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Cestodes from deep-water squaliform sharks in the Azores

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ABSTRACT

The majority of our knowledge on marine tapeworms (cestodes) is limited to taxa that are relatively easy to obtain (i.e., those that parasitize shallower-water species). The invitation to participate in a deep-water research survey off the Condor seamount in the Azores offered the opportunity to gain information regarding parasites of the less often studied sharks of the mesopelagic and bathypelagic zone. All tapeworms (Platyhelminthes: Cestoda) found parasitizing the spiral intestine of squaliform shark species (Elasmobranchii: Squaliformes) encountered as part of this survey, as well as some additional Azorean sampling from previous years obtained from local fishermen are reported. In total, 112 shark specimens of 12 species of squaliform sharks representing 4 different families from depths ranging between 400 and 1290 m were examined. Cestodes were found in the spiral intestines from 11 of the 12 squaliform species examined: *Deania calcea*, *D.* cf. *profundorum*, *D. profundorum*, *Etmopterus princeps*, *E. pusillus*, *E. spinax*, *Centroscyllium fabricii*, *Centroscymnus coelolepis*, *C. cryptacanthus*, *C. crepidater*, and *Dalatias licha*. No cestodes were found in the spiral intestines of *Centrophorus squamosus*. Light microscopy and scanning electron microscopy revealed several potentially novel trypanorhynch and biloculated tetraphyllidean species. Aporhynchid and gilquiniid trypanorhynchs dominated the adult cestode fauna of *Etmopterus* and *Deania* host species, respectively, while larval phyllobothriids were found across several host genera, including, *Deania*, *Centroscyllium*, and *Centroscymnus*. These results corroborate previous findings that deep-water cestode faunas are relatively depauperate and consist primarily of trypanorhynchs of the families Gilquiniidae and Aporhynchidae and larval tetraphyllideans. A subset of specimens of most cestode species was preserved in ethanol for future molecular analysis to allow more definitive determinations of the identification of the larval tetraphyllideans and trypanorhynchs lacking evaginated tentacles and other key diagnostic features.

1. Introduction

Our understanding of the tapeworms that parasitize many of the world's ~1200 species of sharks and rays (i.e., elasmobranchs, see Naylor et al., 2012a, 2012b) has expanded considerably over the past two decades (e.g., see Jensen, 2005; Palm, 2004; Ruhnke, 2011; Tyler, 2006). This work has placed particular emphasis on hosts occupying epibenthic and epipelagic habitats (i.e., depths of ≤200 m) in large part because animals inhabiting these relatively shallow waters are readily accessible and easily procured in collaboration with artisanal fishing enterprises. This is not, however, the case for deeper water elasmobranchs, and sharks in particular. With respect to the two most speciose groups of deep-water sharks (i.e., Squaliformes and Scyliorhinidae), in the most recent comprehensive synopsis of the metazoan parasites of deep-water fishes of the world, Klimpel et al.'s (2001) checklist included cestode (tapeworm) data for only 12 of the 121 species of dogfish sharks (Squaliformes) and only 1 of the 154 species of catsharks (Scyliorhinidae) listed by Compagno et al. (2005). This checklist reveals that the majority of cestodes found parasitizing deep-water hosts consist of larval cestodes in the order Tetraphyllidea and adult cestodes of the order Trypanorhyncha. Although Palm (2004) expanded trypanorhynch cestode records to include 27 species of squaliform hosts and 6 species of scyliorhinid hosts, the tapeworm faunas of the majority of species of deep-water sharks are completely unknown.

The Condor seamount project provided an invaluable opportunity for us to expand knowledge of tapeworms of deeper water sharks, and squaliforms in particular. Some of the sharks examined for the purposes of this study had never previously been examined for cestodes; most had never been examined in this region. In fact, to our knowledge, existing data for adult cestodes from deep-water sharks in the vicinity of the Azores come primarily from M.S. (e.g., Schröder, 1999) and Ph.D. (e.g., Noever, 2009) theses, only a subset of the results of which have been formally published (e.g., Palm and Schröder, 2001), in the latter case for the Great Meteor Bank in the central northeast Atlantic, 1000 km south of

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the Azores. Guiart's (1935) report of cestodes from deep-water sharks in the Azores includes only larvae.

Our goal here is to provide preliminary identifications and basic morphological data using scanning electron microscopy and/or light microscopy for the cestode taxa found parasitizing the squaliform shark species examined over the course of the surveys. With the exception of Noever et al. (2010) formal descriptions of the novel tapeworm taxa encountered during this study have yet to be generated and will be published separately, as will the results of analyses conducted on specimens of each tapeworm species preserved for the generation of molecular data. To aid future work we have provided images of most cestode taxa encountered and have deposited representative voucher material in the Lawrence R. Penner Parasitology Collection, University of Connecticut, Storrs, Connecticut, United States. It is our hope that the preliminary morphological identifications provided here can be addressed using molecular data in the near future.

2. Materials and methods

Sharks collected in May 2006, January 2009, and October 2012 off Faial Island, Azores resulted in a total of 112 individuals of 12 species of deeper water sharks being examined for cestodes. Specimens in 2006 and 2009 were obtained from local fishermen, which were caught as by-catch on longlines. In 2012 sharks were obtained from longline fish surveys conducted by the University of the Azores on the R.V. *Arquipélago* in conjunction with the Condor seamount studies (Giacomello et al., 2013), and were collected at depths between 400 m and 1290 m around the Condor seamount between 38°41'43″N, 29°20'12″W and 38°31'24″N, 28°59'30″W. Data reported here are for all squaliform taxa examined. With two exceptions host identities and family-level classification follow Naylor et al. (2012a, 2012b). Specimens examined were as follows: Centrophoridae: 4 *Centrophorus squamosus*, 13 *Deania calcea*, 1 *Deania profundorum*, and 1 *Deania* cf. *profundorum* (*sensu* Naylor et al., 2012b); Etmopteridae: 11 *Etmopterus princeps*, 15 *Etmopterus pusillus*, 23 *Etmopterus spinax*, and 6 *Centroscyllium fabricii*; Somniosidae: 14 *Centroscymnus coelolepis*, 7 *Centroscymnus cryptacanthus* (treated as *Centroscymnus owstonii* by Naylor et al., 2012a, 2012b), and 10 *Centroscymnus crepidater* (treated as *Centroselachus crepidater* by Naylor et al., 2012a, 2012b); Dalatiidae: 7 *Dalatias licha*. Additional details regarding sex, size, and depth of collection for all shark species are provided in Table 1. The specific identity of 30 of the 112 specimens was verified in the molecular analysis of the NADH2 gene in Naylor et al. (2012b). Voucher tissue samples preserved in 95% ethanol were taken from all 112 specimens; photographs of each shark were also taken. The unique host specimen identification numbers of infected individuals (collection code "AZ" followed by collection number) are provided in relevant sections below, and correspond to those used in Naylor et al. (2012b). Images of most specimens and detailed collection and demographic data of all specimens can be accessed at the following URL: www.elasmobranchs.tapewormdb.uconn.edu by entering the collection code AZ and the appropriate collection number.

Data presented are for taxa parasitizing the spiral intestine only. The spiral intestine of each shark was opened with a mid-ventral longitudinal incision, and was examined for cestodes under a stereomicroscope. Cestodes found were preserved in either 95% ethanol for future molecular work or 4% seawater buffered formalin for further morphological examination. The spiral intestines were then fixed in 4% seawater buffered formalin for approximately a week and then transferred to 70% ethanol for long-term storage and further study. Preparations of cestodes as whole mounts and for examination with scanning electron microscopy (SEM) followed standard protocols (e.g., Pickering and Caira, 2008; Rodriguez et al., 2011).

Museum abbreviations used are as follows: IPCAS, Institute of Parasitology, Academy of Sciences of the Czech Republic, České Budějovice, Czech Republic; LRP, Lawrence R. Penner Parasitology Collection, University of Connecticut, Storrs, Connecticut, United States; MHNFCUP, Museu de História Natural da Faculdade de Ciências da Universidade do Porto, Porto, Portugal; USNPC, United States National Parasite Collection, Beltsville, Maryland, United States. Larval cestode terminology follows Chervy (2002); microthrix terminology follows Chervy (2009). Trypanorhynch classification follows Palm (2004) and Olson et al. (2010).

3. Results

In total, 48 (i.e., 43%) of the 112 squaliform sharks examined were found to host cestodes of any life cycle stage (i.e., larva, juvenile, or adult) in their spiral intestine (see Table 1); 35.7% hosted juvenile or adult cestodes. The overall prevalence of infection with cestodes in each species of shark ranged from 0% (*Centrophorus squamosus*) to 73.9% (*Etmopterus spinax*). Infections ranged from 1 to 3 cestode species per infected shark. The average number of cestode species (regardless of life cycle stage) found per shark species was 1.75 worm species; excluding larval stages, the average was 1.17. However, as noted below in the detailed

Table 1
Detail of squaliform shark species examined for cestodes and number infected by sex.

Shark species	Total no. examined	Depth collected (m)	Shark total length (cm)	No. examined (No. infected) females	No. examined (No. infected) males
Centrophoridae					
Centrophorus squamosus	4	1090–1265	121–129	4 (0)	–
Deania calcea	13	1090–1265	81–112	10 (6)	3 (0)
Deania cf. *profundorum*	1	540	95	1 (1)	–
Deania profundorum	1	451–500	41	1 (1)	–
Dalatiidae					
Dalatias licha	7	401–540	114.5–123	–	7 (4)
Etmopteridae					
Centroscyllium fabricii	6	1260–1290	57–81.5	5 (1)	1 (0)
Etmopterus princeps	11	1090–1290	48–69.5	7 (0)	4 (1)
Etmopterus pusillus	15	500–850	24.5–48.5	7 (2)	8 (1)
Etmopterus spinax	23	540–750	30–43	22 (16)	1 (1)
Somniosidae					
Centroscymnus coelolepis	14	1090–1290	85–118.5	6 (5)	8 (4)
Centroscymnus owstonii	7	1090–1265	84.5–114	6 (3)	1 (0)
Centroselachus crepidater	10	1051–1265	61–84.5	8 (2)	2 (0)