Should monkeys wash their hands and feet: A pilot-study on sources of zoonotic parasite exposure

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**ABSTRACT**

Human exposure to zoonotic parasites via contaminated soil is a much studied area. Less research has been performed on exposure via contact with animals and surfaces such as picnic tables with which infected animals might be in contact. On St. Kitts, wild African green monkeys (AGM; *Chlorocebus aethiops sabaenus*), which are known to have zoonotic parasites, roam freely in areas with no door dining facilities and are used in the tourist industry. In this study, the hands and feet of eight AGM and picnic tables where AGM were known to walk on were examined for parasitic organisms. Six of the AGM had parasitic organisms on their hands and/or feet. *Trichuris* spp. eggs, hookworm larvae and eggs and pinworm eggs were recovered from the tables. The results of this pilot study highlight other potential means of transfer of zoonotic parasites and suggest that surfaces without obvious fecal material can be contaminated with zoonotic parasites.

1. Introduction

African green monkeys (AGM; *Chlorocebus aethiops sabaenus*) were introduced to a few Caribbean islands over 350 years ago \cite{1}. Today, free-roaming AGM are on St. Kitts with population estimates ranging from 14,000 to 50,000 with smaller populations on Nevis, Barbados, Tortola and St. Maarten \cite{1,2}. AGM, as with other non-human primates (NHP), are known to harbour zoonotic parasites \cite{3–5}. In work by Yao et al. \cite{6} over 90\% of feces from wild AGM examined on St. Kitts were *Trichuris* spp. positive and phylogenetic analysis indicated that the detected species was in the same clade of *T. trichiura* found in humans \cite{7}. Other parasites seen in the AGM on St. Kitts with zoonotic potential include hookworms (species not determined), *Strongyloides* spp., *Blas tocystis* spp. and *Giardia* sp. (unpublished data) all of which also occur in NHP in other regions \cite{5,8}. Human infections with these parasites occurs from consumption of infective stages; *Strongyloides* spp. infection also occurs via skin penetration of third stage larvae. Infections can be asymptomatic or result in a range of clinical signs including abdominal pain and diarrhea. Of these parasites, the most severe clinical signs can occur with *Strongyloides* spp. pulmonary infections in immuno-compromised people, which can lead to death.

The AGM are a tourist attraction on St. Kitts \cite{2}. Young AGM, typically wearing diapers, can be held for photographic opportunities. At some beach restaurants wild AGM are provisioned at certain times of the day, enabling tourists to have a close view of them. As the feeding time approaches, AGM of all ages gather in the vicinity and during feeding they run through eating areas including over the tables. If infective parasite eggs or larvae are on the hands or feet of the AGM, they could be transferred to the tables or passed to people holding them.

The purpose of this pilot study was to determine if parasitic stages could be found on the hands and feet of AGM and surfaces with which AGM are known to be in contact.

2. Materials and methods

All procedures involving AGM were in accordance with the ethical standards of Ross University School of Veterinary Medicine (RUSVM), performed under RUSVM Institutional Animal Care and Use Committee approved protocols and all applicable international, national, and institutional guidelines for the care and use of animals were followed. For privately owned AGM, owner consent was obtained prior to any procedures with the animal.

The hands and feet of eight AGM (four wild from troops invading agricultural crops and trapped to remove from the area and four tame) were examined for parasites using procedures modified from Jeandron et al. \cite{9}. Wild AGM were sedated while the tame AGM tolerated the...
procedure without sedation. For the first two wild AGM each hand/foot was placed one at a time in an open container of 120 ml water and 0.05% v/v nonionic detergent (Tween® 20, Sigma). The fingers/toes and up to the wrist/ankle of each hand/foot were gently rubbed using a gloved hand for 2–3 min while the foot/ hand was in the water. The hand/foot was then raised above the water/detergent and rinsed with water. For all AGM thereafter, each hand/foot was washed in a separate plastic bag containing 20–40 ml of water/detergent. The hand/foot was placed inside the bag and the bag closed around the wrist/leg. The bag was rubbed gently against the hand/foot for approximately 2 min after which the hand/foot was held over the bag and gently sprayed with water to remove any soap. The water/detergent mixture was centrifuged for 5 min at 500 G, the supernatant poured off and the pellet mixed with Sheather’s sugar flotation solution (1.27 specific gravity) and centrifuged with a coverslip on the tube for 5 min at 500 G. The coverslip was then examined at 100× magnification for parasitic organisms. Using the same centrifugation/flotation method as for the coverslip was then examined at 100× magnification for parasitic organisms. Using the same centrifugation/flotation method as for the hands/feet, 1 g of feces from the wild AGM, collected from the floor of the traps, also were analysed. Parasite identification was based on morphology [10,11].

Two sets of picnic tables were selected for examination. AGM were not fed in the area of the tables; however, an AGM troop resided in the area and the AGM were seen on the tables typically when people were scarce. Table set 1 (3 tables; approximately 1.4 m² each) was metal grid area and the AGM were seen on the tables typically when people were scarce. Table set 2 (2 tables; approximately 1.4 m² each) was painted wood and located in the open. Due to a decision to power wash the set 1 tables, the set 2 tables, located nearby were added. Prior to this study, tables were powerwashed at most three times a year. Set 1 was examined in November 2015 and April and June 2016. Set 2 was examined in April, May and June of 2016. The tables were wiped with damp sponges (water and 0.05% v/v Tween® 20); sponges were then rinsed in approximately 100 ml water/detergent with all water combined for analysis. Analysis was described for the hands/feet.

3. Results

All of the wild AGM and two of the tame AGM used for tourist interactions were found to have 1 to 10 parasitic stages (T. trichiura or hookworms) on their hands and feet (See Table 1). All of the fecal samples were positive for T. trichiura (27 to 937 eggs per gram) and Strongyloides spp. (1–10 eggs per gram). From the picnic tables potentially zoonotic parasites (2 to 85) were recovered on two of three occasions with set 1 and on all occasions with set 2 (See Table 1).

4. Discussion

Environmental contamination with zoonotic organisms is not a new concept with much research on soil in public places containing zoonotic parasites, particularly from dogs and cats as well as urban populations of wild animals (e.g., foxes and raccoons) [12–15]. In this pilot study we demonstrated that zoonotic parasites can be found in areas where animals walk, climb and rest, even when there is no apparent fecal material, with the results from the hands/feet of the AGM suggesting a form of transfer to these surfaces. While only two of the organisms seen were at the infective stage, the methods used were relatively crude and not designed to recover infective hookworm and Strongyloides larvae or protozoa which can be immediately infective.

Molecular characterization of the organisms from the picnic tables was not performed; therefore, it cannot be concluded that the organisms were from AGM. Feral cats, which are common on St. Kitts, could also have been a source and the pinworm could be of human origin. However, the zoonotic risk is still present since hookworms from cats (Anclylostoma tubaeforme) can cause cutaneous larval migrans. Therefore, retrieval of these organisms from the tables could have implications not only in areas with NHP but also with feral cats, free-roaming pet cats and other wildlife.

Risk of acquiring a zoonosis from environmental sources is related to frequency, type and length of contact as well as infection level of the surface [13,16]. Holding an AGM on St. Kitts for a photograph, a single contact, is unlikely to result in a tourist becoming infected with a parasite, given the low number of recovered organisms. However, the risk might be higher with children due to decreased hand hygiene and more hand-to-mouth activities [13,14]. Also, AGM handlers could be at risk with daily AGM contact. In regards to eating areas, the number of organisms recovered from the tables were low but risk of exposure is high since the surfaces are used for consuming food. Contact with the picnic tables during a single meal is unlikely to pose a high risk to tourists; however, restaurant employees, with daily exposure, could be at higher risk.

Raising awareness of sources of zoonotic parasites in this situation with AGM, and also in areas with wildlife and feral animals, must be done with caution and balanced with the true risk and consequences of human infection and the cost and effort of methods for decreasing contamination [17]. Education at restaurants regarding surface disinfection methods could be beneficial; however, it is important to note that most cleaning agents are not effective against infective parasitic stages. Hand hygiene education of AGM owners and tourists would likely be beneficial. Anthelmintic treatment of owned AGM could decrease risk of owner and tourist exposure. However, administration of anthelmintics to wild AGM is more complicated. While anthelmintics could be mixed with fruits offered to AGM, under dosing could occur resulting in low efficacy and a false sense of addressing the issue. Also, mass drug administration to AGM could destroy refugia which can lead to anthelmintic resistance [18]. Given the potential zoonoses of the parasites, particularly of the T. trichiura, anthelmintic resistance could result in a decreased efficacy of treatments for humans on St. Kitts.

While the intention of this pilot study was relatively simple - to assess if animals walking on surfaces could spread parasites - addressing the findings is complicated. Methods of decreasing these forms of environmental exposure require a multidisciplinary approach that

| Source | Number positive | Parasitic organisms recovered |
|--------|-----------------|------------------------------|
| **Chlorocebus aethiops sabaena** | | |
| Wild adult (4) | M | Hands and feet |
| M | Trichuris trichiura (4 eggs) | Feaces egg² |
| F | T. trichiura (10 eggs) | T. trichiura 27; |
| F | T. trichiura (3 eggs) | Strongyloides 10 |
| | | |
| | T. trichiura (2 eggs) | T. trichiura 299; |
| | | Strongyloides 160 |
| | | T. trichiura 80 |
| Pet juvenile (3) | 1 M | T. trichiura; 2 eggs |
| F | No parasitic organisms recovered |
| | | |
| Pet infant (1) | M | Hookworm²; 1 egg |

| **Picnic tables** |  |
|--------------------|---------------------------|
| Set 1 |  |
| Nov 2015 | Trichuris spp. (2 eggs; one larvated/one non-larvated eggs); Hookworm (16 eggs); Strongyloides spp. (5 eggs with larvae) |
| June 2016 | No zoonotic parasite eggs or larvae seen |
| Set 2 |  |
| April 2016 | Enterobius spp. (1 egg); Hookworm (7 eggs) |
| May 2016 | Enterobius spp. (82 eggs); Hookworm (3 eggs) |
| June 2016 | Enterobius spp. (46 eggs); Hookworm (5 eggs); Trichuris spp. (1 egg) |

² Eggs per gram of feces.

b Species not determined; hookworms identified in C. a. sabaena in the Caribbean include Anclylostoma duodenale and Necator americanus and in cats on St. Kitts A. tubaeforme occurs, all of which are zoonotic.

Table 1 Zoonotic parasitic organisms recovered from hands and feet of Chlorocebus aethiops sabaena and picnic tables.
considers animal health and welfare, population control and education.

Conflict of interest statement

The authors have no conflict of interest to declare.

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Author contributions

All authors contributed to the acquisition of the data and approved the final version of the manuscript submitted. C.G. and J.K.K. contributed to the concept and design of the study, analysis and interpretation of the data and drafting of the article. K.C., J.C. and A.B. critically reviewed the manuscript.

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