**A survey of intestinal parasites infecting captive large cats of central Florida.**

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**Abstract.** Feral domestic cats are known to host a diversity of intestinal parasites, with typical infections including both protozoans and helminths. Common symptoms of infection include vomiting and diarrhea, and consequential anemia and dehydration ultimately weaken the host and its immune system. Infections can be prevented with anti-parasitic treatment monthly. Parasites of domestic cat hosts should be of particular interest, as many species can be transmitted to humans and wildlife. In Florida, large feral cat populations have become commonplace in urban and natural environments, suggesting their parasites could be transmitted to native or non-native large cat species. Little work has previously surveyed the species infecting larger cat species in Florida. This survey sought to determine (1) the protozoan and helminth species infecting captive large cat hosts in reserves within central Florida and (2) the effectiveness of the treatment program. Six species were surveyed: tiger (*Panthera tigris*), lion (*Panthera leo*), leopard (*Panthera pardus*),cougar (*Puma concolor*), serval (*Leptailurus serval*), and bobcat (*Lynx rufus*) from Big Cat Rescue in Tampa, FL and Central Florida Animal Reserve in Cocoa, FL. The cats from Big Cat Rescue were treated with ivermectin monthly and fenbendazole on alternating months, and those from Central Florida Animal Reserve were treated with ivermectin on alternating months. Fecal samples were collected from each species monthly, prepared by centrifugal flotation in Sheather’s sugar solution, and prepared on slides for parasite examination. The results of this survey suggest that the anti-parasitic treatments were generally effective; however, some cats still hosted parasite infections.

**Introduction**

The variety and numbers of feral domestic cats (*Felis catis)* in Florida are large, with estimations nearing 10 million free-roaming cats (Hatley, 2003). Many of these cats live in clowders or colonies, thus breeding successfully and maintaining populations (ACES, 2013). The high numbers of these cats are of particular concern due to the similar variety of parasitic infections these cats can carry (Adams et al., 2008). Because feral cats are likely to feed on any available prey species, depending on its environment (Adams et al., 2008), understanding the effect of these animals on the environment, including the native wildlife, can be a complex task. It has been known that feral cats possess the ability to transmit infections to humans, with serious zoonotic diseases such as toxoplasmosis receiving notable attention (The Wildlife Society, 2011). A study from 2010 found that higher levels of *Toxocara cati* in soil samples from urban parks compared to rural areas were associated with urban feral cats (Lee et al.). Children are common hosts to these parasites, due to increased contact with soil or sandboxes, and to a decrease in hand-washing practice (CDC, 2014). In addition, because many of these parasite eggs or oocysts are found in garden soil, gardeners are also at risk of infection (Robertson and Thompson, 2002). Thus, understanding the prevalence of these parasites is important to study, and much work has been done to reach this understanding.

 Many of these parasites are fairly flexible in their requirements of definitive hosts, so it is possible that large cats could serve as the definitive host for some of these parasites. In Florida, a number of wildlife refuges exist, and many of these house large cats. These cats are of particular interest, because they are genetically similar to an already large group of “wild” animals in Florida, the feral cats. This makes them a remarkably good potential host for these parasites. In addition, many of these refuges are non-profit facilities and function based on the work of volunteers, admission prices of visitors, and donations. Due to this, it is likely that many of these facilities cannot afford the highest technological facilities, and potentially cannot afford the highest quality of preventative treatment. It follows, then, that these facilities may be housing cats with parasite infections common in feral domestic cats.

 The foremost purpose of this study was to survey the prevalence of gastrointestinal parasites infecting captive big cats in central Florida. Big cat reserves, as with many wildlife refuges, house animals in enclosures typically surrounded by chain-link fences. These enclosures allow for the potential entry of numerous intermediate hosts, thus begging the question of the possibility of transfer of infections from native fauna to the big cats. We surveyed two reserves: Big Cat Rescue and Central Florida Animal Reserve. Big Cat Rescue (BCR) is located in Tampa, Florida, and is home to approximately 100 wild cats, rescued from owners, performance acts, or from the wild to protect from hunters. They are a non-profit organization accredited by the Global Federation of Sanctuaries (Big Cat Rescue website). Central Florida Animal Reserve (CFAR) is located in Cocoa, Florida and houses over 40 big cats. All cats at CFAR were captive-born, being removed from “pet situations”, rescued, or born at the facility. This facility is a non-profit organization (Central Florida Animal Reserve website).

 The secondary purpose of this study was to compare the effectiveness of two treatments plans. BCR utilizes an all-encompassing preventative treatment plan, giving appropriate doses of ivermectin monthly, and fenbendazole on alternating months. CFAR utilizes a less inclusive preventative treatment plan, dosing with ivermectin on alternating months. Both facilities also treat any infections that may be seen with the appropriate treatment (W. Hecht, personal communication, April 10, 2013). These preventative treatment plans utilize well known products acceptable for use in big cats. Ivermectin is an antiparasitic drug used against roundworms and arthropod parasites, as well as heartworm (Campbell and Benz, 1984). Fenbendazole, often referred to by the brand name “Panacur”, is used against roundworms, hookworms, whipworms, and tapeworms, as well as some protozoa, including *Giardia spp.* (Foster and Smith, 2007). We expected that the less-inclusive plan used by CFAR would be associated with a higher prevalence of infection between the two sites, due to the less frequent dosages, and the less-inclusive nature of ivermectin as a treatment. It would thus follow that we would see more hookworms, whipworms, or tapeworms at CFAR, considering ivermectin does not target these helminths specifically.

**Methods**

 Collections were made from the two reserves over a total of six months, beginning in October 2013, and ending in March 2014. Samples were taken from BCR in October, November, January, February, and March. One sample was taken each month from each of the five cats sampled for a total of 25 different samples. The cats sampled at Big Cat Rescue were: tiger (*Panthera tigris*), leopard (*Panthera pardus*), cougar (*Puma concolor*), serval (*Leptailurus serval*), and bobcat (*Lynx rufus*). Samples were taken from CFAR in October, November, and March. The November collections from CFAR were not prepared due to timing of the collection; we were not able to prepare the samples due to the Florida Southern College calendar. One sample was taken each month for each of the four cats for a total of 8 samples. The cats sampled at CFAR were: tiger (*Panthera* tigris), lion (*Panthera leo*), leopard (*Panthera pardus*), and cougar (*Puma concolor*).

 Upon collection, the samples were refrigerated and stored for no more than 72 hours. Slides were prepared by centrifugal fecal flotation using Sheather’s sugar solution, specific gravity 1.27. A standard fecal flotation procedure was followed (Veterinary Learning Systems, 2006). Two to five grams of feces were mixed with 10 mL of flotation solution, and then strained. The strained mixture was poured into a 15 mL centrifuge tube, and the tubes were spun in a centrifuge at 3400 rpm for 5 minutes. The tubes were removed, filled with flotation solution to form a slight meniscus, and coverslips were placed on the tubes. After ten minutes, the coverslips were placed on microscope slides. Most slides were observed within the following month, often after the sugar solution had dried. Slides were examined on 40x objective light microscopy for parasite eggs. Any potential parasites were photographed and identified using *Feline Clinical Parasitology*. When necessary for photographic evidence, eggs were examined on 100x objective under oil immersion and photographed. After slide preparation, fecal samples were stored in 85% ethanol. To observe for coccidians, samples were stored in potassium dichromate solution for preservation and then prepared by the fecal flotation procedure described above. Potassium dichromate solution is recommended for the preservation of coccidian oocysts (Bush, 2009). Slides were then observed under oil immersion on 100x objective for species identification.

**Results**

 Five helminth species were identified, including one nematode (*Toxocara cati*), two trematodes (*Alaria marcianae* and *Paragonimus kellicotti*) and two tapeworms (*Taenia taeniaeformis* and *Spirometra mansonoides*). Some species were found in more than one cat, but all were found within only one reserve (Figures 1 and 2).

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| --- | --- | --- | --- |
| **Parasite species** | **Host** | **Location** | **Date** |
| *Toxocara cati* | Cougar | CFAR | October 2013 |
| Leopard | CFAR | October 2013 |
| Lion | CFAR | March 2014 |
| *Alaria marcianae* | Lion | CFAR | March 2014 |
| *Paragonimus kellicotti* | Leopard | BCR | January 2014 |
| Serval | BCR | January 2014 |
| Serval | BCR | February 2014 |
| *Taenia taeniaeformis* | Leopard | BCR | January 2014 |
| *Spirometra mansonoides* | Bobcat | BCR | February 2014 |

**Figure 1.** Helminth parasites and their respective hosts, locations, and dates identified from fecal samples.



**Figure 2.** Helminth eggs photos at 400x. **a.** *Toxocara cati* **b.** *Alaria marcianae* **c.** *Paragonimus kellicotti* **d.** *Taenia taeniaeformis* **e.** *Spirometra mansonoides*.

 Upon observation at 100x objective, coccidians were observed, but were not identifiable to species. Prevalence of helminth infection in all cats sampled was 24%. Prevalence at Big Cat Rescue and Central Florida Animal Reserve independently were 20% and 50%, respectively.

**Discussion**

 We found that many cats carried low-level infections, despite preventative anti-parasitic treatment. Each species of parasite was found at only one site, so it does not appear that any species is more likely to be found in big cats universally.

 We used an ecological explanation of infection to confirm the identification of parasite species. *T. cati* and *T. taeniaeformis* utilize similar hosts in their life cycles, both involving a small mammal as either a paratenic or intermediate host, respectively. This small mammal can be a mouse or rat, or potentially another small cat or raccoon. *T. cati* completes its life cycle when infective eggs are ingested by the definitive host. Infective eggs are those that have developed outside the definitive host for at least one month. However, *Toxocara* eggs can survive much longer than one month in the environment, as they can resist harsher environmental conditions. *T. cati* can be passed to a paratenic host, such as a small mammal, when that host ingests the infective eggs from the definitive host’s feces. Infective eggs can be ingested from either the environment or the paratenic host by the definitive host, in this case a big cat. When the eggs are ingested by a cat, the larvae hatches and migrates to the lungs. There, development continues into a third-stage larvae, which travels toward the throat. There, larvae elicit coughing, which often leads to their swallowing, where they return to the intestinal tract. In the intestines, larvae mature, mate, and produce eggs, which are then passed in the feces (Johnstone, 2000). *T. taeniaeformis* strays from this life cycle slightly, as it requires an intermediate, rather than paratenic, host for development. When a small mammal ingests the eggs from a definitive host’s feces, the eggs hatch in the intestines, and the larvae move to the liver, where they mature to the strobilocercus stage, the infective stage. When the definitive host ingests the intermediate host, the larval tapeworm attaches to the gut wall, and the adult tapeworm develops. The adult then produces eggs, which are shed in the definitive host’s feces (Nolan, 2004).

To understand the transmission of these two parasites to the big cats of the study, we must consider the environment of the enclosures. All the cats are kept in caged enclosures, typically enclosed in a chain-link style fencing. These enclosures aim to keep out larger mammal species, such as domestic feral cats or raccoons, but cannot keep out smaller mammals such as mice or rats. Both sites have the potential for this transmission to occur. This understanding validates the explanation of transmission to the big cats, and helps explain a potential method of parasite infection control.

The life cycle of *P. kellicotti* involves a snail and crustacean as the first and second intermediate hosts. Similarly, the life cycle of *A. marcianae* requires a snail and tadpole first and second intermediate host. The *S. mansonoides* life cycle involves a copepod and amphibian or fish first and second intermediate host. *P. kellicotti* involves a number of intermediate stages. After egg maturation, the miracidium hatches in water, where it locates and penetrates a snail. The snail hosts development of the sporocysts, redia, and cercariae, which emerge from the snail. The cercariae are free-swimming and penetrate a crustacean, where they encyst as metacercariae. When the crustacean is consumed by the definitive host, the metacercariae excyst and penetrate the gut wall. These flukes migrate to the lungs, where they are often found in a cyst. The cyst opens to a bronchiole, where the eggs are released. When the eggs are moved upward toward the throat, they are swallowed and passed out through the feces (Nolan, 2004). The life cycle of *A. marcianae* is very similar to that of *P. kellicotti*, with the eggs hatching in fresh water and the miracidium entering snails, where sporocysts and cercariae develop. The cercariae penetrate a tadpole, where they develop into mesocercariae. The mesocercariae can remain undeveloped in a variety of paratenic hosts, until a cat ingests the tadpole or paratenic host. When the cat ingests mesocercariae, they migrate into the lungs, where they develop into metacercariae, move to the trachea, and are swallowed. The adult worm develops within the small intestine, produces eggs, and the eggs are shed in the feces (Nolan, 2004).The life cycle of *S. mansonoides* is a bit distinct, as the egg hatching in water releases a free-swimming coracidium, which is ingested by a copepod. Within the copepod, a procercoid develops, and when an amphibian second intermediate host ingests the copepod, the pleurocercoid develops. The pleurocercoid can be passed to various paratenic hosts without developing further. When the pleurocercoid is ingested by the definitive host, either in the second intermediate host or the paratenic host, the worm attaches to the small intestine and develops proglottids, which eventually release eggs that are shed in the feces (Nolan, 2004).

These three life cycles require access to a water source for the completion of the life cycle. According to topological data and photographs of the site found on Google Maps, BCR has enclosures with significant access to natural and artificial water sources. This explains how multiple species requiring an aquatic environment at some point during their life cycle could be transmitted to a terrestrial host. At CFAR, there is less access to natural water, as there is a ten-foot high barrier separating all the enclosures from these natural water features (S. Farrar, personal communication, April 7, 2014). However, if the enclosures feature smaller, artificial water features, these could be sufficient to host snails and tadpoles. Similarly, the movement of frogs around terrestrial areas of Florida, especially during rainy nights, is very common, and it is not unlikely that the cats could catch and consume a frog hosting the mesocercariae of *A. marcianae*. It is logical that *P. kellicotti* and *S. mansonoides* were not identified in the cats at CFAR, because hosting a crustacean or copepod population significant enough to spread infection is far less likely given the lack of natural water sources available. At BCR, this is a higher possibility, and these life cycles explain the likely source of infection.

 Understanding how animals are infected is the first step in controlling infection. There are two potential ways to control parasitic infection, either by preventing infection through the parasite life cycle, or by treating or preventing infection through medications. The problems with medicinal control are two-fold; not only are the cats likely experiencing a higher level of infection, but the treatment costs are also likely to be higher, because infections will likely reoccur. An alternative method to treatment is preventing infection by controlling the parasite life cycle. In these situations, this will likely mean controlling access of the first and second intermediate hosts or paratenic hosts to the cats. These potential controls will look different at the two sites. For Big Cat Rescue, this will mean controlling access to the enclosures. This would likely be a difficult task, as mice and frogs are generally small and difficult to keep out of an enclosure, especially with chain-link style fences. For future purposes, moving enclosures away from natural water sources could be a first step for control. In addition, gaining a better understanding of the cats’ behavior, specifically when keepers are not directly overseeing would help to discover which animals are serving as intermediate hosts, by seeing which hosts the cats may be eating. This could be accomplished through 24-hour camera surveillance within the reserve. Knowing the exact hosts gives a target for control, and therefore a better basis for developing a control plan. For Central Florida Animal Reserve, similar methods could be utilized, though the cats are already separated from natural water sources. At this site, the focus should fall more on enclosure structure and more intensive surveillance of the cats.

 The more common method for controlling parasites is using medication for preventative treatment or for infections that occur. Both sites currently use this method of control, but do so at different levels. Comparing the prevalence of infection at the two sites, the prevalence is much higher at CFAR. With the understanding that samples sizes are both small and not consistent between the two sites, we use these values as a starting ground for discussion of prevention effectiveness. The higher prevalence at CFAR is likely a reflection of the nature of the dosage chosen: ivermectin does not target as many parasites as fenbendazole, and also is not used as often as at BCR. Thus, a dosage plan including a wider-covering medication, as well as increasing the frequency of dosage, would likely lower the prevalence of parasite infections. However, parasites of many types were present at BCR as well, despite the more intensive treatment.

 The final question to address is the practical need for a control plan. Control methods are, realistically, directly dependent on the funds available. Especially for these wildlife sanctuaries, funds must be used very responsibly, as they are non-profit organizations. None of the cats surveyed showed signs of heavy infection, and this begs the question of the necessity of complete control of parasites. The keepers at these reserves can notice parasite infection, and each site has a control plan for parasites that do cause a level of infection sufficient to cause symptoms. At the low intensity seen in the cats surveyed, it is likely that none of these infections are high enough to cause symptoms. We also saw that with only one exception, no cats held an infection within a species over multiple months. The exception to this is the serval at BCR. The servals tested in January and February 2014 hosted an infection of *P. kellicotti*. Samples came from unique cats each month, so the presence of this parasite was found in two distinct cats, not in the same cat both months. Because cats of the same species are kept in closer proximity than cats of differing species, it is possible that this infection was spread from one cat to another, but it is also possible that the infection was gained independently. No other cats appeared to pass the infections to other cats of their species with any consistency. With this understanding, we can say that the preventative treatments at both sites are generally effective.

 We conclude that though infections do occur in the big cats at both reserves, that the intensities are indeed low, and therefore the preventative treatments used by the two sites are generally effective. We have included recommendations for further parasite control, but conclude that the preventative treatment appears to be sufficient to protect the cats in these sanctuaries. Further work on this study should include the use of fecal sedimentation tests to ensure detection of all parasite eggs, additional samples and samples from more sites, and consistency in collection times during the treatment cycle. Gathering more data will develop a more accurate understanding of infection intensity.

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