

Preliminary survey on endoparasitism in *Ophiophagus hannah* (Reptilia: Serpentes: Elapidae) in Mizoram, India

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There is no record on the prevalence of parasites of snakes in Mizoram. This study was undertaken in an attempt to identify common gastrointestinal parasites in the king cobra *Ophiophagus hannah*. In this preliminary survey, necropsy was performed on four individuals collected from different localities within Mizoram during January 2015 to June 2016. Of the 4 snakes examined, 1 was found infected with cestode, nematode and acanthocephalan. The *Capillaria* sp. and *Kalicephalus* sp. collected from *O. hannah* in the present study was morphologically identified but molecular identification could not be done due to insufficient specimen. Acanthocephalan cystacanths were collected from the intestinal lining and were morphologically identified as belonging to the family Gigantothorhynchidae. Metacestode of *Taenia* sp. was collected from the body cavity and was morphologically identified while molecular characterization could not be done as only a single specimen was collected. Accurate identification is very important in understanding the geographical distribution and biodiversity of parasites. However, the identification of parasites to the species level based solely on morphological analysis might be risky.

Keywords: *Ophiophagus hannah*, cestode, nematode, parasite, Mizoram.

INTRODUCTION

The class Reptilia is composed of four extant orders, namely Chelonia (tortoises and turtles), Crocodilia (alligators, caimans, crocodiles, gharial), Rhynchocephalia (tuataras), and Squamata (lizards, worm lizards, and snakes). The first known reptiles appeared during the Paleozoic era, about 300 million years ago (Jacobson, 2007). During the Mesozoic, often called the Age of Reptiles, there were 17 orders. Thus, reptiles are far less diverse now. The exact phylogenetic relationship among chelonians, crocodilians, and squamata is not resolved. Particularly, the relationship between chelonians and the reptile clade Diapsida (crocodilians, lizards, and snakes) remains controversial (Jacobson, 2007). For example, molecular analyses indicate a relationship between chelonians and crocodilians (Zardoya and Meyer, 1998; Hedges and Poling, 1999), while osteological data indicate turtles within the diapsids as a sister group of the Sauopterygia, a group of Mesozoic reptiles (Rieppel and Reisz, 1999).

Ophidia (Serpentes) is the most recent group (order) of the present-day reptiles. The snake body is noticeably supple, capable of twisting and other movements that are impossible for other vertebrates. The structure of the backbone restricts any space for vertical movements, so that it is impossible for snakes to perform vertical undulatory movements. Thus, snakes move sideways (Daniel, 2002). Snakes are highly diverse and can be classified into 14 to 17 families. For instance, boas and pythons are sometimes placed in the family Boidae, while some authors separate them to Boidae and Pythonidae respectively (Jacobson, 2007). 3567 species of snakes have been recorded (Uetz, 2016), with 297 species India (Anon., 2014). In Mizoram 52 species of snakes have been reported by Lalremsanga and Lalronunga (2017). Lalbiakzuala and Lalremsanga (2017) made the first report of *Lycodon fasciatus* (banded wolf snake) from the state.

There is little information on parasitic infection in wild reptiles, particularly snakes. It is important to understand the degree and types of infection for wild life con-

servation. Some helminths can be directly dangerous and even lethal, while some can be mildly pathogenic (Noryoska and Sokót, 2015). This study shows helminth infection in the king cobra, which is perhaps the first study in Mizoram.

MATERIALS AND METHODS

For cestodes, freshly recovered parasites were stretched in warm water and gently flattened between a slide and a cover glass and was fixed in 70% ethyl alcohol. The specimen was stained with borax carmine, and dehydrated through ascending grades of ethyl alcohol, then cleared in methyl benzoate and mounted with DPX.

For nematodes, the worms were stretched in warm 70% alcohol. For permanent mounting, the worms were cleared in ascending grades of glycerine and finally double mounted using Kaiser's glycerine jelly (50 mL water + 80 gm gelatin + 50 mL glycerol + 0.1 gm phenol); for temporary mounting, the specimen were immersed in lactophenol overnight and temporarily mount with the same solution on the glass slide.

For Acanthocephala, the parasites were allowed to relax in hot saline and narcotized by adding a few drops of 70% alcohol and further processed following usual procedure of flattening and permanent mounting.

Generic identification of the parasites was done following standard reference works (Yamaguti, 1959a, 1959b, 1961, 1971; CIH Keys to Nematodes 1–10; Khalil et al., 1994).

RESULTS

During the study period, i.e. 2016–2017, *O. hannah* caught from the wild were immediately kept in a terrarium. Road killed specimen were also collected and kept immediately in a cold storage. A total of 4 individuals were examined and found positive with helminth parasitic infection viz. *Taenia* sp., *Capillaria* sp., *Ophidascaris* sp. and Acanthocephala.

DISCUSSION

Endoparasites of vertebrates are representatives of two phyla – the acelomate Platyhelminthes and the pseudocoelomate Nematoda. Parasitic infections are often chronic and in optimal habitat and maintenance conditions do not give clear clinical symptoms. Stress of transport, inadequate microclimate in the terraria, concentration of animals or improper nutrition, can suppress the immune system and lead to clinical form of parasitic diseases. But not all parasites are harmful to the reptile host; those that just pass through the intestinal tract are usually not (Schneller and Pantchev, 2008).

Plasmodium infections are suggested to cause changes in behavior and physiology of certain popula-

tions of lizards (Schall, 1996); the coccidian *Caryospora* was responsible for mortality in captive (Leibovitz et al., 1978) and wild (Gordon et al., 1993) marine turtles, and spirorchiid trematodes and their eggs caused severe lesions in multiple visceral sites in marine turtles (Gordon et al., 1998). A range of amoebiasis, cryptosporidiosis, and various helminth infections are noted (Jacobson, 2007).

Species of *Bothridium* and *Bothriocephalus* are mainly parasitic to boid snakes. Heavy infections have been reported, but the extent of infection is not investigated. A pseudophyllidean cestode, *Spirometra* is one of the most widely distributed parasites in snakes, which are in most cases intermediate or paratenic hosts. These cestodes release eggs from the definitive host that grow into larvae (coracidia). The larvae are ingested by a copepod, and develop into procercooids. Second intermediate host (such as amphibian, reptile, or mammal) ingest the procercooids that develop into plerocercoids, which are known as *spargana*. These are distributed throughout the body. Spargana may result in soft swellings of the body surfaces. Inside the tissues, they can cause edema and hemorrhage. Many cestodes of the families Ochetosomatidae (*Dasymetra*, *Lechriorchis*, *Zeugorchis*, *Ochetosoma*) and Plagiorchiidae (*Stomatrema*, *Styphlodora*) are commonly present in the oral cavity, pulmonary system and upper esophagus of snakes. They are collectively called the renifiers (Jacobson, 2007). Many species of *Hexametra* are reported from lizards and snakes in Africa and Southeast Asia (Sprent, 1978).

Roundworm infections are prevalent in wild snakes, and pathologic lesions are usually associated. Third-stage larvae of *Polydelphis quadrangularis* is known to cause gastric ulceration in Neotropical rattlesnake *Crotalus durissus terrificus*, particularly if they are less than 173 days of age (Araujo, 1971). Pythons are commonly infected with different species of nematodes, where adults lodged in the gastric mucosa and larvae migrate through visceral tissues creating mechanical damage. The wandering larvae are responsible severe inflammatory responses. A Papuan python (*Apodora papuana* [formerly *Liasis papuanus*]) held in captivity for 4 years and died after 3 weeks of anorexia was found with adhesions between the aorta and esophagus (Hamir, 1986). The aortic wall had many fibrous nodules, the endothelial surface was roughened, and there were numerous aneurysms throughout the aorta and the peritoneum of the coelomic cavity. One nematode was identified as *Ophidascaris papuanus*; of which the adults were found in the intestine. A bronchopneumonia and a large area of necrosis of one lung was also noted. Other ascarids observed by Sprent (1963; 1970a, 1970b) in pythons include *Ophidascaris moreliae* from the carpet python (*Morelia spilota variegata*), *Polydelphis anoura* from carpet pythons, scrub pythons (*Morelia amethistina*), and the woma python (*Aspidites ramsayi*), and *Amplicaecum robertsi* in the carpet python, where encapsulated larvae

Figure 1: Cestode recovered from intestinal lining of *O. hannah*.



Figure 2: Cystacanth of Acanthocephala recovered from intestinal lining of *O. hannah*.



Figure 3: Cystacanth of Acanthocephala recovered from intestinal lining of *O. hannah*.





Figure 4: *Ophiophagus hannah* (host species of *Capillaria* sp., *Ophidiascaris* sp., *Metacestode of Taenia* sp., and *Acanthocephala*.



Figure 5: Head portion of female *Capillaria* sp.



Figure 6: Mid-portion of gravid female *Capillaria* sp.



Figure 7: Female *Capillaria* sp. with magnified eggs with polar plug in uterus (single row).



Figure 8: Female *Ophidascaris* sp.



Figure 9: Male *Ophidascaris* sp.

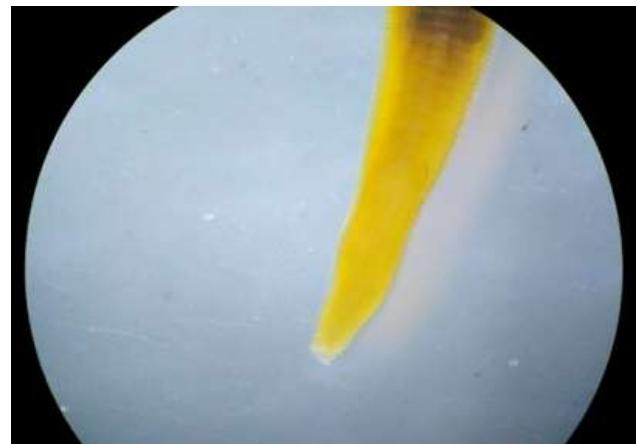


Figure 10: Anterior end of male *Ophidascaris* sp.

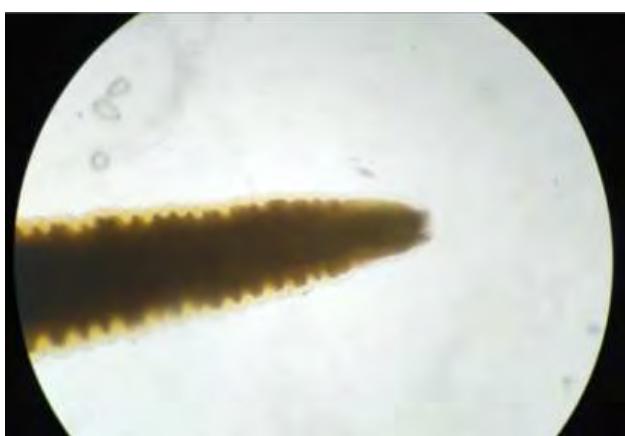


Figure 11: Anterior end of female *Ophidascaris* sp.



Figure 12: Posterior end of male *Ophidascaris* sp.

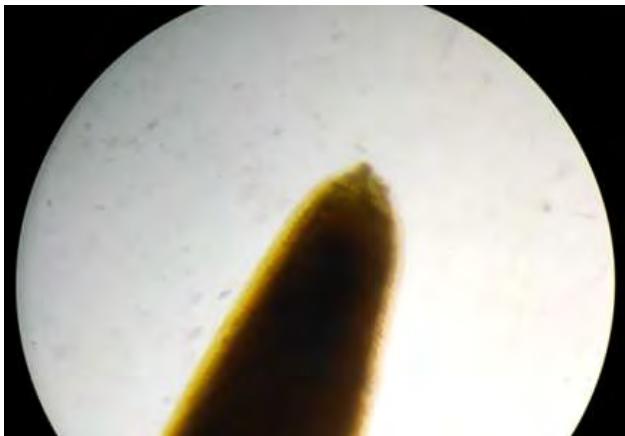


Figure 13: Posterior end of female *Ophidascaris* sp.



Figure 14 (a): Metacestode of *Taenia* sp. Anterior end showing scolex.

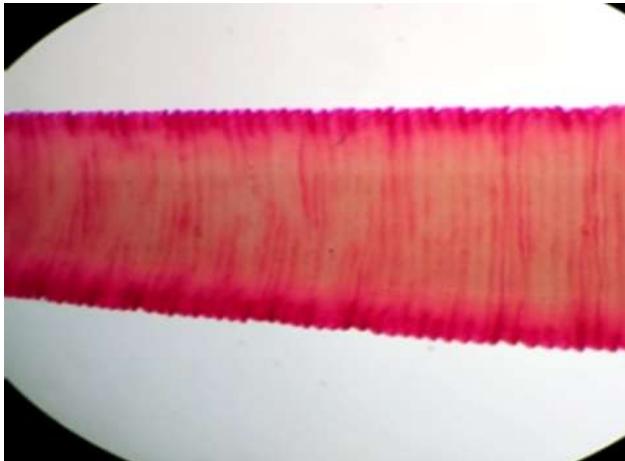


Figure 14 (b): Metacestode of *Taenia* sp. Mid-section showing segments.



Figure 14 (c): Metacestode of *Taenia* sp. Posterior end showing bulb.



Figure 15: Cystacanth of Acanthocephala.



Figure 16: Acanthocephala with no pseudosegmentation.

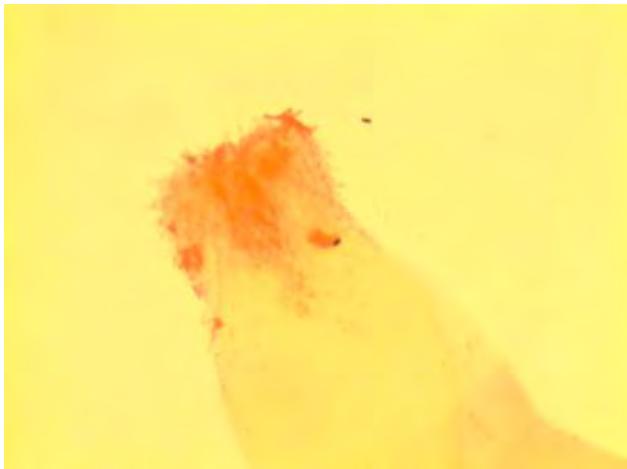


Figure 17: Anterior part of Acanthocephala with hooks.

were seen in the aorta (Sprent, 1963). Third-stage larvae of *Ophidascaris moreliae* were either found free or attached to the surface of the lungs (Sprent 1970a). Adults of *Ophidascaris moreliae* are found in the stomach and intestine (Jacobson, 2007). An adult king cobra (*Ophiophagus hannah*) was infected with mature *Sphaerchinorhynchus serpenticola* (Jacobson, 2007). Cystacanths of the acanthocephalan *Macracanthorhynchus ingens* were identified in several species of snakes in Louisiana (Elkins and Nickol, 1983). Cystacanths of *Centrorhynchus spinosus* and *Macracanthorhynchus ingens* were identified in eastern indigo snakes in Florida (Foster et al., 2000)

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