Wildlife Species as Potential Sources of Human Exposure to Parasitic Pathogens in Accra, Ghana

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Abstract

Handling and consumption of wildlife species pose risks of exposure to the infective stages of parasitic pathogens and disease transmission. This study assessed protozoan and helminth infections in some wildlife species commonly consumed as meat in Accra, Ghana. Using Zinc Sulphate centrifugal flotation and microscopic techniques, seven protozoan species (*Entamoeba* sp., *Giardia* sp. *Iodamoeba* sp., *Cryptosporidium* sp., *Balantidium* sp., *Endolimax* sp. and *Eimeria* sp.) and 11 genera of intestinal helminths including *Haemonchus, Trichostrongylus, Ascaris, Monieza, Schistosoma* and *Trichuris* were identified. Kruskal-Wallis tests showed significant difference in intensity of protozoan [$\chi^2(3) = 11.59$; $p = 0.009$] and helminth [$\chi^2(3) = 31.41$; $p < 0.0001$] infections among the four groups of wildlife species. For protozoans, the differences were observed between the mean cyst and oocyst (*Cryptosporidium* sp.) intensities of Maxwell's Duiker and Bushbuck ($U = 5$, $p = 0.006$) as well as Grasscutter and Bushbuck ($U = 3$, $p = 0.004$). However, the differences in mean helminth egg intensities were observed between Grasscutter and Maxwell's Duiker ($U = 6$, $p < 0.0001$), Grasscutter and Bushbuck ($U = 16$, $p < 0.0001$) as well as Grasscutter and Royal Antelope ($U = 4$, $p < 0.0001$). The results highlight the need for awareness of the risks of parasitic diseases as a prerequisite for their effective prevention.

Introduction

Bushmeat is an important nutritional resource for hundreds of millions of people especially in developing countries (Golden *et al.*, 2011). It provides various healthy nutritional requirements such as high protein (Tulley *et al.*, 2000; Wilkie *et al.*, 2011), low cholesterol and healthy fatty acid profiles (Tulley *et al.*, 2000; Hoffman, 2008) including essential omega-3 fatty acids (Okoye *et al.*, 2015).

Bushmeat is also a major source of livelihood for rural households living in extreme poverty (de Merode *et al.*, 2004) as well as poor-to-middle income households (Kümpel *et al.*, 2010). It also contributes significantly to the economies of West and Central African countries (Bowen-Jones *et al.*, 2003; Ntiamoa-Baidu, 1987) and serves several other needs including zootherapy (Alves and Alves, 2011).

Wild animals consumed as meat serve as reservoir hosts for many parasites such as protozoans (Opara, 2012) and helminths (Robinson and Dalton, 2009) that can be transmitted to humans. Majority of global infectious diseases are caused by zoonotic pathogens with about 71% percent of them originating from wildlife (Cutler *et al.*, 2010; WHO, 2012). The frequent dependence of humans on bushmeat as a vital resource poses increasing risks of zoonotic pathogen and disease transmission (Friant *et al.*, 2015) through its handling and consumption (FAO, 2015; Karesh *et al.*, 2009; Wolfe *et al.*, 2005; Daszak *et al.*, 2007; Kuukyi *et al.*, 2014) particularly by people with occupational exposures such as hunters, slaughters, traders and the final consumer (Futagbi *et al.*, 2010).

In this study, we report on protozoan and helminth infections in some wildlife species commonly consumed as meat in Ghana and highlight the importance of awareness of zoonotic disease risks as a prerequisite for

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effective disease control.

Materials and methods

Study design and area

A cross-sectional study involving four commonly-consumed wildlife species was carried out between January and April, 2016 to assess their endoparasitic infection status. The sampling site, Kantamanto Market in the Central Business District of Accra (*5.548° N, 0.2123° W*), is one of the major markets in Ghana where fresh wildlife species consumed as meat are readily available (Ntiamoa-Baidu, 1997). The wildlife species were *Thryonomys swinderianus* (Grasscutter, n = 41), *Philantomba maxwellii* (Maxwell's Duiker, n = 56), *Tragelaphus scriptus* (Bushbuck, n = 35) and *Neotragus pygmaeus* (Royal Antelope, n = 68). These were brought to the market from three major towns in Ghana notably Mankessim in the Mfantsiman Municipal District (*5.2728° N, 1.0155° W*), Swedru in the Agona District (*5.5298° N, 0.7058° W*) and Afram Plains in the Kwahu North District (*6.9209° N, 0. 1495° W*).

Sample collection

Fresh faecal samples from individual animals of each species were collected prior to singeing, kept in labelled zip lock transparent rubber bags and immediately placed in an ice chest containing ice cubes and transported to the Parasitology Laboratory, Department of Animal Biology and Conservation Science (DABCS), for processing.

Sample processing, data collection and analysis

The Zinc Sulphate centrifugal flotation method (Faust and Melency, 1924) was used to process the faecal samples. The prepared slides were observed at 10 X and 40 X under a Leica DME Compound Microscope. Parasitic identification was based on standard morphological characteristics of protozoan parasite cysts, oocysts (for *Cryptosporidium* sp.) and helminth eggs (Chiodini *et al.*, 2001; WHO, 1994). The prevalence of protozoan and helminth infections for each wildlife species was calculated. Protozoan cysts, oocysts and helminth eggs for each infected sample were counted and multiplied by 50 as required by the protocol. The intensities of protozoan parasite cysts, oocysts and helminth eggs were calculated as their respective mean numbers per gram of faecal matter.

Using the Statistical Package for Social Sciences (SPSS) [version 20] to check for normality, the data were observed to be skewed. Kruskal-Wallis test, using correction of ties, was therefore performed to assess variability of mean protozoan cyst, oocyst and helminth egg intensities in the wildlife species. A post hoc test was performed to identify the wildlife species accounting for the observed difference in mean protozoan cyst, oocyst and helminth egg intensities.

Results

Protozoan species identified as present in the wildlife species were *Cryptosporidium* sp., *Giardia* sp., *Iodamoeba* sp., *Endolimax* sp., *Entamoeba* sp., *Balantidium* sp. and *Eimeria* sp. *Thryonomys swinderianus* was identified to have the highest overall prevalence (56.10%) of protozoan infections compared with those of *Philantomba maxwellii* (28.57%), *Tragelaphus scriptus* (17.14%) and *Neotragus* *pygmaeus* (10.29%). It also dominated in prevalence of individual protozoan infections (Figure 1).

In all, 11 genera of intestinal helminths (Table 1) belonging to two phyla, Platyhelminthes and Nematoda, were identified as present in the wildlife species studied. The nematode, *Trichostrongylus* sp., was the most common helminth (Figure 2) in all the wildlife species with *T. swinderianus* dominating in prevalence (Figure 2) of individual helminth infections as well.

A Kruskal-Wallis test was performed to determine whether mean cyst and oocyst (*Cryptosporidium* sp.) intensities of protozoans varied in the wildlife species. The results show significant differences in intensity of protozoan infections, $\chi^2(3) = 11.59$; $p = 0.009$. These differences were observed between the mean intensities of Maxwell's Duiker and Bushbuck ($U = 5$, $p = 0.006$) and Grasscutter and Bushbuck ($U = 3$, $p = 0.004$) rather than Grasscutter versus Royal Antelope ($U = 10.5$, $p = 0.067$) and Maxwell's Duiker versus Royal

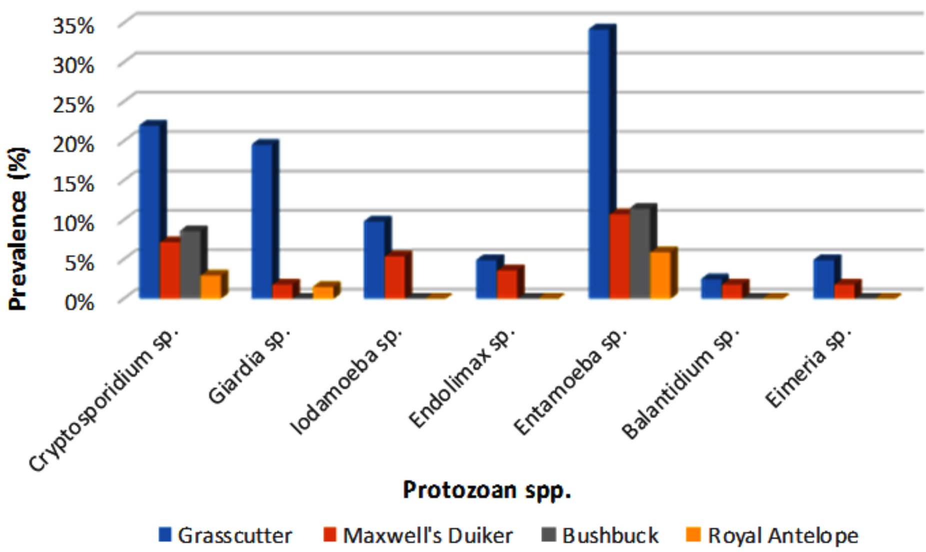

Figure 1: Prevalence of protozoans in wildlife species

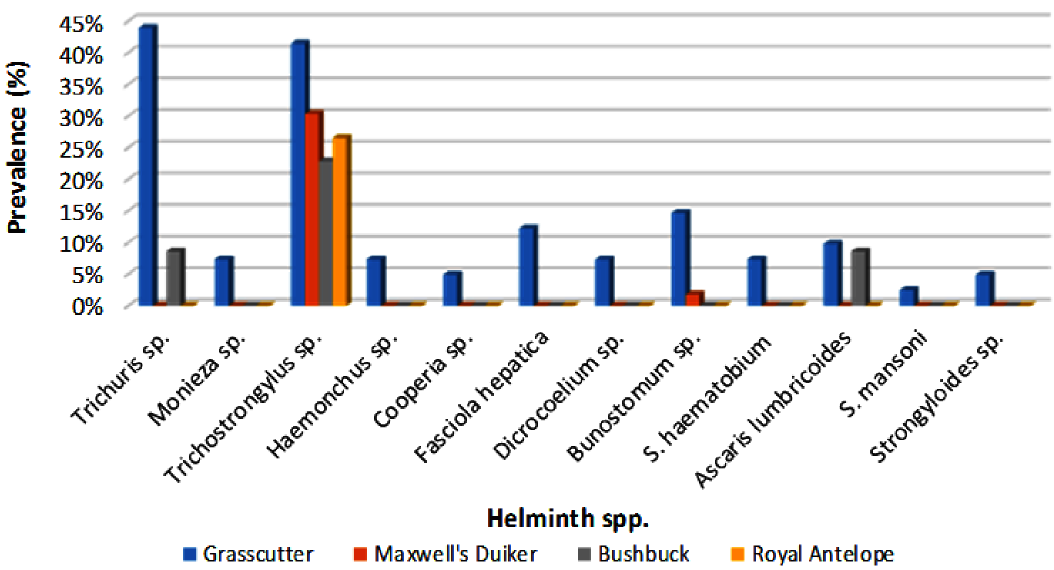

Figure 2: Prevalence of helminths in wildlife species

Antelope ($U = 14.5$, $p = 0.178$).

Mean egg intensities of the different species of helminths (Table 1) differed significantly among the four groups of wildlife species, $\chi^2(3) = 31.41$; $p < 0.0001$. This difference was observed between the mean intensities of Grasscutter and Maxwell's Duiker ($U = 6$, $p < 0.0001$), Grasscutter and Bushbuck ($U = 16$, $p < 0.0001$) as well as Grasscutter and Royal Antelope ($U = 4$, $p < 0.0001$) rather than Maxwell's Duiker and Bushbuck ($U = 70.5$, $p = 0.330$), Maxwell's Duiker and Royal Antelope ($U = 78$, $p = 0.548$) and Bushbuck and Royal Antelope ($U = 65$, $p < 0.146$).

Discussion

Bushmeat is an important nutritional resource, which serves as a major source of livelihood for the poor. In Ghana, Grasscutter, Maxwell's Duiker, Bushbuck (Yeebo, 2016; Kuukyi *et al.*, 2014; Binnquist *et al.*, 2004; Owusu *et al.*, 2004) and Royal Antelope (Yeebo, 2016) are some of the most patronised wildlife species consumed as meat. This study assessed protozoan and helminth infections in such wildlife species.

Protozoan and helminth species from seven and 11 genera respectively were identified in

TABLE 1
Number of protozoan and helminth-infected wildlife and their mean cysts, oocyst (for *Cryptosporidium* sp.) and egg intensities (*mean intensities in brackets*)

Type of parasite	*Thryonomys swinderianus*	*Philantomba maxwellii*	*Tragelaphus scriptus*	*Neotragus pygmaeus*
Protozoa				
Cryptosporidium sp.	9 (116.67)	4 (75)	3 (50)	2 (50)
Giardia sp.	8 (75)	1 (50)	-	1 (250)
Entamoeba sp.	14 (353.57)	6 (66.67)	4 (50)	4 (62.50)
Iodamoeba sp.	4 (50)	3 (50)	-	-
Endolimax sp.	2 (50)	2 (50)	-	-
Eimeria sp.	2 (100)	1 (50)	-	-
Balantidium sp.	1 (50)	1 (50)	-	-
Helminth				
Trichuris sp.	18 (227.80)	-	3 (100)	-
Monieza sp.	3 (116.70)	-		-
Ascaris lumbricoides	2 (75)	-	1 (50)	-
Infertile A. lumbricoides	2 (50)	-	2 (50)	-
Trichostrongylus sp.	17 (238.20)	17(64.65)	8 (68.75)	18 (52.30)
Haemoncus sp.	3 (50)	-	-	-
Cooperia sp.	2 (75)	-	-	-
Fasciola hepatica	5 (120)	-	-	-
Dicrocoelium sp.	3 (66.70)	-	-	-
Bunostomum sp.	6 (212.50)	1(50)	-	-
Schistosoma mansoni	1 (50)	-	-	-
S. haematobium	3 (50)	-	-	-
Strongyloides sp.	2 (1250)	-	-	-

(-) No evidence of parasitic infection

the wildlife species at varying frequencies and intensities. Among the four groups of wildlife species, significant differences in intensity of protozoan and helminth infections were observed. In particular, Grasscutter dominated in prevalence (Figures 1 and 2) and intensity (Table 1) of both protozoan and helminth infections in conformity with previous investigations in several parts of Africa, including Ghana (Mpoame, 1995; Adu *et al.*, 1999; Yeboah and Simpson, 2004; Ajayi *et al.*, 2007; Opara and Fagbemi, 2008) compared with the other wildlife species. These results may be attributed to their feeding behaviour and some habitat characteristics.

Grasscutter is a monogastric herbivore (Hemmer, 1990) that feeds on a large variety of grasses, as well as leaves of other food crops such as maize, sugar cane, oil palm and pineapple (Opara, 2012) and often practices coprophagy (Hemmer, 1990; Opara, 2012). It has often been encountered in the vicinity of water courses (Kingdon, 1974) and is known to inhabit places with dense grass, especially wet and damp areas with grasses (Asibey, 1974). These places are also the preferred areas for protozoans and helminths, which thrive better under moist conditions (Akinbo *et al.*, 2011) and whose availability is a key determinant of the magnitude of pasture infectivity (Rossanigo and Gruner, 1995; O'Connor *et al.*, 2007). These risk factors may have predisposed Grasscutter to parasitic infections. However, the other species are browsers, with Royal Antelope having a preference for fresh foliage and shoots, and occasionally feeding on fruits and fungi. Maxwell's Duiker feeds more on fruits, dicotyle leaves and less grass (Hofmann and Roth, 2003) with Bushbuck feeding more on shrubs, perennial woody plants, herb species, forbs and algae (Smits, 1986). These feeding materials are less likely to be contaminated with infective stages of parasites as the results indicated.

Several species of parasites from different genera of zoonotic importance including *Giardia*, *Cryptosporidium, Haemonchus, Trichostrongylus, Monieza, Strongyloides* and *Fasciola* were identified. The most important cycles of transmission for maintaining *Giardia* and *Cryptosporidium*, the most common enteric parasites of domestic animals (Fayer, 2004; Thompson, 2004; Thompson and Monis, 2004), involve humans, livestock, companion animals and wildlife (Thompson *et al.*, 2008). *Cryptosporidium parvum* genotype 1 is exclusive to humans and a single nonhuman primate, and genotype 2 has a broader host range, including livestock, wild animals and humans (Spano *et al.*, 1998; McLauchlin *et al.*, 1999; Morgan *et al.*, 1999). *C. parvum* genotypes 1 and 2 now known as *C. hominis* and *C. parvum* respectively (Morgan-Ryan *et al.*, 2002) have been identified to be involved in several outbreaks related to drinking water (McLaughlin *et al.*, 2000; Anim-Baidoo *et al.*, 2015), swimming pool, family, children's nursery (McLaughlin *et al.*, 2000), paediatric hospital (Anim-Baidoo *et al.*, 2015) and vegetables (Petersen *et al.*, 2014).

Some species of *Haemonchus* (Pestechian *et al.*, 2014), *Trichostrongylus* (Bradbury, 2006; Sato *et al.*, 2011), *Ascaris* (Nejsum *et al.*, 2005), *Monieza* (el-Shazly *et al.*, 2004), *Strongyloides* (Labes *et al.*, 2011; Pourrut *et al.*, 2011) and *Fasciola* (Curtale *et al.*, 2007) have been associated with human infections. Human handling and consumption of wildlife, coupled with the complex interactions with livestock, companion animals and the environment, pose increasing risk of exposure to the infective stages of zoonotic pathogens and disease transmission. Good wildlife handling practices particularly by hunters, slaughters, bushmeat traders and consumers as well as sustained health education on the risk of zoonotic disease transmission are fundamental to the reduction of health and economic losses.

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