

# Parasitic infections in African pangolin (*Manis temminckii*) from Edo State, southern Nigeria

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## Abstract

Pangolins are anteaters and they play important roles in regulating ant and termite levels in the ecosystem. They have been reported as useful tools in traditional medicine as well as host to parasites that can cause diseases in humans. Market-derived pangolins (*Manis temminckii*) from Ekenwan Village, a rain forest location in Ovia North East Government Area of Edo State, Nigeria, were used in the study. All 12 pangolins examined were infected with parasites giving a 100% prevalence of infection. The parasites isolated from pangolins included, a cestode (*Oochoristica sp.*), a nematode (Strongylidae), a pentastomid (*Armillifer sp.*) and an ectoparasite (*Amblyomma sp.*). *Oochoristica sp.* (100%) and *Amblyomma sp.* (75%) were the most prevalent parasites. Both male and female pangolins recorded equal prevalence (100%) of infection, however, mean intensity of parasites was higher in males ( $13.33 \pm 4.34$ ) than females ( $8.00 \pm 3.52$ ). Both adult and juvenile pangolins showed equal prevalence (100%) of infection, but mean intensity of parasites was higher in adults ( $23.89 \pm 3.61$ ) than in juveniles ( $11.00 \pm 5.75$ ). Pangolins collected during the dry and rainy seasons recorded equal prevalence (100%) of infection, however, mean intensity of parasites was higher during the dry season ( $24.62 \pm 3.67$ ) than the rainy season ( $14.75 \pm 3.42$ ). The findings in this study are new records for pangolin *Manis temminckii* in Nigeria.

**Keywords:** pangolins, *Manis temminckii*, parasites, Edo State, Nigeria.

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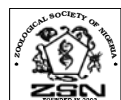
## Introduction

Pangolins play important roles in the ecosystem (Craig, 2012). As anteaters, they help regulate population of ants and termites in the ecosystem. Conversely, pangolins act as sources of food to predators, like lions, tigers, leopards and hyenas. Pangolins have been reported as useful tools in traditional medicine around the world (Adeola, 1992; El-kamal 2000; Kakati and Doulo, 2002; Soewu, 2008; Durojaye and Ibukun, 2009). These animals have been reported as hosts to ectoparasites (ticks such as *Amblyomma javanese*) and endoparasites (tapeworms and nematodes) that can cause diseases in humans (David, 2001, and Mohammed *et al*, 2012)). Aside predation and parasitism, pangolins are also threatened by poaching for meat and use of parts of the animals for traditional medicine. Hence the need to pay more attention to the study of these animals, as the only documented studies in Nigeria are those connected with their uses in traditional medicine (Adeola, 1992; Soewu, 2008; Durojaye and Ibukun, 2009).

## Materials and methods

Market-derived pangolins (*Manis temminckii*) from Ekenwan Village were used for the study. Ekenwan Village (Latitude  $5^{\circ}40'N$  and  $5^{\circ}05'E$ ) is a rain forest location in Ovia North East Local Government Area of

Edo State, southern Nigeria. The specimens were transported to the laboratory, euthanized and examined for parasites. The ectoparasites were isolated from the specimens. Thereafter, the specimens were dissected and various parts of the gastro-intestinal tract as well as the lungs, gall bladder and urinary bladder were teased out and examined in normal saline (0.72% NaCl) under a dissecting microscope. The body cavity was also examined visually for parasites. Ticks were preserved in 70% ethanol while pentastomids were preserved in alcohol-saline. Isolated cestodes were flattened and fixed with 5% formol-saline under cover slip pressure on a microscope slide for 30 minutes, after which they were transferred to specimen bottles containing 5% formol-saline. The nematodes recovered were fixed in hot 70% alcohol and preserved in fresh 70% alcohol. For the purpose of examining pentastomid hooks for identification, the worms were decapitated and flattened on a microscope slide under a cover slip, by applying gentle pressure after adding a drop of Hoyer's medium. Nematodes were prepared for identification by mounting in a clearing agent (Lactophenol). While cestodes were washed in several changes of tap water to remove formalin preservative. Thereafter, the worms were stained overnight with a dilute solution of acetocarmine, dehydrated in ethanol series, cleared in xylene and permanent mount made in Canada balsam.



Photographs were taken using biological microscope (model BMX2CBA) to which was attached a digital camera (3.34 mega pixels). Parasites were identified with the aid of appropriate keys (Bray *et al*, 1994 and Yamaguti, 1971). Prevalence rate of parasites was calculated as a percentage of the number of host infected with a specific parasite divided by the total number of host examined. The mean intensity of infection was based on the average number of parasites per infected host.

## Result

A high prevalence (100%) of infection was recorded for pangolin in this study, three types of endoparasites, comprising pentastomids, a nematode and a cestode as well as an ectoparasite (a tick) were recovered from the pangolins. The pentastomids were identified as *Armilifer sp.*, cestode as *Oochoristica sp.*, and tick as *Amblyomma sp.* the nematode has so far been identified as a member of family strongylidae. *Oochoristica sp.* was the most prevalent parasite, and it occurred in all 12 pangolins. This parasite also had the highest mean intensity of parasite (12.67±2.92). Parasites isolated, prevalence of infection, mean intensity of parasites and location of parasites in the host are shown in Table 1. The prevalence

of *Amblyomma sp.* was 75% while the mean intensity was 7.2±1.51. For the strongylid nematode, the prevalence was 50% and mean intensity 6.00±5.75. *Armilifer sp.* was isolated from only one pangolin, therefore had the lowest prevalence (8.3%) and mean intensity of parasite (3.00±0.00). Parasites prevalence and mean intensity of parasites in relation to pangolin's life stages is shown in Table 2. All nine adults and three juveniles pangolins examined were infected, however, mean intensity of parasites was higher in adults (23.89±3.61) than juveniles (13.66±2.91). Parasite prevalence and mean intensity of infection in relation to pangolin's sex is shown in Table 3. All seven male and five female pangolins examined were infected, but with different mean intensity of infection. Male pangolins mean intensity was 13.33±4.34 while for females, it was 8.00±3.52.

Prevalence of infection and mean intensity ( $\bar{x} \pm SD$ ) of parasites in relation to season is represented in Table 4. Pangolins collected both in dry season (8) and rainy season (4) were all infected but mean intensity of parasite was higher during dry season (24.62±3.67) than rainy season (14.75±3.42).

**Table 1:** Prevalence and mean intensity of parasites of the African pangolin.

Parasite	No. of host examined	No. of host infected	Total No. of parasites	Prevalence (%)	Mean intensity ( $\bar{x} \pm SD$ )	Location of parasites
Pentastomida <i>Armilifer sp.</i>	12	1	3	8.3	3.00±0.00	Liver
Nematoda Strongylid nematode	12	6	36	50	6.00±5.75	Stomach/small intestine
Cestoda <i>Oochoristica sp.</i>	12	12	152	100	12.67±2.92	Intestine
Acarina <i>Amblyomma sp.</i>	12	9	65	75	7.20±1.51	Under scale

**Table 2:** Parasites prevalence and mean intensity in relation to pangolin's life stages.

Age	No. of host examined	No. of host infected	Prevalence %	Total No. of parasites	Mean intensity ( $\bar{x} \times SD$ )
Adult	9	9	100	215	23.89±3.61
Juvenile	3	3	100	41	13.66±2.91
<b>Total</b>	<b>12</b>	<b>12</b>	<b>100</b>	<b>256</b>	<b>21.33±6.52</b>

**Table 3:** Parasites prevalence and mean intensity in relation to pangolin's sex.

Sex	No. of host examined	No. of host infected	Prevalence %	Total No of parasites	Mean intensity ( $\bar{x} \times SD$ )
Male	7	7	100	160	13.33±4.34
Females	5	5	100	96	8.00±3.52
<b>Total</b>	<b>12</b>	<b>12</b>	<b>100</b>	<b>256</b>	<b>21.33±7.86</b>

**Table 4:** Parasites prevalence and mean intensity in relation to seasons.

Season	No. of host examined	No. of host infected	Prevalence %	Total No. of parasites	Mean intensity ( $\bar{x} \pm SD$ )
Dry	8	8	100	197	24.62±3.67
Rainy	4	4	100	59	14.75±3.42
<b>Total</b>	<b>12</b>	<b>12</b>	<b>100</b>	<b>256</b>	<b>21.33±3.32</b>

## Discussion

Information on the parasites of pangolins in the literature is scanty. Other closely related mammals belonging to the same Manidae family that have been investigated include armadillos and sloths.

All pangolins, *Manis temminckii*, examined in this study were infected with one or more species of ectoparasite (ticks) and endoparasites (cestode, nematode and pentastomids). Similarly, all armadillos, 2 *Euphractus sexinctus* and 2 *Dasybus novemcinctus*, examined in Paraguay by Osamu *et al* (1995) were infected with one or more species of nematodes. However, 21.5% prevalence of infection with gastro-intestinal parasites (coccidia, cestode and spiruridea) was reported in sloths, *Bradypus variegatus* and *Choloepus hoffmanni* in Costa Rica (Karem, 2009). Luwis (2008) however, indicated that due to insectivorous diet of pangolins, their helminth fauna largely comprises hymenolepid and dilepidid cestodes, brachylaemid and durocoelid digeneans.

The prevalence of *Amblyomma sp.*, the only ectoparasite recorded in the present study was 75% and the mean intensity of the parasite was 7.2±1.5. Furthermore, both adults (66.7%) and juvenile (33.3%) pangolins were infected. Yang (2001) recorded 68.63% prevalence of *Amblyomma javanense* infection in Malayan pangolin *Manis javanica* and noted that juveniles were not infected. Mohammed *et al* (2012) also isolated *A. javanense* from Malayan pangolin, but recorded 100% prevalence in juveniles compared to 63.6% in adult. Ectoparasites including *Sarcoptes scabies* (mite) and *Amblyomma varium* (tick) have been recovered from sloths which were in captivity in Costa Rica (Karem, 2009, Keli, 2012).

The endoparasites isolated from pangolin in this study differ from those isolated previously by other researchers. Whereas, nematode recorded in this study was identified as strongylid nematode, a nematode recorded previously from pangolin, *Manis pentadactyla*, by other researchers (Travassos, 1957; Meyer, 1960; Cameron and Meyers, 1960; Chin, 1961) was named *Manisstrongylus meyeri*.

Osamu *et al* (1995) however, recovered 12 species of nematodes comprising 9 genera including aspidoderid nematodes from 4 armadillos captured in Paraguay, while Papadopoulos *et al* (1996) isolated 3 nematodes from 4 armadillos obtained from rural areas in Greece. Jimenz (2003) also isolated 245 aspidoderid nematodes, namely, *Aspidodera binansata* and *Lauroia bolivari* from Bolivian armadillos.

Other nematodes, apart from aspidoderidae, recorded also in armadillos (*Euphractus sexinctus*) from Brazil include, *Ancylostoma caninum*, *Trichoelax tuberculata*, and *Hadrostrongylus ransomi* (Hoppe, 2009). Satya (2009) described a new nematode species *Leiperinema leiperi* (strongyloididae) from pangolin (*Manis pentadactyla*) in India. A new species of nematode (family subuluridae) named *Cyclobulura* was also isolated by Graciela *et. al.* (2010) from armadillos (*Zaedyus pichiy* and *Chaetophractus vellerosus*) from Argentina.

Previous researchers (Papadopoulos *et al*, 1996; Karem, 2009; Keli, 2012) were not specific about the identity of cestodes isolated from armadillos and sloths, however, the cestode recorded in the present study from pangolin was identified as *Oochoristica sp.*

Pentastomids such as *Porocephalus crotali*, *Armillifer armillatus*, *Linguatula serrata* and *L. nuttalli* have been recorded in different mammals by various researchers (James, 1964; Young, 1975; Brookis, 2013). However, no previous report was recorded for armadillos, pangolins and sloths, whereas, 3 pentastomids identified as *Armillifer sp.* were isolated from 1 pangolin in the present study. Brookis (2013) reported that mammals serve as intermediate hosts (nymphs in form of encysted parasites were found in the gastro-intestinal tract of a dog) while snakes are the definitive hosts.

Another parasite that has been reported, though not commonly, in armadillos, *Tolyteutes matacus* is the acanthocephalan, *Oligacanthorhynchus carinni* (Lesley, 2007). It is note worthy that acanthocephalans were not recorded in this study.

The observation that male and female pangolins showed equal prevalence of infection is contrary to that observed by other researchers. Mohammed *et al* (2012) observed higher prevalence (88.9%) in males compared to females (42.9%). Zuk (2008) reported that sexually mature male vertebrates are often more susceptible to infection and carry higher parasite burdens in the field. These differences have been attributed to ecological and physiological differences including differential exposure to pathogens due to sex specific behaviours or morphology and the association between testosterone and the immune system.

Previously, Mohammed *et al* (2012) recorded a 100% prevalence of infection in juveniles compared to 63.6% in adult pangolins, whereas in the present study both adult and juveniles showed equal prevalence (100%) of infection. Payne *et al* (1998) earlier proffered an explanation in support of Mohammed *et al* (2012) that

juveniles tend to be more exposed to parasites in the environment since they are involved in many activities such as learning, exploring, playing and so on. Furthermore in nature juvenile animals commonly get infected by parasites through contamination from parents (Payne *et al.*, 1998).

Although pangolins collected in the present study during the rainy and dry season showed equal prevalence of infection. Previous studies (Fister, 1993) reported rainy season (June to September) represent a period of high parasites burden in mammals.

The fact that all pangolins examined presently were infected with parasites is an indication that parasitic infections in pangolins is common in the study-area. It is noteworthy that single species infection with parasites in the current study was rare (83.3% were multiple infections) and concerned only *Oochoristica sp.*, which was not only the most prevalent parasite but also had the highest mean intensity of infection.

Keli (2012), opined that these parasitic infections could be of serious problems to the infected animals, in particular the ectoparasites which have been known to be associated with nutritional deficiencies and systemic diseases especially in animals in captivity. Furthermore, David (2001) reported that anteaters harbour parasites that can cause diseases in humans.

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