16 Parasites of Marine Turtles

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CONTENTS

16.1	Introduction	428	
	16.1.1 Methods of Helminth Recovery and Parasite Diagnosis		
	16.1.2 Trematodes		
	16.1.3 Nematodes		
	16.1.4 Cestodes	434	
	16.1.5 Protozoa		
	16.1.5 Protozoa 16.1.6 Annelids and Arthropods	a068434	cb50
	16.1.7 Parasitological Principles	434	brary
16.2	Loggerhead (<i>Caretta caretta</i>) Parasites		
	16.2.1 Aspidogastrid Flukes		
	16.2.2 Blood Flukes (Digenea: Spirorchiidae)		
	16.2.3 Gastrointestional Flukes (Digenea: Brachycoelidae, Pachypsolidae,	5161202169/MD/3	
	Plagiorchiidae, Pronocephalidae, Styphlotrematidae, and Telorchiidae)		
	16.2.4 Liver Flukes (Digenea: Calycoididae)		
	16.2.5 Urinary Bladder Flukes (Digenea: Gorgoderidae)		
	16.2.6 Larval Tapeworms (Cestoda: Trypanorhyncha)		
	16.2.7 Nematodes (Nematoda: Cuculidae, Kathlanidae, Anisakidae)		
	16.2.8 Comparison with Other Helminth Surveys		
	16.2.9 Protozoa (Eucoccidiorida: Eimeridae)		
16.3	Green Turtle (Chelonia mydas) Parasites		
	16.3.1 Blood Flukes (Digenea: Spirorchiidae)		
	16.3.2 Gastrointestinal Flukes (Digenea: Brachycoelidae, Plagiorchiidae,		
	Pronocephalidae, Rhytidodidae, Telorchiidae, Microscaphidiidae,		
	and Cladorchiidae)		
706013	16.3.3 Liver Flukes (Digenea: Rhytidodidae and Plagiorchiidae)		
ary	16.3.4 Urinary Bladder Flukes (Digenea: Gorgoderidae)		
	16.3.5 Coccidians (Eucoccidiorida: Eimeridae, Cryptosporidae)		
	16.3.6 Ciliates (Vestibuloferida; Balantidiidae)		
	16.3.7 Flagellates (Diplomonadida: Hexamitidae, Trichomonadida: Trichomonadida		
	16.3.8 Amoebae (Entamoebidae)		
	16.3.9 Comparison with Other Helminth Surveys		
16.4	Hawksbill (Eretmochelys imbricata) Parasites		
	16.4.1 Blood Flukes (Digenea: Spirorchiidae)		
	16.4.2 Gastrointestinal Flukes (Digenea: Plagiorchiidae, Pronocephalidae,		
	and Rhytidodidae		
	16.4.3 Urinary Bladder Flukes (Digenea: Gorgoderidae)		
	16.4.4 Comparison with Other Surveys and a Listing of Species Known		
	from Hawkshills	443	

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16.5 Leatherback (Dermochelys coriacea) Parasites	
16.5.1 Comparison with Other Surveys and a Listing of Species	
Known from Leatherbacks	
16.6 Kemp's Ridley (Leptochelys kempii) Parasites	
16.7 Olive Ridley (Lepidochelys olivacea) Parasites	
Acknowledgments	
References	

16.1 INTRODUCTION

The parasite fauna of marine turtles is varied and diverse. It includes helminths, protozoa, arthropods, and annelids. The amazing thing is that some of these marine turtles migrate great distances through the oceans and yet they must reside in waters that allow them to be infected by microscopic infective stages at some point in their journeys. These parasites are evidently well adapted to their reptilian hosts as some are very common parasites and others are more rarely detected. The parasites have been associated with these turtles for a very long time as many organs of the turtles harbor brary parasites. Marine turtles are usually the final or definitive host of the parasites, but sometimes they serve as a source of infection to other vertebrates that prey upon them. Due to the intended shortness of this chapter, focus will be on the parasites in the marine turtles in Florida waters as work on these turtles has been done in my former laboratory over the past 25 years. There is a wealth of information on parasites of marine turtles, but much of it is based on examinations of one or few turtles. My research reflects taking advantage of turtles that have died in Florida. Carcasses were submitted to my laboratory through cooperation with turtle rehabilitation facilities and state and federal turtle biologists. This work was done through federal permits held by Dr. Elliott R. Jacobson.

16.1.1 METHODS OF HELMINTH RECOVERY AND PARASITE DIAGNOSIS

The plastron and legs were removed from the carcass and the viscera were removed into a container. The entire gastrointestinal system was separated from the remainder. The esophagus and stomach were placed into separate containers and the intestine was divided into equal thirds and each section labeled and isolated. Each section was slit open and the mucosa was washed to free most parasites into the container and then the mucosa was visually inspected. One end of a section of intestine was then placed between two fingers of one hand and the entire length was pulled through the fingers to ebrar strip remaining worms and ingesta into the container. Solid organs like the liver and kidney were sliced and pressed in a container with water to allow worms in the blood vessels and bile ducts to be expressed into the water. The urinary bladder was removed and opened into its own container. The heart and associated major vessels were slit open and washed into a container. The contents of each container were then placed into a standard #50 sieve separately and the contents were washed with gentle water spray. This process cleaned and concentrated the worms. The sieve was back flushed into a container and contents were then viewed in a light box. The worms were removed, separated by species, counted, fixed, and stored in 70% ethanol in vials with host ID and organ and species name. It becomes obvious that helminths have a primary normal site of infection within the turtle, but you must realize that post mortem these worms move into adjacent organs. So, depending upon the time span between the turtle death and the necropsy, where the parasites might be found might change.

The primary means of diagnosing mature helminths is by fecal examination to detect eggs. Two basic methods are used. The most common is the fecal flotation in which a salt or sugar solution of an appropriate specific density will be used to cleanse and concentrate many helminth eggs, protozoan cysts, and coccidian oocysts. I prefer sodium nitrate, available commercially as Fecasol, as I find it floats out a diversity of parasite stages. Whereas some parasitologists feel one must use centrifugation to find these stages, I disagree and use a standing flotation. The problem with marine turtle parasite diagnostics is that the majority of helminths in these hosts are trematodes. Their eggs

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Wyneken, Jeanette. Marine Biology, Volume 14 : Biology of Sea Turtles, Volume III. : CRC Press, . p 448 http://site.ebrary.com/id/10657402?ppg=448 Copyright © CRC Press. . All rights reserved. May not be reproduced in any form without permission from the publisher, except fair uses permitted under U.S. or applicable copyright law. do not float and thus we use a fecal sedimentation method in which the cleansing and concentrating are done by allowing the eggs to sediment to the bottom of a tube, rather than float to the top. A very simple system is to place a drop or two of liquid dish soap in a 500 mL squeeze bottle, fill it with water, and shake to mix. Feces (1-2 g if possible) is placed into a small container such as a urine sample cup and 30-40 mL of the soapy water is added. The feces must be broken apart as well as possible. Place a single layer of gauze over the opening and pour the contents into a 50 mL centrifuge tube. You may need to rewash the sediment back into the container and strain the contents into the tube a second time. Do not allow the tube to overflow. Place the tube in a rack so it remains vertical and allow it to stand for 5 min. Decant or aspirate the fluid down close to the sediment and then refill with the soapy water. You will repeat this twice and then switch from the soapy water to plain water as this will reduce the soap bubble problem. Continue the process until the diluent remains clear and then let it settle once more. Aspirate off the diluent down to the sediment. All of the aspirated diluent may be discarded. Place some of the sediment onto a microscope slide and add a coverslip. If you add too much sediment, you may miss the eggs. Scan the coverslip systematically until it is all viewed. You may wish to make and scan more than one slide preparation. You should echoo also know that all diagnostic stages including those that float, such as nematode eggs and oocysts, brary will sediment. Some eggs will be numerous and others will be in low numbers. Compare what you see with either procedure to the photos of eggs of some of the helminths of turtles (Figure 16.1, 23 through 50). If you are able to examine very fresh feces, you may also use a direct smear procedure to look for motile protozoa. Place a few drops of normal saline on a microscope slide and with an applicator stick, add some feces and mix it thoroughly in the saline and then add a coverslip. Then scan the entire coverslip as described earlier. You will possibly find eggs on these slides, but not in the quantity you will after concentrating them. You need to have an ocular micrometer on your microscope that is calibrated so you can measure the eggs as that is a crucial element in the identification of these eggs along with shape, and the presence of absence of an operculum.

16.1.2 TREMATODES

Flukes are the most diverse and numerous parasites in marine turtles. Most of these in marine turtles are digenetic trematodes (flukes), which have complex life cycles sometimes requiring one to three other species of animal to complete their life history. They are flat worms, no body cavity is present, and both sets of reproductive systems are present in each adult. They may be found in most body organs in these turtles, but are usually restricted to a primary site for development. Because different species of marine turtles eat different things as a normal dietary regimen, this might influence the diversity of flukes present in turtles, as those that have a more select or restricted diet such as hawksbills and leatherbacks seem to have a less diverse fauna than omnivores as loggerheads. Flukes exist in all the major marine turtle species and sometimes numbered in the thousands per infected host. A single species of aspidograstrid trematode, a distinct and separate type of fluke, occurs only in the loggerhead.

16.1.3 NEMATODES

Nematodes are another component of the helminthic community and these are not nearly as diversified as the trematodes in marine turtles. They are referred to as round worms as they are tubular in morphology, they have a body cavity, and have a single sex in each adult. Their life cycles may be complex, but the most of those in marine turtles usually only infect the turtles and have direct life cycles. Most nematode parasites of marine turtles reside in the gastrointestinal tract, but one taxon lives in the lungs. Infections of one species of nematode (*Anisakis*) resulted from captive greens being fed sardines and larvae did not develop to maturity and were found embedded in the stomach walls at necropsy causing gastric ulcerations (Burke and Rogers, 1962), and this parasite has been reported in free-ranging turtles in the Mediterranean region and will be discussed later.

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The Biology of Sea Turtles, Volume III



FIGURE 16.1 Identifying adult trematodes requires measurements of size, examination of the shape, realizing the relative position and number of the testes, position of the ovary, extent and position of the vitellaria, one versus two suckers and their location on the body, length of the intestinal caeca, and characteristics of the termination region of the male. Some of these flukes may not be in proper proportion with respect to others in order to assemble the Plate. Figures 1 through 12 are adult flukes from loggerheads and 13 through 22 are from green turtles. 1. Lophotaspis vallei, 7 mm, 2. Carettacola bipora, 8 mm, 3. Hapalotrema mistroides, 5 mm, 4. Neospirorchis pricei, 10 mm, 5. Cymatocarpus undulatus, 6 mm, 6. Pachypsolus irroratus, 4 mm, 7. Pleurogonius trigonocephalus, 4 mm, 8. Diaschistorchis pandus, 5 mm, 9. Rhytidodes gelatinosus, 8 mm, 10. Calycodes anthos, 12 mm, 11. Orchidasma amphiorchis, 9 mm, 12. Plesiochorus cymbiformis, 6 mm,

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FIGURE 16.1 (continued) Identifying adult trematodes requires measurements of size, examination of the shape, realizing the relative position and number of the testes, position of the ovary, extent and position of the vitellaria, one versus two suckers and their location on the body, length of the intestinal caeca, and characteristics of the termination region of the male. Some of these flukes may not be in proper proportion with respect to others in order to assemble the Plate. Figures 1 through 12 are adult flukes from loggerheads and 13 through 22 are from green turtles. 13. Learedius learedi, 5 mm, 14. Enodiotrema megachondrus, 9 mm, 15. Cricocephalus albus, 3.5 mm, 16. Deuterobarus proteus, 8 mm, 17. Pronocephalus obliquus, 4.5 mm, 18. Microscaphidium reticulare, 5 mm, 19. Angiodyctium parallelum, 8 mm, 20. Octangium sagitta, 4 mm, 21. Rhytidoides similis, 5 mm, 22. Schizamphistomoides spinulosus, 7 mm,

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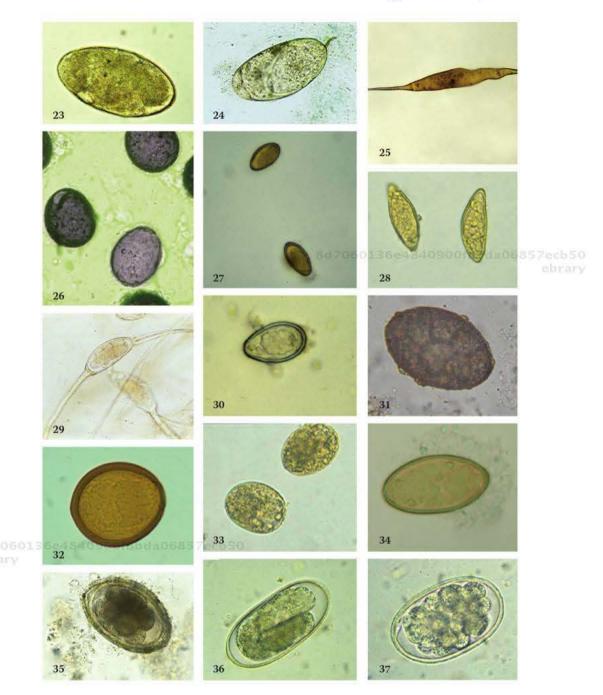


FIGURE 16.1 (continued) Fluke eggs are based on size and morphology. Most of them have an operculum (trap door for larval escape, see Figure 45 which has its operculum open), what has developed inside the egg, shape of the egg, and if there are polar spines or filaments and how many and their length. 23. Lophotaspis vallei egg 169×68 μ m, 24. Carettacola bipora egg 88×38 μ m, 25. Hapalotrema mistroides egg 414×36 μ m, 26. Neospirorchis sp. egg 45×35 μ m. 27. Cymatocarpus undulatus egg 37×16 μ m, 28. Pachypsolus irroratus egg 51×18 μ m, 29. Pleurogonius sindhi egg capsule 52×22 μ m, 30. Diaschistorchis pandus egg 40×22 μ m, 31. Rhytidodes gelatinosus egg 75×53 μ m, 32. Orchidasma amphiorchis egg 60×50 μ m, 33. Plesiochorus cymbiformis egg 41×28 μ m. 34. Styphlotrema solitaria egg 32×20 μ m, 35. Sulcascaris sulcata egg 91×57 μ m. 36. Tonaudia/Kathlania egg 104×55 μ m, 37. Cuculanus carretae egg 91×57 μ m,

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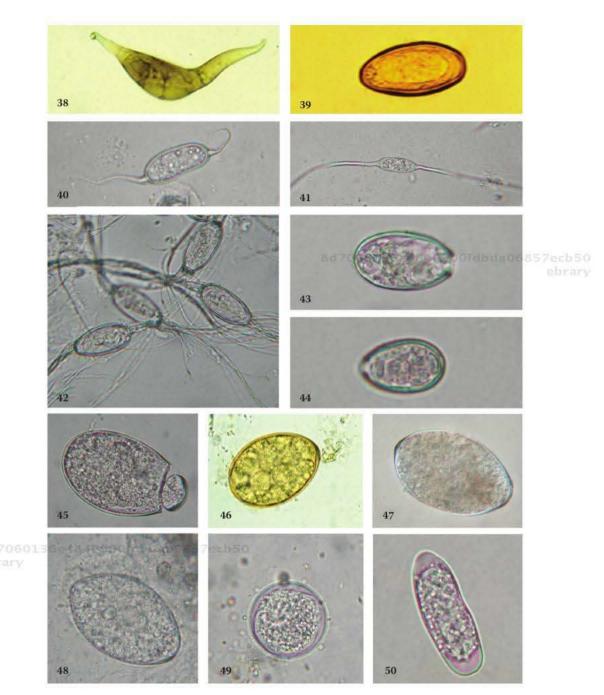


FIGURE 16.1 (continued) Fluke eggs are based on size and morphology. Most of them have an operculum (trap door for larval escape, see Figure 45 which has its operculum open), what has developed inside the egg, shape of the egg, and if there are polar spines or filaments and how many and their length **38**. *Learedius learedi* egg 297×42 µm, **39**. *Enodiotrema megachondrus* egg capsule 42×21 µm, **40**. *Cricocephalus albus* egg capsule 33×14 µm, **41**. *Pronocephalus obliquus* egg capsule 33×14 µm, **42**. *Desmogonius desmogonius* egg capsule 32×13 µm, **43**. *Charaxicephalus robustus* egg 26×16 µm 44. *Angiodyctium parallelum* egg 25×15 µm, **45**. *Deuterobarus proteus* egg 94×34 µm, **46**. *Rhytidodes gelatinosus* egg 68×37 µm, **47**. *Schizamphistomum sceloporum* egg, 98×60 µm, **48**. *Octangium sagitta* egg 89×57 µm, **49**. *Balantidum bacteriophorus* cyst 58 µm, and **50**. *Caryospora cheloniae* sporocyst 41×17 µm.

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16.1.4 CESTODES

Adult cestodes are segmented and are flat worms with solid bodies and both sexes are in the same individual worm. Tapeworms need an intermediate host to complete their life cycles. Marine turtles function as intermediate hosts for tapeworms (trypanorhynchs) that mature in sharks. The shark becomes infected by feeding on turtles containing these larvae. The ornate larvae have four long spiny tentacles, which anchor the worm into the intestinal mucosa of the final host. These larvae may be distributed throughout the peritoneal cavity, often associated with the mesenteries or attached to the peritoneal surface of the gut.

16.1.5 PROTOZOA

While there are many types of these single-celled eukaryotic parasites, only a few have been reported from marine turtles. Two species of coccidian have been named from marine turtles along with one species of ciliate. Amoebae and flagellates also have been reported. All of these appear to have a direct life cycle; and while some develop within cells, others remain in the lumen of the intestines.

16.1.6 ANNELIDS AND ARTHROPODS

Some parasites live inside the host and others live on the outside and the latter are ectoparasites. Most arthropods would be ectoparasites, but one mite is internal and that is *Chelonacarus elongatus*, which lives in the wall of the cloaca of green turtles (Pence and Wright, 1998). Another arthropod, which is associated with marine turtles occasionally, is a biting midge. The concept of female turtles being fed upon by hematophagous arthropods while they are on shore laying eggs has been demonstrated. A biting midge (*Culicoides phlebotomus*) has been recovered feeding on nesting leatherbacks in Costa Rico (Borkent, 1995). The epibiota will be dealt with in Chapter 10 by Frick and Pfaller and will not be covered here.

16.1.7 PARASITOLOGICAL PRINCIPLES

The intrigue of parasites is amazing. Many questions remain unsolved, such as how host-specific these helminths are. We know that many parasites demonstrate loose host specificity, but some are only able to infect a single definitive host species. Others fit on a continuum between these extremes. Examples of species-specific host specificity in marine turtles would include Lophotaspis vallei, Learedius learedi, and Styphlotrema solitaria (see Tables 16.1, 16.2, 16.3 and 16.4). Species with a wider host range would include Plesiochorus cymbiformis, Rhytidodes gelatinosus, Enodiotrema carettae, and Pleurogonius trigonocephalus (see Tables 16.1, 16.2, 16.3 and 16.4). These conclusions are based on findings from turtles examined in Florida and other regions of the world as will be realized in the discussions on individual turtles. How much harm these parasites cause to their final hosts is poorly understood. We know that the Spirorchiidae (blood flukes) cause a great deal of damage (Stacy et al., 2010a). There might be a threshold in numbers of a single species when the host will suffer consequences and when one adds up insults caused by the presence of several parasite species. The extent of damage could vary from case to case depending on the species involved. The site where most of the individuals of a species were recovered is indicated in the Tables, but this does not show the range of habitats utilized by each species. The beauty of this variation is that the helminthic community has evolved to use a wide range in habitats and niche selection. Moreover, not all parasites infect all hosts. Variation in the intensity of infection may be influenced by the complexity of the life cycle, the availability of intermediate hosts where they are needed, interspecific effects of one parasite species on another, immune response by the host, and turtle population density. Obviously if too many of a pathogenic species of helminth are present,

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		Prevalence (%)	Intensity		Total	Normal	
Species	#Pos.		Mean	Range	Worms	Habitata	
Lophotaspis vallei	10	22.7	30.7	2-119	307	St	
Carettacola bipora	19	43.1	4.9	1-13	94	H, Ar	
Hapalotrema mistroides	11	25.0	2.6	1–9	29	Lv	
Neospirorchis pricei	3	6.8	11.0	10-12	33	H, Ar	
Cymatocarpus undulatus	25	56.8	288.8	2-1,350	7,224	UI	
Pachypsolus irroratus	14	31.8	54.6	3-202	764	Es, St	
Enodiotrema megachondrus	13	29.5	57.9	2-574	753	St	
Enodiotrema carettae	16	36.3	7.7	1-42	123	UI	
Pleurogonius trigonocephala	24	54.5	51.7	1-289	1,280	MI	
Diaschistorchis pandus	11	25.0	53.17	062213860	4845840	Of Es, St 06	857ecb50
Pyelosomum renicapite	6	13.6	16.8	1–54	101	MI, LI	ebrary
Pyelosomum chelonei	4	9.1	79.5	2-286	318	LI	
Rhytidodes gelatinosus	23	52.6	161	1-882	3,703	UI	
Styphlotrema solitaria	11	25.0	70.1	1-395	771	UI	
Orchidasma amphiorchis	25	56.8	825.8	6-3,918	20,644	UI	
Calycodes anthos	12	27.3	61.6	1-164	739	L, GB	
Plesiochorus cymbiformis	30	68.2	68.4	2-190	2,051	UB	
Trypanorhynch cysts	2	4.5	8	2007	<u>200</u> 3	Me	
Cuculanus carretae	20	45.4	180.4	1-465	3,609	UI	
Kathlania leptura	19	43.2	713.6	2-10,212	13,558	MI	
Tonaudia tonaudia	22	50.0	529.8	1-3,714	11,655	LI	
Sulcascaris sulcata	23	52.7	230.6	1-2,416	5,072	St	

TABLE 16.1 Summary of Adult Worms from 44 Loggerheads 1991–2006 from Florida

No loggerheads without worms present. Number of worm species/infected host mean=8.7 (2-17).

^a Ar, aorta; H, heart; Lv, liver blood vessels; L, liver; GB, gall bladder; Es, esophagus; St, stomach; UI,

upper intestine; MI, middle intestine; LI, lower intestine; UB, urinary bladder; Me, mesenteries.

Bd70 there could be a negative impact on the parasite as well because if the host dies, there will be no **ebrar** more progeny from it. So through evolutionary change in long-standing relationships, parasites usually become adapted to co-exist with their hosts.

Terms used in this chapter that need definition include the following (see Bush et al., 1997):

- Prevalence is the number of hosts infected by a parasite species divided by the number of hosts examined and then multiplied by 100 and is expressed as a percentage.
- Intensity is the number of parasites of a species divided by the number of hosts infected with that species.
- Mean intensity is the total number of individual parasites of a species divided by the number of infected hosts.

16.2 LOGGERHEAD (CARETTA CARETTA) PARASITES

Seventeen trematodes species, four species of nematodes, and larval trypanorhynch tapeworms were recovered from 44 loggerheads from Florida at necropsy (Table 16.1). The number of species per host ranged from 2 to 17 with an average of 8.7 species per turtle. The intensity of infection ranged from 1 to 10,212 for individual species. All loggerheads were infected with helminths. While this

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		Prevalence	Int	tensity	Total	Normal	
Species	#Pos.	(%)	Mean	Range	Worms	Habitatª	
Learedius learedi	10	13.1		1-16	53	H, MV	
Neospirorchis sp.	2	2.6	3.5	1-7	7	H, MV	
Cymatocarpus undulatus	1	1.3	26	26	26	UI, MI	
Enodiotrema megachondrus	6	8.1	13.8	1–79	85	LI	
Enodiotrema carettae	2	2.6	40.5	2-79	81	L, GB	
Pleurogonius trigonocephalus	3	3.9	71	1-216	219	MI	
Pleurogonius sindhi	15	19.7	41.1	1-221	617	MI	
Pleurogonius longisculus	7	9.4	8.8	2-17	62	MI	
Pleurogonius lobatus	3	3.9	8.0	2-18	24	LI	
Pleurogonius linearis	2	2.6	8170	6013§e48	840900	fd <mark>s</mark> tda06857	ecb50
Diaschistorchis pandus	1	1.3	10	10	10	Es, St	ebrary
Cricocephalus albus	47	63.5	57.5	1-524	2,702	St	
Pronocephalus obliquus	30	40.5	74.4	1-416	2,232	MI	
Metacetabulum invaginatum	27	36.5	15.6	1-172	421	UI	
Desmogonius desmogonius	4	5.4	6.0	1-11	24	Es, St	
Charaxicephalus robustus	8	10.8	9.5	2-31	76	St	
Rhytidodes gelatinosus	10	13.5	10.1	1-49	101	UI	
Rhytidoides similis	16	21.6	19.0	1-98	304	L, GB	
Orchidasma amphiorchis	4	5.4	56.2	10-114	225	UI	
Plesiochorus cymbiformis	4	5.4	2.5	1-6	9	UB	
Deuterobarus proteus	30	40.5	125	1-1,006	3,750	MI, LI	
Angiodictyum paralellum	17	22.9	112.8	1-787	1,918	MI	
Microscaphidium reticulare	20	27.0	244.3	1-2,449	4,887	UI	
Neoctangium travassosi	25	33.8	67.2	1-117	1,680	MI	
Octangium sagitta	41	55.4	378.6	1-4833	15,524	MI	
Polyangium linguatula	18	24.3	38.3	1-128	689	MI	
Schizamphistomoides spinulosus	35	47.3	14.3	1-63	501	MI	
Schizamphistomum sceloporum	1	1.3	4	4	4	LI	
Trypanorhynch larvae d a 0685	7e1450	16.9	200	_	STIT	ME	

TABLE 16.2 Summary of Adult Worms from 74 Green Turtles 1991–2006 from Florida

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Six greens contained no helminths. Number of worm species/infected host mean and range=6.3 (1–16). ^a H, heart; MA, major arteries; L, liver; GB, gall bladder; Es, esophagus; St, stomach; UI, upper intestine;

MI, middle intestine; LI, lower intestine; UB, urinary bladder; Me, mesenteries.

chapter will deal primarily with parasites recovered in the parasitology laboratory at the College of Veterinary Medicine, University of Florida, there are other worms that have been reported from loggerheads not found in this survey.

16.2.1 ASPIDOGASTRID FLUKES

A single species of this order of trematodes infects loggerheads and no other marine turtle species. *L. vallei* (Figure 16.1, 1) has a subdivided sucker (= holdfast) covering most of its ventral surface, which readily distinguishes it from all other flukes in these hosts. It is believed that the aspidogastrids in vertebrates use a single gastropod as an intermediate host (Wharton, 1939). It was found in 22.7% of the loggerheads with an average of 30.7 individuals per infected host.

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TABLE 16.3 Summary for Adult Worms from Four Hawksbills from Florida 1991–2006

		Inte	nsity	Total	Normal	
Species	#Pos./#Exam.	Mean	Range	Worms	Habitata	
Neospirorchis sp.	1/4	9	9	9	В	
Rhytidodes gelatinosus	3/4	16	9-45	54	UI	
Enodiotrema carettae	1/4	1	1	1	L	
Cricocephalus albus	2/4	3	2-4	6	St, UI	
Diaschistorchis pandus	2/4	12	12	24	St	
Plesiochorus cymbiformis	1/4	3	3	3	UB	
Trypanorhynch cysts	1/4		31-12		<u>201</u> 3	

All hawksbills contained parasites. Number of worm species/infected host mean=3 (2-4). bda06857ecb50 ^a B. brain; L, liver; St, stomach; UI, upper intestine; UB, urinary bladder.

TABLE 16.4

Summary of Adult Worms from Four Kemp's Ridley Turtles from Florida 1991–2006

	Species		Ir	ntensity	Total	Normal Habitatª	
		#Pos./#Exam.	Mean	Range	Worms		
	Carettacola bipora	1/4	1	1	1	L vessels	
	Cymatocarpus undulatus	2/4	5,904	222-11,809	11,809	MI	
	Cricocephalus albus	2/4	4	1–7	8	St	
	Pleurogonius trigonocephalus	1/4	5	5	5	MI	
	Rhytidodes gelatinosus	1/4	2	2	2	UI	
	Enodiotrema megachondrus	1/4	22	22	22	UI	
1706013	Enodiotrema carettae	7 och 51/4	5	5	5	UI	
rarv	Calycodes anthus	1/4	4	4	4	UI	
	Tonaudia tonaudia	2/4	932	639-1,226	1,865	MI	
	Sulcascaris sulcata	1/4	1	1	1	St	
	Trypanorhynch cysts	2/4				St, UI walls	

All Kemp's Ridleys were infected. Number of worm species/infected host mean=6 (2-10).

^a L, liver; St, stomach; UI, upper intestine; MI, middle intestine.

16.2.2 BLOOD FLUKES (DIGENEA: SPIRORCHIIDAE)

Adults of three species of this family were recovered from blood vessels. They were *Carettacola bipora* (Figure 16.1, **2**), *Hapalotrema mistroides* (Figure 16.1, **3**), and *Neospirorchis pricei* (Figure 16.1, **4**). These are the most pathogenic of the parasites in these turtles. A recent mortality of log-gerheads in Florida suggested that these flukes may have contributed to this die off (Jacobson et al., 2006). The massive infections in the brain are undoubtedly causing neurological damage, but the extent of the damage caused by less intense infections or infections in other sites is unknown (Stacy, 2012, personal communication). The adults sometime are numerous enough that they inhibit blood

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Wyneken, Jeanette. Marine Biology, Volume 14 : Biology of Sea Turtles, Volume III. : CRC Press, . p 457 http://site.ebrary.com/id/10657402?ppg=457 Copyright © CRC Press. . All rights reserved. May not be reproduced in any form without permission from the publisher, except fair uses permitted under U.S. or applicable copyright law. flow. Maybe more importantly, if these flukes are to pass their progeny onto new hosts, the eggs have to exit the host in some manner. So the eggs have to exit the vessels, often those in association with the gut, then pass through the intestinal wall to gain access to the lumen of the gut. They may cause extensive damage as they pass through normal tissue. Often eggs are found in most organs when the host has a number of blood flukes present. Some spirorchiids distribute their eggs close to the site of the adult and others embolize their eggs throughout the host. If they gain entrance into the gut lumen, they may pass along with the feces and exit the host. Many eggs are trapped in the tissues by host response. Finding adults of this family is difficult as they are not always washed out of the vessels, especially when they occur in small vessels. Therefore, the prevalence for these parasites here is underestimated. Stacy et al. (2010a) reported 96% of the loggerheads were infected with Neospirorchis spp., 78% with Hapalotrema spp., and 22% for Carettacola spp. From the data in Table 16.1, C. bipora is the most common blood fluke in loggerheads in Florida, but from related data species of *Neospirorchis* are more common than *C. bipora*. These blood flukes are usually found in low numbers, and whether this is a true reflection of their intensities or failure to locate the adults at necropsy is uncertain. Stacy et al. (2010a) demonstrated the normal sites for adult echoso H. mistroides are heart, aorta, and mesenteric arteries and H. pambanensis (not found in my survey) brary was the heart and aorta, whereas C. bipora is in hepatic vessels and N. pricei also uses the heart and associated great vessels.

Stacy et al. (2010a) have recorded infections of *Neospirorchis* in the thymus, endocrine organs, intestine, and brain. We are unsure of how flukes in vessels feeding the brain are capable of exiting the turtle. The data in this reference (resulting from the PhD dissertation of Dr. Brian Stacy) were collected in an incredibly meticulous manner producing these fantastic results and giving us an indepth understanding of these parasites. Because zoological species are based on morphology and because the adults could not be isolated from these other organs, these taxa have not been named. Stacy et al. (2010a) also discusses the pathology associated with these spirorchiids in great depth, and the reader is referred to this paper as the detail is beyond what will be discussed in the present chapter. For extensive bibliographies on spirorchiids (see Smith, 1973, 1997).

16.2.3 GASTROINTESTIONAL FLUKES (DIGENEA: BRACHYCOELIDAE, PACHYPSOLIDAE, PLAGIORCHIIDAE, PRONOCEPHALIDAE, STYPHLOTREMATIDAE, AND TELORCHIIDAE)

Most digenetic flukes live in the gastrointestinal tract. Their distribution begins in the esophagus with *Pachypsolus irroratus* (Figure 16.1, **6**) and *Diaschistorchis pandus* (Figure 16.1, **8**) and range through to the lower intestine where species of *Pyelosomum* reside (Table 16.1). The stomach is home to *Enodiotrema megachondrus* and the two species found in the esophagus. The intestine is divided by distribution of fluke habitats as the upper intestine houses *Cymatocarpus undulatus* (Figure 16.1, **5**), *E. carettae*, *R. gelatinosus* (Figure 16.1, **9**), *Orchidasma amphiorchis* (Figure 16.1, **10**), and *S. solitaria*, the middle region has *Pleurogonius trigonocephala* (Figure 16.1, **7**) and *Pyelosoma renicapite*, and the lower gut also has *P. renicapite* and *P. chelonei* (Table 16.1). The highest numbers and diversity of fluke are present in the upper intestine and this may reflect the quality and/or quantity of nutrients there before the turtle removes them to nourish itself. *C. undulatus*, *O. amphiorchis*, and *P. trigonocephala* were the more numerous and more common flukes found in loggerheads (Table 16.1).

16.2.4 LIVER FLUKES (DIGENEA: CALYCOIDIDAE)

One species resides in the liver and gall bladder and that is Calycoides anthus (Figure 16.1, 10).

16.2.5 URINARY BLADDER FLUKES (DIGENEA: GORGODERIDAE)

Only one species, P. cymbiformis (Figure 16.1, 12), lives in the lumen of the urinary bladder.

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16.2.6 LARVAL TAPEWORMS (CESTODA: TRYPANORHYNCHA)

The only tapeworms detected were larval forms of the Trypanorhyncha. We have not identified these to even genus, so we are unsure of how many species might be present in these turtles.

16.2.7 NEMATODES (NEMATODA: CUCULIDAE, KATHLANIDAE, ANISAKIDAE)

The nematodes, like the flukes, have evolved to use different habitats. *Sulcascaris sulcata* resides in the stomach, *C. carettae* in the upper intestine, *Kathlania leptura* in the middle gut, and *Tonaudia tonaudia* in the lower intestine. *S. sulcata* is the largest of these and is attached to the mucosa and may cause ulcers, but most specimens will be found unattached in necropsies as the worms will detach after the host is dead. This species has been found in post-hatchling loggerheads and may have a greater potential to cause damage due to the large size of the worm in a small host (Stacy, 2012, personal communication). This species uses a variety of bivalves and snails as intermediate hosts (Lester et al., 1980; Lichtenfels et al., 1980; Berry and Cannon, 1981). The other intestinal inhabiting nematodes have direct cycles. Although I did not find lungworms in our hosts (maybe cb50) due to their fragility, tiny size, and condition of the carcasses), *Angiostoma carettae* was detected in living loggerheads from Florida (Bursey and Manire, 2006; Manire et al., 2008). I did not find larval *Anisakis* in the stomach walls of our turtles, but they have been reported and were recently identified by molecular methods as *A. pegreffi* (Santoro et al., 2010b).

16.2.8 COMPARISON WITH OTHER HELMINTH SURVEYS

More studies on loggerheads have been done in the Mediterranean Sea than elsewhere. Five studies from that region examined a total of over 300 turtles and found a relatively depauperate helminth community. These references are for loggerheads in Italy, Spain, and Portugal (Manfredi et al., 1998; Aznar et al., 1998; Piccolo and Manfredi, 2002; Valente et al., 2009; Santoro et al., 2010a). While many studies list parasites found in a single turtle or name a new parasite, these have reported on the prevalence and intensity or abundance of helminths in loggerheads. Collectively they recovered 10 species of digenetic flukes, 4 species of nematode, 2 larval acanthocephalans, and 1 post larval tapeworm. E. megachondrus was the most common and most widely distributed fluke and it had the highest intensities. It was followed by R. gelatinosus and C. anthos. The remaining flukes were more restricted in geographical distribution and had lower prevalence and intensities and these 8d706were O. amphiorchis, P. trigonocephalus, P. cymbiformis, P. renicapite, P. irroratus, S. solitaria, ebrar and Adenogaster serialis. Nematodes included S. sulcata, C. carettae, K. leptura, and Anisakis sp. Sey (1977) examined loggerheads from Egypt and added only D. pandus to the known helminths from Mediterranean marine turtles. The prevalence of R. gelatinosus was the only species that was comparable with those in Florida turtles and the prevalence of the remaining species were usually less than half of that determined in Florida. All of these species were recovered from Florida loggerheads, with the exception of A. serialis and Anisakis. In all cases the intensity of infections was much lower in the Mediterranean as compared to Florida. Twelve juvenile loggerheads were examined in Brazil (Werneck et al., 2008c). They recovered 5 species including O. amphiorchis in 4 loggerheads with a range of 1-335 adults per turtle, 12 specimens of P. renicapite in 1 host, a single C. anthos in 1, S. sulcata in 2 with 15-18 adults and K. leptura in 2 hosts and 51-406 worms present. Chen et al. (2012) examined two loggerheads in Taiwan for spirorchiids and found none present.

16.2.9 PROTOZOA (EUCOCCIDIORIDA: EIMERIDAE)

A single coccidian species, *Eimeria carettae*, has been described from loggerheads. This is a typical appearing oocyst of *Eimeria* containing four sporocysts each with two sporozoites, except that there are thin filaments originating from the Stieda bodies on the sporocysts (Upton et al., 1990). Nothing more has been published on this parasite since it was described.

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16.3 GREEN TURTLE (CHELONIA MYDAS) PARASITES

Twenty-eight digenetic trematodes and larval cestodes were removed from 74 green turtles (Table 16.2). No helminths were recovered from 6 green turtles. An average of 6.2 and a range of 1–16 fluke species were recovered per infected turtle. The intensity of infections ranged from 2 to 15,524 for individual species. No adult nematodes were found in green turtles, but larval forms have been seen in histological sections from green turtles from Florida (Stacy, 2012, personal communication). Protozoa include two coccodia, *Caryospora cheloniae* and *Cryptosporidium* sp., unidentified species of *Octomitus* (flagellate) and *Entamoeba* (ameba) have been isolated as has a species of *Balantidium*. More papers have been published on green turtles parasites than any of the other marine turtles, and it is beyond the scope of this chapter to deal with that wealth of information.

16.3.1 BLOOD FLUKES (DIGENEA: SPIRORCHIIDAE)

Two species of blood-inhabiting flukes were found in green turtles and these were *L. learedi* (Figure 16.1, **13**) and a species of *Neospirorchis* both from the heart and major associated vessels. This family was discussed in the section on loggerheads and most of the same information applies here. *L. learedi* is the only spirorchiid for which we have an indication of the intermediate host based on molecular methods and it is the knobby keyhole limpit (*Fissurella nodosa*) (Stacy et al., 2010b). The pathological changes observed in greens associated with spirorchiids were also reported by (Stacy et al., 2010a). Again, there is a specialization in *Neospirorchis* species by organ selection as there was in loggerheads, and there are new species in vessels of the brain, thyroid gland, and gastrointestinal tract (Stacy et al., 2010a).

16.3.2 GASTROINTESTINAL FLUKES (DIGENEA: BRACHYCOELIDAE, PLAGIORCHIIDAE, PRONOCEPHALIDAE, RHYTIDODIDAE, TELORCHIIDAE, MICROSCAPHIDIIDAE, AND CLADORCHIIDAE)

Greens have a more diverse fluke fauna than found in loggerheads. The gastrointestinal tract had the most diverse parasite fauna as was the case with the loggerheads. Their distribution (Table 16.2) within the host varied from the esophagus (*D. pandus* and *Desmogonius desmogonius*) to the lower intestine *E. megachondrus* (Figure 16.1, 14), *Pleurogonius lobatus*, and *Schizamphistomum sceloporum*. Flukes from the stomach included *D. pandus*, *D. desmogonius*, *Charaxicephalus robustus*, and *Cricocephalus albus* (Figure 16.1, 15). Upper intestine flukes were *C. undulatus*, *Metacetabulum invaginatum*, *R. gelatinosus*, and *Microscaphidium reticulare* (Figure 16.1, 18). The middle intestine community comprised *P. trigonocephalus*, *P. signhi*, *P. longisculus*, *Deuterobarus proteus* (Figure 16.1, 16), *Pronocephalus obliquus* (Figure 16.1, 17), *Angiodictyum parallelum* (Figure 16.1, 19), *Neoctangium tranvassoi*, *Octangium sagitta* (Figure 16.1, 20), *Polyangium linguatula*, and *Schizamphistomoides spinulosus* (Figure 16.1, 22). The lower gut inhabitants were *E. megachondrus*, *P. lobatus*, *D. proteus*, and *S. sceloporum*. The middle intestine had the highest diversity of fluke species. *O. sagitta* was the most numerous species followed by *M. reticulare* and *D. proteus*, all of which are in the Microscaphidiidae.

16.3.3 LIVER FLUKES (DIGENEA: RHYTIDODIDAE AND PLAGIORCHIIDAE)

E. carettae and Rhytidoides similis (Figure 16.1, 21) lived in the liver (bile ducts) and gall bladder.

16.3.4 URINARY BLADDER FLUKES (DIGENEA: GORGODERIDAE)

P. cymbiformis was the sole helminth in the urinary bladder lumen.

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16.3.5 COCCIDIANS (EUCOCCIDIORIDA: EIMERIDAE, CRYPTOSPORIDAE)

A species of *Caryospora* resides in greens. It has eight sporozoites, which were originally thought to be in a single sporocyst within an oocyst which defines *Caryospora*, but it may be in a sporocyst without an oocyst. *C. cheloniae* was named from mariculture-reared green turtle hatchlings at the Turtle Farm in Grand Cayman (Leibovitz et al., 1978). They indicated it was pathogenic and included some descriptions of the damage done. More recently, an epizootic caused by this coccidian in free-ranging adult green turtles off the coast of Australia was published (Gordon et al., 1993). They necropsied 24 of the 70 that died within a 6 week time span. This coccidian was present in the intestinal as well as non-intestinal sites and it caused encephalitis well as enteritis. Much more is known from the dissertation of Dr. Anita Gordon (1995) in which she has demonstrated developing stages in small intestine, stomach, large intestine, urinary bladder, brain, thyroid gland, kidney, and adrenal gland. Her evidence suggests this is a serious pathogen of greens in the wild and we have detected oocysts in a few green turtles from Florida, but know nothing about its pathogenicity here.

Cryptosporidium oocysts have been reported from greens in Hawaii, but numerous loggerhead and green turtles histologic sections from Florida did not reveal evidence of this parasite (Grazyk et al., 1997; Stacy, 2012, personal communication).

16.3.6 CILIATES (VESTIBULOFERIDA; BALANTIDIIDAE)

The ciliate is *Balantidium bacteriophorus* and it was found in the stomach of a green turtle collected off the shore of Nicaragua (Fenchel, 1980). Nothing further has been published on this ciliate. It is presumably present in greens and loggerheads in Florida, based upon the cyst-like structures found in feces in the current study.

16.3.7 FLAGELLATES (DIPLOMONADIDA: HEXAMITIDAE, TRICHOMONADIDA: TRICHOMONADIDAE)

Octomitus sp. and a trichomonad whose identification was not taken to genus exist in greens (Fenchel, 1980). A brief description and a line drawing of the *Octomitus* are the extent of our knowledge of these flagellates.

16.3.8 Amoebae (Entamoebidae)

Entamoeba sp. was recorded from green turtles. This parasite is poorly characterized and the one citation suggests it might kill turtles (Frank et al., 1976). It has been seen in histological sections from both greens and loggerheads in Florida (Stacy, 2012 personal communication).

16.3.9 COMPARISON WITH OTHER HELMINTH SURVEYS

Green turtle populations have been examined for parasites in Egypt (Sey, 1977), mariculture-reared greens in Grand Cayman (Greiner et al., 1980), Hawaii (Dailey et al., 1992), and nesting greens in Costa Rica (Santoro et al., 2006). The Mediterranean green turtles were infected with 8 species of digenetic flukes and the highest prevalence was 42.8 (3/7) which was detected for *M. reticulare*, *O. sagitta*, and *P. linguatula*. The remainders were in 28% or fewer, namely *C. robustus*, *D. proteus*, *A. parallelum*, *Microscaphidium aberrans*, and *Cricocephalus resectus* and the intensity of infections were much lower than those found in Florida. That study only examined the gastrointestinal tract, gall bladder, lungs, and urinary bladder. The same organs plus the heart of greens were examined in Hawaii. Seven digenetic flukes were recovered and those were *L. learedi*, *Carettacola hawaiiensis*, two species of *Haplalotrema*, *P. linguatula*, *Angiodyctium longum*, and *Pyelosoma cochlear*. The spirorchiid fauna was more diverse than in Florida in the sample used in this chapter, but other data of Stacy et al. (2010a) erase that difference. The intensity of infections in Hawaii was less than those

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Wyneken, Jeanette. Marine Biology, Volume 14 : Biology of Sea Turtles, Volume III. : CRC Press, . p 461 http://site.ebrary.com/id/10657402?ppg=461 Copyright © CRC Press. . All rights reserved. May not be reproduced in any form without permission from the publisher, except fair uses permitted under U.S. or applicable copyright law. in Florida as well. Only two digenes were collected from the mariculture-reared greens on Grand Cayman and they were L. learedi in 40% of the greens with an intensity of 1-49 with a mean of 14 per infected turtle. A single specimen of *Pleurogonius mehrai* was removed from the upper intestine. The most recent study was in Costa Rica and that study had a sample of 40 nesting females and is a nice example of how such studies should be done. Twenty-nine species of digenetic flukes were recovered including 4 spirorchiids (L. learedi, Hapalotrema postorchis, Monticellius indicum, and Amphiorchis solus*), 5 angiodictyids (M. reticulare, P. linguatula, Deuterobarus intestinalis, Octangium hyphalum,* and Microscaphidium warui,* 1 clinostomid (Clinostomum complanatum*), 2 cladorchiids (S. sceloporum and Schistosomoides erratum), 2 rhytidodids (R. similis and Rhytidoides intestinalis), and 15 pronocephalids (P. cochlear, C. resectus, C. magastomus, C. albus, P. obliquus, D. desmogonius, Pleurogonius linearis,* P. longisculus, P. singhi, P. lobatus,* P. solidus,* Pleurogonius sp.,* Charaxicephaloides sp., C. robustus, and Rameshwarotrema uterocrescens). Eight species* of these were found in only one or two turtles with only one or two specimens in each, and only two of these minor flukes were identified from Florida greens and they were P. lobatus and P. linearis, which also were detected in very low prevalence. Costa Rican data indicated that species with the highest echoso intensities were similar to those in Florida as M. recticulare was their highest followed by D. intestinalisabrary (see Table 16.2 for comparison). It is possible that the two studies identified their flukes differently in some instances and that will have to be sorted out with future research. Typically greens from Florida had higher intensities when the same taxon was in both locations. A major difference between the two studies was that the Costa Rica turtles were all nesting females and the Florida specimens ranged in size from 23 cm straight carapace length to mature adults (unfortunately size data for a number of Florida turtles were lost). Four other recent papers examined blood flukes in green turtles in different locations. Three studies in Brazil found a single A. solus (Werneck et al., 2011), M. indicum was found in two juvenile greens (Werneck et al., 2008a), and in the final study 11 juveniles were examined and 6 were infected with L. learedi (Werneck et al., 2006). A study in Taiwan recovered L. learedi in 11/13 turtles, H. mehrai in 7/13, H. postorchis in 6/13, and C. hawaiiensis in 2/13 (Chen et al., 2012).

16.4 HAWKSBILL (ERETMOCHELYS IMBRICATA) PARASITES

Relatively few studies have been conducted on the parasites of the hawksbills. Only four hawksbills were examined in my laboratory culminating in a fauna of six species of digenetic trematodes and trypanorhynch larvae. An average of three species (range of two to four) per infected turtle was detected. The mean intensity ranged from 3 to 16 flukes (Table 16.3). This species primarily eats sponges, which would limit the potential for ingesting molluskan hosts serving as intermediate hosts to helminths. Thus the low number of parasites and low diversity of parasites might reflect this behavior. No nematodes or protozoa have been reported from this species.

16.4.1 BLOOD FLUKES (DIGENEA: SPIRORCHIIDAE)

One hawksbill was infected with a species of *Neospirorchis* in the cerebral vasculature (Table 16.3). Nine adults were observed, but it is impossible to extract them from these vessels due to the fact that the diameter of the adult fluke approximates the lumen diameter of the infected vessels and thus species identification was not possible. Whether this is the same undescribed species in the cranial vessels of the green turtle is unknown.

16.4.2 GASTROINTESTINAL FLUKES (DIGENEA: PLAGIORCHIIDAE, PRONOCEPHALIDAE, AND RHYTIDODIDAE

A single specimen of *E. carettae* was recovered from one individual. Two hawksbills were infected with *C. albus* and two to four flukes were in each and one *D. pandus* was in one turtle. *R. gelatinosus* was in three individuals and from 9 to 45 were present in each (Table 16.3).

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16.4.3 URINARY BLADDER FLUKES (DIGENEA: GORGODERIDAE)

A single species, *P. cymbiformis*, was present in the urinary bladder of one hawksbill and four adults were recovered.

16.4.4 COMPARISON WITH OTHER SURVEYS AND A LISTING OF SPECIES KNOWN FROM HAWKSBILLS

Because of the few hawksbills examined, the following is a list of helminths reported from this species. Fischthal and Acholonu (1976) have the most complete listing from hawksbills from their necropsies in Puerto Rico and which included seven new species, namely Amphiorchis caborojoensis, Epibathra stenobursata, Glyphicephalus latus, Pleurogonius laterouterus, P. puertoricensis, Pachypsolus puertoricensis, and Calycodes caborojoensis and 21 previously reported species which were L. orientalis, Hapalotrema spirorchis, Amphiorchis amphiorchis, M. reticulare, O. sagitta, O. travassosi, S. sceloporum, P. cymbiformis, C. albus, C. magastomus, P. lin-act 50 earis, P. trigonocephalus, P. lobatus, Pyelosoma posteriorchis, D. pandus, M. invaginatum, brary Pachypsolus ovalis, Enodiotrema reductum, S. solitaria, O. amphiorchis, and R. gelatinosus. Most of their intensities were less than five worms per infected turtle and D. pandus was the most prevalent species followed by O. sagitta and O. tranvassosi, which also had the highest intensities. More recent reports from hawksbills from Puerto Rico provided Angiodictyum mooreae, A. anteroporum, and A. parallelum (Dyer et al., 1995a), and an extensive checklist of hawksbill helminths was provided by Dyer et al. (1995c). Chattopadhyaya (1972) added Octangium microrchis and Pyelsomum solum, both from the lower intestine. Two more species from Australia were Pleurogonius truncatus and Pyelosomum parvum, both new species (Prudhoe, 1944). Two other new species from Bermuda were Diaschistorchis megas and Pachypsolus brachus (Barker, 1922). The only report of Neospirorchis sp. and E. carettae is from the Florida sample. Five Amphiorchis cabarojoensis and two Carettacola stunkardi were recovered from a single Hawksbills from Brazil (Werneck et al., 2008b).

16.5 LEATHERBACK (DERMOCHELYS CORIACEA) PARASITES

Two Florida leatherback turtles were examined and one of those was still partially frozen. The only **8d70** parasites from it were three *Pyelosomum renicapite* from the intestine. These specimens were about **ebrar** 2.5–3 cm in length, the largest flukes I have encountered in marine turtles. A fresh leatherback was killed by a trawler and it provided a better look at the parasites present. This resulted in *P. renicapite*, *C. undulates, Enodiotrema instar, C. anthos*, and larval tapeworms.

16.5.1 COMPARISON WITH OTHER SURVEYS AND A LISTING OF SPECIES KNOWN FROM LEATHERBACKS

A north Atlantic leatherback examination occurred in Newfoundland where two leatherbacks were killed. Three digenetic flukes were recovered and they were *P. renicapite*, *C. anthos*, and *Cymatocarpus* sp. (Threlfall, 1979). An examination of one leatherback in Puerto Rico yielded 25 *P. renicapite* from the intestine (Dyer et al., 1995b) and two of the same species were removed from a leatherback in France (Almore et al., 1989). A Mediterranean leatherback yielded 44 *P. renicapite*, and 60 *E. instar* from the stomach and 71 *E. carettae* from the liver (Manfredi et al., 1996); and a second leatherback collected a few years later had the same three flukes (Piccolo and Manfredi, 1999). Four of eight leatherbacks examined from Brazil and Uruguay were infected with *P. renicapite*, and the one from Uruguay contained a single *C. anthos* (Werneck et al., 2012).

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16.6 KEMP'S RIDLEY (LEPTOCHELYS KEMPII) PARASITES

Four Kemp's Ridleys were examined in our study. They yielded eight species of digenetic trematodes and two species of nematodes and trypanorhynch larvae (Table 16.4). Each was infected with a mean of 6 species with a range of 2–10. Mean intensities ranged from 1–5904 and no more than 2 of the 4 were infected with the same species of parasite (Table 16.4). *C. undulatus* had the highest intensity, with one turtle having nearly 12,000 in its intestine. This turtle species shared its entire parasite fauna with loggerheads, but the fauna was more restricted than in the loggerheads (see Tables 16.1 and 16.4). One study examined cold-stunned Kemp's Ridleys in Massachusetts; and while they examined tissues histologically, they only identified a trypanorhynch post-larva as presumptively identified as *Tentacularia coryphaenae*, but did not attempt other identifications (Innis et al., 2009). No other reports were found on the parasites of this turtle species.

16.7 OLIVE RIDLEY (LEPIDOCHELYS OLIVACEA) PARASITES

We did not examine any olive ridleys as they rarely occur in Florida. However, two published reports present an indication of their parasite fauna. One study in Mexico examined the gastrointestinal tracts of 32 olive ridleys and recovered 8 digenetic flukes and 8 turtles were not infected. These were *A. serialis*, *P. lobatus*, *P. renicapite*, *P. irroratus*, *E. megachondrus*, *O. amphiorchis*, *C. anthos*, and *Prosorchis psenopis*. All of these except the last one are shared with other marine turtles. *E. megachondrus* had the highest abundance and had the second highest prevalence, following *A. serialis* (Perez Ponce de Leon, 1996). A study of 3 olive ridleys in Costa Rica recovered 3 *E. megachondrus* in the upper intestine from 1 turtle, 1 *P. irroratus* from the stomach and 31 *P. cymbiformis* from the urinary bladder of another host (Santoro and Morales, 2007). Nothing more is known on the parasites of this marine turtle.

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