5	03.07.21	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Summer	2020-2024
33	*P. l. melanochaita*	6	03.07.21	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Summer	2020-2024
34	*P. l. melanochaita*	4	21.10.20	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Autumn	2020-2024
35	*P. l. melanochaita*	1	21.10.20	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Autumn	2020-2024
36	*P. l. melanochaita*	5	21.10.20	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Autumn	2020-2024
37	*P. l. melanochaita*	6	21.10.20	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Autumn	2020-2024
38	*P. l. melanochaita*	7	10.07.06	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Summer	2005-2009
39	*P. l. melanochaita*	10	17.01.96	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	1995-1999
40	*P. l. melanochaita*	8	17.01.96	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	1995-1999

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#	Species	Animal	Date	Tests											Parasites									Season	Year Span
				$NaNO_3$ Flot.	$ZnSO_4$ Flot.	Sugar Flot.	NaCl Flot.	Non-Specified Flot.	Direct Exam	Sedimentation	Stain MIF	Acid Fast	Baermann	Serum Analysis	*Cystoisospora*	Coccidia	Flagellates	Nematodes	*T. leonina*	Toxocaridae	*T. gondii*	Sporul. Oocysts	Non-Sporul. Oocysts		
41	*P. l. melanochaita*	11	17.01.96	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	1995-1999
42	*P. l. melanochaita*	9	17.01.96	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	1995-1999
43	*P. l. melanochaita*	10	28.02.95	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	1995-1999
44	*P. l. melanochaita*	12	28.02.95	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	1995-1999
45	*P. l. melanochaita*	9	28.02.95	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	1995-1999
46	*P. l. melanochaita*	10	04.01.95	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	1995-1999
47	*P. l. melanochaita*	12	04.01.95	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	1995-1999
48	*P. l. melanochaita*	9	04.01.95	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	1995-1999
49	*P. l. melanochaita*	10	08.12.93	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	1990-1994
50	*P. onca*	13	08.05.23	1	0	0	0	0	1	0	0	0	0	0	0	1	0	0	0	0	0	0	0	Spring	2020-2024
51	*P. onca*	13	14.11.22	1	0	0	0	0	1	0	0	0	0	0	0	0	0	1	0	0	0	0	0	Autumn	2020-2024
52	*P. onca*	13	14.09.20	1	0	0	0	0	1	0	0	0	0	0	0	0	0	1	0	0	0	0	0	Autumn	2020-2024
53	*P. onca*	14	06.10.92	0	0	0	0	0	0	1	1	1	0	0	0	0	1	0	0	0	0	1	1	Autumn	1990-1994
54	*P. onca*	14	13.03.90	0	0	1	1	0	1	0	0	0	0	0	1	1	0	0	1	0	0	0	0	Spring	1990-1994
55	*P. onca*	15	13.03.90	0	0	1	1	0	1	0	0	0	0	0	1	1	0	0	1	0	0	0	0	Spring	1990-1994
56	*P. onca*	13	20.11.23	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Autumn	2020-2024
57	*P. onca*	13	14.03.22	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	2020-2024
58	*P. onca*	16	14.03.22	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	2020-2024
59	*P. onca*	16	29.09.21	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Autumn	2020-2024
60	*P. onca*	13	29.09. 21	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Autumn	2020-2024

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#	Species	Animal	Date	Tests											Parasites									Season	Year Span
				NaNO3 Flot.	ZnSO4 Flot.	Sugar Flot.	NaCl Flot.	Non-Specified Flot.	Direct Exam	Sedimentation	Stain MIF	Acid Fast	Baermann	Serum Analysis	*Cystoisospora*	Coccidia	Flagellates	Nematodes	*T. leonina*	Toxocaridae	*T. gondii*	Sporul. Oocysts	Non-Sporul. Oocysts		
61	*P. onca*	16	06.03.21	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	2020-2024
62	*P. onca*	13	06.03.21	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	2020-2024
63	*P. onca*	16	01.02.21	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	2020-2024
64	*P. onca*	14	02.03.95	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	1995-1999
65	*P. onca*	14	06.01.95	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	1995-1999
66	*P. onca*	14	09.10.92	0	0	0	0	0	0	1	1	1	0	0	0	0	0	0	0	0	0	0	0	Autumn	1990-1994
67	*P. onca*	14	10.05.90	0	0	0	1	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	1990-1994
68	*P. onca*	15	10.05.90	0	0	0	1	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	1990-1994
69	*P. onca*	14	14.03.90	0	0	1	1	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	1990-1994
70	*P. onca*	15	14.03.90	0	0	1	1	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	1990-1994
71	*P. onca*	14	07.03.90	0	0	1	1	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	1990-1994
72	*P. onca*	15	07.03.90	0	0	1	1	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	1990-1994
73	*P. p. kotiya*	17	14.11.22	1	0	0	0	0	1	0	0	0	0	0	0	0	0	1	0	0	0	0	0	Autumn	2020-2024
74	*P. p. kotiya*	18	14.11.22	1	0	0	0	0	1	0	0	0	0	0	0	0	0	1	0	0	0	0	0	Autumn	2020-2024
75	*P. p. kotiya*	19	13.03.08	1	0	0	0	0	1	0	0	0	0	0	0	1	0	0	0	0	0	0	0	Spring	2005-2009
76	*P. p. kotiya*	19	10.03.08	1	0	0	0	0	1	0	0	0	0	0	0	1	0	0	0	0	0	0	0	Spring	2005-2009
77	*P. p. kotiya*	20	09.10.92	0	0	0	0	0	0	1	1	1	0	0	0	0	0	0	1	0	0	0	0	Autumn	1990-1994
78	*P. p. kotiya*	21	09.10.92	0	0	0	0	0	0	1	1	1	0	0	0	0	0	0	1	0	0	0	0	Autumn	1990-1994
79	*P. p. kotiya*	17	20.11.23	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Autumn	2020-2024
80	*P. p. kotiya*	18	20.11.23	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Autumn	2020-2024

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#	Species	Animal	Date	Tests											Parasites									Season	Year Span
				$NaNO_3$ Flot.	$ZnSO_4$ Flot.	Sugar Flot.	NaCl Flot.	Non-Specified Flot.	Direct Exam	Sedimentation	Stain MIF	Acid Fast	Baermann	Serum Analysis	*Cystoisospora*	Coccidia	Flagellates	Nematodes	*T. leonina*	Toxocaridae	*T. gondii*	Sporul. Oocysts	Non-Sporul. Oocysts		
81	*P. p. kotiya*	17	08.05.23	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	2020-2024
82	*P. p. kotiya*	18	08.05.23	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	2020-2024
83	*P. p. kotiya*	17	14.03.22	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	2020-2024
84	*P. p. kotiya*	18	14.03.22	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	2020-2024
85	*P. p. kotiya*	17	29.09.21	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Autumn	2020-2024
86	*P. p. kotiya*	22	29.09.21	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Autumn	2020-2024
87	*P. p. kotiya*	17	06.03.21	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	2020-2024
88	*P. p. kotiya*	22	06.03.21	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	2020-2024
89	*P. p. kotiya*	17	14.09.20	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Autumn	2020-2024
90	*P. p. kotiya*	22	14.09.20	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Autumn	2020-2024
91	*P. p. kotiya*	17	08.03.20	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	2020-2024
92	*P. p. kotiya*	22	08.03.20	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	2020-2024
93	*P. p. kotiya*	23	13.03.08	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	2005-2009
94	*P. p. kotiya*	23	10.03.08	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	2005-2009
95	*P. p. kotiya*	19	04.03.08	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	2005-2009
96	*P. p. kotiya*	23	04.03.08	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	2005-2009
97	*P. p. kotiya*	19	03.03.08	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	2005-2009
98	*P. p. kotiya*	23	03.03.08	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	2005-2009
99	*P. p. kotiya*	20	30.07.96	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Summer	1995-1999
100	*P. p. kotiya*	20	28.02.95	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	1995-1999

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#	Species	Animal	Date	Tests: NaNO3 Flot.	ZnSO4 Flot.	Sugar Flot.	NaCl Flot.	Non-Specified Flot.	Direct Exam	Sedimentation	Stain MIF	Acid Fast	Baermann	Serum Analysis	Parasites: *Cystoisospora*	Coccidia	Flagellates	Nematodes	*T. leonina*	Toxocaridae	*T. gondii*	Sporul. Oocysts	Non-Sporul. Oocysts	Season	Year Span
101	*P. p. kotiya*	21	28.02.95	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	1995-1999
102	*P. p. kotiya*	20	05.01.95	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	1995-1999
103	*P. p. kotiya*	21	05.01.95	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	1995-1999
104	*P. p. kotiya*	20	13.10.92	0	0	0	0	0	0	1	1	1	0	0	0	0	0	0	0	0	0	0	0	Autumn	1990-1994
105	*P. p. kotiya*	21	13.10.92	0	0	0	0	0	0	1	1	1	0	0	0	0	0	0	0	0	0	0	0	Autumn	1990-1994
106	*P. p. kotiya*	20	06.10.92	0	0	0	0	0	0	1	1	1	0	0	0	0	0	0	0	0	0	0	0	Autumn	1990-1994
107	*P. p. kotiya*	21	06.10.92	0	0	0	0	0	0	1	1	1	0	0	0	0	0	0	0	0	0	0	0	Autumn	1990-1994
108	*P. p. kotiya*	20	09.03.90	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	1990-1994
109	*P. p. kotiya*	21	09.03.90	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	1990-1994
110	*P. p. kotiya*	20	08.03.90	0	0	1	1	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	1990-1994
111	*P. p. kotiya*	21	08.03.90	0	0	1	1	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	1990-1994
112	*P. p. kotiya*	20	07.03.90	0	0	1	1	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	1990-1994
113	*P. p. kotiya*	21	07.03.90	0	0	1	1	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	1990-1994
114	*P. t. sumatrae*	24	11.02.23	0	0	0	0	1	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	Winter	2020-2024
115	*P. t. sumatrae*	26	06.01.95	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	Winter	1995-1999
116	*P. t. sumatrae*	27	06.01.95	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	Winter	1995-1999
117	*P. t. sumatrae*	28	29.01.24	1	0	0	0	0	1	0	0	0	1	0	0	0	0	0	0	0	0	0	0	Winter	2020-2024
118	*P. t. sumatrae*	24	29.01.24	1	0	0	0	0	1	0	0	0	1	0	0	0	0	0	0	0	0	0	0	Winter	2020-2024
119	*P. t. sumatrae*	28	28.10.23	1	0	0	0	0	1	0	0	0	1	0	0	0	0	0	0	0	0	0	0	Autumn	2020-2024
120	*P. t. sumatrae*	24	28.10.23	1	0	0	0	0	1	0	0	0	1	0	0	0	0	0	0	0	0	0	0	Autumn	2020-2024

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#	Species	Animal	Date	Tests											Parasites									Season	Year Span
				$NaNO_3$ Flot	$ZnSO_4$ Flot	Sugar Flot	NaCl Flot.	Non-Specified Flot.	Direct Exam	Sedimentation	Stain MIF	Acid Fast	Baermann	Serum Analysis	*Cystoisospora*	Coccidia	Flagellates	Nematodes	*T. leonina*	Toxocaridae	*T. gondii*	Sporul. Oocysts	Non-Sporul. Oocysts		
121	*P. t. sumatrae*	28	17.07.23	1	0	0	0	0	1	0	0	0	1	0	0	0	0	0	0	0	0	0	0	Summer	2020-2024
122	*P. t. sumatrae*	24	17.07.23	1	0	0	0	0	1	0	0	0	1	0	0	0	0	0	0	0	0	0	0	Summer	2020-2024
123	*P. t. sumatrae*	28	08.05.23	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	2020-2024
124	*P. t. sumatrae*	24	08.05.23	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Spring	2020-2024
125	*P. t. sumatrae*	28	11.02.23	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	2020-2024
126	*P. t. sumatrae*	24	16.07.22	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Summer	2020-2024
127	*P. t. sumatrae*	28	16.07.22	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Summer	2020-2024
128	*P. t. sumatrae*	24	03.07.21	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Summer	2020-2024
129	*P. t. sumatrae*	28	03.07.21	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Summer	2020-2024
130	*P. t. sumatrae*	24	04.12.20	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	2020-2024
131	*P. t. sumatrae*	28	04.12.20	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	2020-2024
132	*P. t. sumatrae*	24	11.08.20	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Summer	2020-2024
133	*P. t. sumatrae*	28	11.08.20	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Summer	2020-2024
134	*P. t. sumatrae*	24	03.02.20	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	2020-2024
135	*P. t. sumatrae*	28	03.02.20	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	2020-2024
136	*P. t. sumatrae*	25	25.09.08	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Autumn	2005-2009
137	*P. t. sumatrae*	25	23.09.08	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Autumn	2005-2009
138	*P. t. sumatrae*	25	03.09.08	0	0	0	0	0	0	0	1	0	0	1	0	0	0	0	0	0	0	0	0	Autumn	2005-2009
139	*P. t. sumatrae*	26	12.01.07	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	2005-2009
140	*P. t. sumatrae*	26	28.02.95	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	1995-1999

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#	Species	Animal	Date	Tests											Parasites									Season	Year Span
				$NaNO_3$ Flot	$ZnSO_4$ Flot	Sugar Flot	NaCl Flot.	Non-Specified Flot.	Direct Exam	Sedimentation	Stain MIF	Acid Fast	Baermann	Serum Analysis	*Cystoisospora*	Coccidia	Flagellates	Nematodes	*T. leonina*	Toxocaridae	*T. gondii*	Sporul. Oocysts	Non-Sporul. Oocysts		
141	*P. t. sumatrae*	27	28.02.95	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	Winter	1995-1999
142	*P. t. sumatrae*	24	04.04.18	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	Spring	2015-2019
143	*P. t. sumatrae*	25	09.09.08	1	0	0	0	0	1	0	0	0	0	1	0	0	0	0	0	0	1	0	0	Autumn	2005-2009
144	*P. t. sumatrae*	25	29.06.07	1	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	1	0	0	Summer	2005-2009
145	*P. t. sumatrae*	25	24.10.08	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	1	0	0	Autumn	2005-2009
146	*P. t. sumatrae*	24	10.10.08	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	1	0	0	Autumn	2005-2009