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Sirenian Pathology and Mortality Assessment

ROBERT K. BONDE, ANTONIO A. MIGNUCCI-GIANNONI, AND GREGORY D. BOSSART

Previous chapters in this book have discussed how rapid technological development in recent history has accelerated human impact on sirenians. Determining the causes of the numerous threats to manatees, especially those that result in mortality or disease (tables 17.1 and 17.2) will enable implementation of appropriate protection measures. As a conservation tool, cause of death determination and interviews with the local population are vital steps for establishing successful research and conservation programs[1]. Such information can identify threats to the species and provides an opportunity to collect information on population distribution, diseases, parasites, diet, reproductive condition, genetics, and morphological anomalies.

Anthropogenic Causes of Mortality

Beyond being exploited for food[2], manatees and dugongs are faced with additional human-originated threats. In Florida many manatees are killed each year as a result of collisions with water vessels (figure 17.1). Twenty-five percent of Florida manatee mortalities are due to such collisions[3]. From 1990 to 2006, 19.8% of all known manatee deaths (n = 121) in Puerto Rico were identified to be due to boat strikes[4]. Most boat strikes are single events, affecting one individual. However, both in Puerto Rico and Florida, scientists have documented boat strikes of up to five manatees at one time, all assumed to be part of a mating herd when struck. Manatees in mating herds are preoccupied with breeding and may be especially vulnerable to collisions.

Most adult manatees in Florida, and some in Puerto Rico, bear scars as evidence of nonfatal encounters with boats, and sirenians with propeller scars are observed in other countries as well[5]. Many manatees do not appear to be permanently affected by moderate to light vessel strikes, but others suffer very serious injuries that may affect reproduction and migration or lead to death. In fatal cases the trauma can be acute or chronic in nature[6]. In acute cases, the injury is often massive, deep, or severe

Table 17.1. Determined cause of death for 68 fresh manatee carcasses from Florida by age/size and sex in decreasing order of frequency, January 1996 to January 2004.

		Adult		Subadult		Perinatal	
Cause of Death	Total	M	F	M	F	M	F
Trauma	32	7	6	12	7	.	.
Cold stress syndrome	12	1	.	4	7	.	.
Inflammatory/infectious disease	8	2	0	3	2	.	1
Suspected brevetoxicosis	6	2	1	2	1	.	.
Cachexia	4	.	.	.	.	2	2
Unknown	4	2	.	1	1	.	.
Intestinal foreign body	1	.	.	.	1	.	.
Cardiomyopathy	1	1	.	.	.	.	.
Total	68						

Source: Bossart et al. 2004, reprinted with permission.

Table 17.2. Predominant cause of stranding for 121 West Indian manatees recovered from Puerto Rico between 1990 and 2006 with comparison to previously tabulated data.

		Prior to 1990		Total		Total	
	Causes	Cases	%	Cases	%	Cases	%
Natural		6	10.9	53	43.8	59	33.5
	Dependent calf	6	10.9	28	23.1	34	19.3
	Illness	0	0	21	17.4	21	11.9
	Stillborn	0	0	3	2.5	3	0.6
	Predation	0	0	1	0.8	1	0.6
Human related		31	56.4	35	28.9	66	37.6
	Drowning	0	0	1	0.8	1	0.6
	Entanglement	1	1.8	1	0.8	2	1.2
	Gunshot	2	3.6	2	1.7	4	2.3
	Pollution	0	0	2	1.7	2	1.2
	Capture	22	40	5	4.1	27	15.3
	Watercraft	6	10.9	24	19.8	30	17
Undetermined		18	32.7	33	27.3	51	28.9
Total		55		121		176	

Source: Adapted and updated from Mignucci-Giannoni et al. 2000, with permission.

Figure 17.1. Fatally injured West Indian manatee in Florida with characteristic propeller lesions on dorsum. (Courtesy of U.S. Geological Survey.)

enough for the animal to succumb to shock or drowning, and death is rapid. In chronic cases some individuals develop bone lesions that can become systemic and eventually cause death[7].

Another form of human-related death in Florida is attributed to entrapment in the gates of canal locks or flood control structures resulting in crushing or drowning[8]. Poor nutritional condition in manatees can also be anthropogenic in nature, as it is often related to habitat loss and possibly high contaminant exposure levels[9].

Entanglement in fishing gear or debris is usually not life threatening but can lead to severe tissue damage and sometimes self-amputation of one or both flippers as blood flow is restricted. Entanglement can involve several types of abandoned fishing gear, but commonly, monofilament fishing lines and thicker nylon ropes are implicated. Sirenians can be bound or restricted when the lines become wrapped around their flippers or body. If detected early, the lesions often can be treated by removal of the constricting material and debridement of necrotic tissue. Conservation education can inform fishers of the potential damage caused by reckless discarding of fishing gear. Ingestion of debris, hunting, and vandalism are also of major concern in sirenian conservation throughout the world[10].

Trauma

Categories of traumatic injuries are based on historic criteria established in Florida[11] and may be a result of human-inflicted injury, including sharp penetrating trauma from boat propellers or blunt trauma from the impact of a boat hull or outdrive skeg (figure 17.2). It also may include penetrating wounds produced by firearms, spear guns, or harpoons. Based on gross and microscopic findings, death by trauma can be further subdivided into the categories of acute (i.e., injury considered severe enough to have caused death immediately or within a few hours with no pathologic evidence of preexisting disease) and chronic (i.e., injury that was not immediately fatal but resulted in secondary inflammatory disease and death).

Most often, trauma is associated with internal wounds, hemorrhage, blood clots, and severe damage to tissues and organ systems. Care should be taken to assess and determine whether the trauma in question is post- or antemortem in nature. Samples should be collected and submitted for histopathological analyses. Trauma from shark, alligator, or crocodile predation would fall into the natural category but should be distinguished from postmortem scavenging of a carcass that may have died of other causes.

Drowning

By itself, drowning does not have consistent clinical signs, and often there may not be any associated pathology to identify drowning as the specific cause of death. Drowning is therefore usually a diagnosis of exclusion. Just prior to death, agonal (labored, irregular) breathing

Figure 17.2. Injured West Indian manatee in Florida with severe propeller lesions on tail stock. (Courtesy of Kit Curtin.)

is present and aspiration of foreign bodies (e.g., mud, sand, plant material) may be apparent. Drowning in a marine mammal is often secondary to some other process or etiology.

Toxins

Sirenian mortality may be associated with toxins and chemical pollution from biotoxins, pharmaceuticals, and industrial discharge or spills[12]. Suspected cases of anthropogenic toxins impacting sirenians have been documented[13], but to date no direct related morbidity or mortality has been verified. Some suspect cases were reported during the Iran/Iraq War in the Persian Gulf during the 1980s, where dugongs were exposed to large quantities of spilled petroleum products[14] that may have contributed to their death. During this die-off there were also rumors of potentially fatal algal blooms in the area that may have contributed to the dugong mortality. Under these circumstances, the examiner can directly detect the caustic substance on the animal, but cause and effect in a fatality may be very difficult to prove[15]. In subtle cases, where small quantities of these products might have been ingested, a necropsy is needed to examine stomach contents, and tissue samples should be collected for contaminant analyses. Other pollutants directly affecting sirenians may include pesticides and herbicides. Impacts from exposure to these toxins could affect changes in reproductive, metabolic, and immune functions without killing the animal and may be more detrimental to overall population health and recovery. Causes for these biological stressors are difficult to identify, and in cases where toxins are suspected, blood, brain, liver, kidney, muscle, and blubber samples should be taken and frozen in chemically clean glass containers with Teflon lids (available from scientific supply houses; clean aluminum foil can be used for organic contaminants if such containers are unavailable). These samples should then be submitted to an appropriate toxicology laboratory for analyses.

Natural Causes of Mortality

Disease

Based on clinical findings, West Indian manatees appear to be remarkably resistant to disease and the sublethal effects of traumatic injury[16]. Their disease-resistant traits may result partially from a highly efficient and responsive immune system[17], which in some cases may take the manatee to a weakened state, even to the extent of cachexia (severe malnutrition), over a long period of time (figure 17.3). Notable exceptions to non-traumatic deaths in manatees from Florida are found with mortality associated with prolonged cold-water exposure and the inhalation and/or ingestion of red tide toxins (see discussion to follow)[18]. However, the general pathologic aspects of disease associated with mortality in other manatee species and the dugong have not been well characterized.

Verminous and bacterial pneumonia and enteritis are the most common diseases found in manatees from Florida and Puerto Rico[19]. Pneumatosis intestinalis (gas-filled areas in the bowel wall of the gastrointestinal tract) has been detected in manatee calves from both Florida and Puerto Rico[20].

Over the last 15 years field biologists have observed papillomatous lesions on some captive manatees[21] and papilloma-suspect dermal lesions on wild manatees in northwest Florida[22]. These lesions were not present decades ago during detailed surveillance of manatees in Crystal River as part of the photo-identification program[23]; research efforts are under way to ascertain whether they are the result of environmental and/or physical changes or stressors in the habitat or due to a compromise of the manatee's immune system function.

Preliminary studies suggest that unlike manatees from Florida, West Indian manatees throughout the rest of their range do not have pathologic findings consistent with either cold stress syndrome (CSS) or brevetoxicosis. This is not surprising as the prolonged exposure to low water temperatures necessary to induce CSS or the frequent red tide blooms consisting of the brevetoxin-producing dinoflagellates are rarely found in the Antillean manatees' habitat.

Recently it has been postulated that sirenians may be good sentinels for ocean and human health[24]. In this context, the characterization of sirenian mortality factors could provide early warning indicators of stressors, contaminants, or habitat changes in the aquatic environment. Disease characterization among sirenians would potentially permit us to manage related impacts on human and animal health associated with our coastal ecosystems. Thus as we indentify environmental degradation affecting sirenian health, we may also be identifying factors affecting human health.

Harmful Algal Blooms

Recent epizootics (animal epidemics) have been associated with potent marine neurotoxins known as brevetoxins, which are produced in Florida by the "red tide" dinoflagellate *Karenia brevis*[25]. Brevetoxins are known to kill large numbers of fish and to cause illness in humans who ingest filter-feeding shellfish contaminated with the brevetoxin (neurotoxic shellfish poisoning) or who

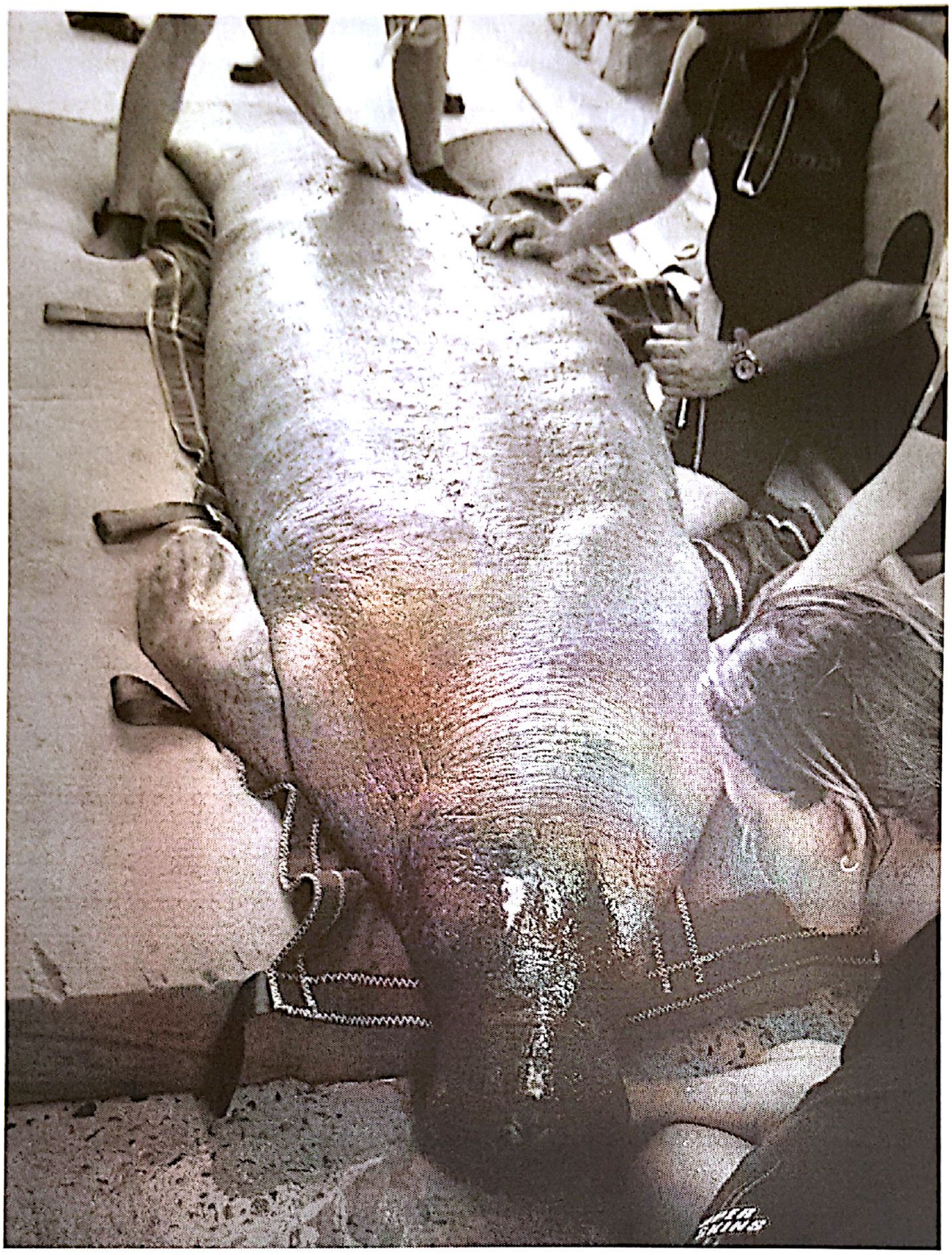

Figure 17.3. Chronically sick West Indian manatee rescued in the British Virgin Islands in a severe state of cachexia and debilitation. (Courtesy of Puerto Rico Manatee Conservation Center.)

inhale the toxic aerosols. The pathogenesis of manatee brevetoxicosis is suspected to involve direct inhalation and/or ingestion of toxins from the environment. Important new data indicate that brevetoxin vectors such as epiphytes on seagrasses or incidentally ingested invertebrates can result in delayed or remote manatee exposure causing intoxication in the absence of active toxin-producing dinoflagellates[26]. Thus, unexpected toxin vectors may account for manatee deaths long after, or remote from, a dinoflagellate bloom.

Diagnosis of brevetoxicosis in manatees is typically by exclusion and may be based on pathologic findings and postmortem demonstration of the toxins in fluids and tissues. Gross findings of inhalational brevetoxicosis in manatees include irritation of the upper respiratory tract and excessive congestion and edema in the lungs. Present data suggest that manatee mortality resulting from brevetoxicosis may not necessarily be acute but rather occurs after chronic inhalation and/or ingestion[27]. The inhalational route of brevetoxin exposure appears to be unique among marine mammals and humans. Increases in human pulmonary emergency room diagnoses often are temporally related to "red tide" occurrences, which may be increasing in frequency along the Florida coastline[28].

Neurotoxic signs of clinical brevetoxicosis include seizure, disorientation, ataxia, hyperflexion, muscle fasciculations, flaccid paralysis, and dyspnea. Manatees with brevetoxicosis may be treated symptomatically with corticosteroids and nonsteroidal anti-inflammatory drugs. Supportive care includes removal from toxic waters, providing fluids, nutritional supplementation, and providing water buoyancy devices to prevent submersion and subsequent drowning. It is essential that all findings be discussed with an experienced veterinarian and pathologist.

Cold Stress Syndrome

Chronic exposure to cold water produces a cascade of clinical signs and disease processes termed manatee cold stress syndrome (CSS)[29]. All age and sex categories are affected by CSS; however fewer cases are detected among the perinatal age category. Additionally, large adults tend to be affected by CSS less frequently than small adults and subadults, owing to their greater experience at utilizing known warm water sites, their increased competence at heeding the signs of approaching cold weather, or the ability to decrease body heat loss due to a reduced surface area to volume ratio. Pathologic features of CSS include emaciation, fat store depletion, serous fat atrophy, lymphoid depletion, epidermal hyperplasia, pustular dermatitis, enterocolitis, and degeneration of heart muscle. The data indicate that CSS is a complex disease process that involves compromise to metabolic, nutritional, and immunologic balances and culminates in secondary diseases. The pathogenesis of manatee CSS appears to involve a series of events that initially involves chronic exposure to water temperatures well below 20°C. This exposure, in turn, results in lethargy, decreased food intake, dehydration, and constipation, which further compromise nutritional, metabolic, and immune balances. The latter would ultimately result in additional immunologic compromise, predisposing CSS manatees to opportunistic infections and other unusual lesions that may have infectious, nutritional, and/or metabolic components. Therefore, treatment for rescued manatees with CSS should address these specific factors with special emphasis on reestablishing normal gastrointestinal function and fluid/nutritional status.

The long-term sublethal effects of CSS may be more insidious and involve increased susceptibility to disease and impacts on fecundity and calf survival. The preliminary evidence of the combined suppressive immunologic effects of CSS and exposure to red tide toxins needs further investigation. Additionally, understanding the pathophysiologic mechanisms of manatee CSS is becoming critically important for developing future management strategies. The availability of a network of warm water refugia may be one of the most important and challenging long-term needs for manatees[30]. Most Florida manatees rely on natural or artificial warm water refuges to survive cold winters and typically return to the same refuges year after year. Over the past 30 years in Florida, warm water effluent sources at electric power plants have provided life-sustaining refugia for many manatees during the winter. These refugia become particularly significant as winter manatee counts at a single power plant warm water refuge may exceed 500 animals or approximately 15% of the remaining population[31]. Serious concern exists about the inevitable shutdown and/or deregulation of aging power plants on which manatees have become dependent during cold weather[32]. Because manatees are unable to adapt quickly to changes in the availability of warm water, elimination of these warm water refugia could have profound effects on increasing mortality and altering population distributions in this already vulnerable species[33].

Parasites

Heavy parasitic loads in manatees are common, but generally these loads are not of pathologic concern. Parasites have been studied in detail in manatees from Florida[34], the Dominican Republic[35], and Puerto Rico[36], and in dugongs[37]. The most common parasite in Florida manatees is the trematode *Chirochis fabaceus*, whereas *C. groschafti* is found in manatees from the Caribbean. Both are flukes inhabiting the lower gastrointestinal tract. Another fluke common in Florida and Antillean manatees is *Pulmonicola cochleotrema*, located in the nasal passages, which has been implicated in rare cases of verminous pneumonia[38]. An ascarid nematode, *Heterochelius tunicatus*, is often present in the mucosa of the stomach and duodenum or free within the lumen[39]. Additionally, manatees in Florida, Belize, and Puerto Rico may have up to five species of barnacles on their dermis[40], and their skin is often encrusted with algae and other epiphytes. Recently a tanaid crustacean, *Hexapleomera robusta*, was discovered imbedded in the skin of manatees in Mexico[41] and Belize[42] (figure 17.4), and a new species of nematode has been identified, yet undescribed, in the urine of manatees from Belize[43]. A copepod, *Harpacticus pulex*, was isolated on the skin of a captive manatee in Florida[44] and recently from wild captured manatees from Puerto Rico and Mexico[45]. In dugongs, one species of protozoan, a nematode, and 19 trematode species[46] have been found. Most of these species only occur in the dugong and not other animals. Sharksucker species (*Echeneis naucrates* and *E. neucratoides*) are common associates with West Indian manatees and dugongs in marine environments[47].

Perinatal

The social structure of all sirenians entails a strong bond between the cow and calf, which can last as long as three years in some pairs. If for some reason a mother is separated from her offspring, the nutritionally and socially dependent calf usually dies, though there is some evidence that orphaned calves have been adopted by other lactating females. Calves can be separated from their

Figure 17.4. A tanaid (*Hexapleomera robusta*), an epibiont found embedded in the skin of a West Indian manatee from Belize. (Courtesy of U.S. Geological Survey.)

mothers for several reasons. The mother may have died, the two could have been separated by human or other curious manatee disturbance resulting in flight avoidance, the cows or calves may not be behaviorally compatible and were separated, or they could be genetically unfit and die in the natural culling process. From 1990 to 2006, 35.5% of the manatee carcasses recovered from Puerto Rico were calves[48]. Deaths in this group are generally attributed to cachexia and pathologic lesions consistent with prolonged protein-caloric deficiency[49]. The clinicopathologic features are similar to those often seen in orphaned manatees rescued and presented to rehabilitation facilities for medical care[50].

Quantitatively, this grouping of dependent calf deaths is deemed important and could have a direct impact on the growth rate of the overall population. Dependent calves that die generally do so in their first year. In Florida, these are manatees that are often shorter than 175 cm in total length. By process of elimination, the proximate cause of death in these cases is not likely to be human-related[51].

Predation

Sirenians are presumed to be opportunistically attacked and sometimes consumed by large predators, although documentation of this interaction is very rare. Dugongs are preyed upon by sharks, including tiger sharks (*Galeocerdo cuvier*), killer whales (*Orcinus orca*), probably by salt-water crocodiles (*Crocodylus porosus*), and hammerhead sharks (*Sphyrina* spp.)[52]. Amazonian manatees are said to be occasionally preyed upon by sharks, jaguars (*Panthera onca*), and caimans[53]. No natural predation on West African manatees has been documented[54], and it has been reported that the West Indian manatee has no natural predators[55]. However, Falcón-Matos et al.[56] documented the first case of an antemortem predatory attack on a manatee in Puerto Rico, probably by a large tiger shark. Recently, shark predation events have been recorded from Florida and Belize[57], and now West Indian manatees are suspected to be opportunistically preyed upon as well by the American crocodile (*Crocodylus acutus*) and American alligators (*Alligator mississippiensis*).

Stochastic Events

Stochastic or unpredictable events such as hurricanes, also can result in mortality[58]. During storms animals may be swept from their normal distributional range into inappropriate or unsuitable habitat. One such case has been documented where a manatee was displaced from its natural Florida habitat to waters around an island in the Bahamas, hundreds of kilometers away. Hurricane Georges off Puerto Rico in September 1998 was associated with a manatee mother and calf separation, resulting in the need to rescue the calf. In September 1996 a manatee calf death in Puerto Rico was directly related to Hurricane Hortense. At present, the extent of storm-induced mortality and the consequences of global warming affecting range dispersal on sirenian populations are not well understood.

Mortality Documentation

Recovery and History

If there is a report of a carcass, it should be examined as soon as possible. The examiner should obtain as much information as possible, including the location, circumstances, date, and time of events and the name and contact information for the initial observer or informer. Most marine mammals, and particularly sirenians, even dead ones, are protected by local and national laws and regulations. Make sure you are familiar with applicable laws and understand the implications of recovering a carcass. Obtain proper permits prior to conducting research on any marine mammal. When recovering a carcass, observe and note the weather and environmental conditions where the animal was found, as this may help in determining the cause of death. In some cases these factors may be associated with the demise of the animal (e.g., red tide, other dead wildlife, signs of water contamination, recent passage of a hurricane or storm, etc.). Take full body photographs, with appropriate reference scales, of each side of the animal and its ventral and dorsal surfaces before moving the carcass. Transfer the animal to the examination site, taking care not to damage the carcass, and note especially signs of disease or trauma that may be altered by the ropes, nets, stretchers, or cranes used during recovery. If the animal is transported, if possible apply bags of ice around the carcass to reduce the rate of decomposition until it can be examined. Carcasses which are going to be examined in the near future may be better if refrigerated rather than frozen. Freezing artifact or burn will damage cells and complicate the interpretation of tissues collected at necropsy.

Necropsy Examination

Although there is no set standard for necropsy examination of a sirenian carcass, some useful guides have been produced[59]. Experience and a thorough, consistent approach are important in a necropsy examination, especially when dealing with sirenians and their unusual morphological aquatic adaptations. Try to determine abnormal conditions even though this may be difficult if you are not familiar with normal sirenian or mammal anatomy and pathology. The initial examiner should take detailed notes and photographs, which, along with submitted tissues, can be examined later by a pathologist to attempt to provide a definitive cause of death. One should carefully examine the carcass

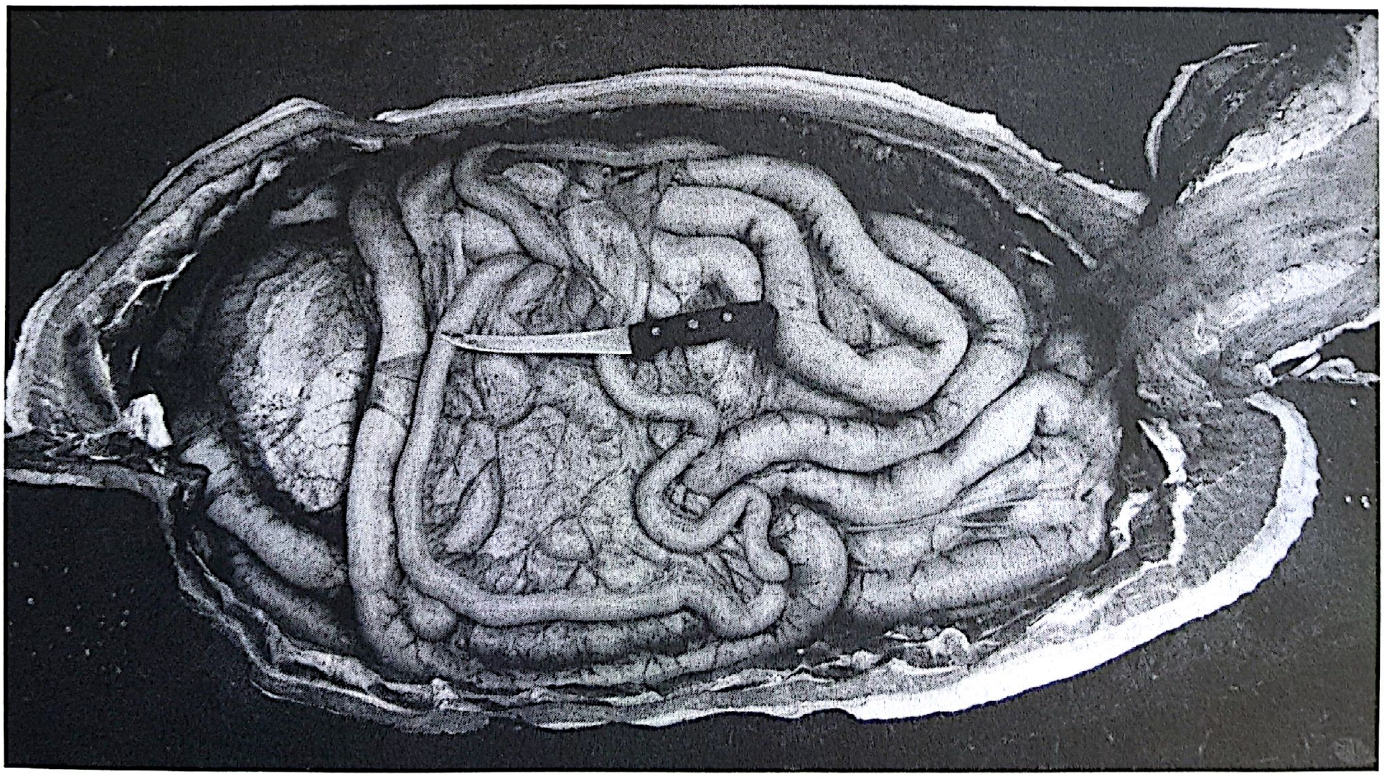

Figure 17.5. Necropsy of a West Indian manatee, showing dorsal recumbency position used for examination of organs in situ. (Courtesy of U.S. Geological Survey.)

for unusual marks, lesions, or trauma. Complete morphometrics, photographs, and video documentation should be taken as well. Place the carcass on its back so that it can be opened ventrally, exposing all organ systems to the examiner (figure 17.5). Tissue samples from each organ should be collected and properly preserved. For pathology purposes, a sample should be collected from each major organ (heart, lungs, liver, spleen, stomach, duodenum, small intestine, large intestine, testes or ovaries, kidneys, urinary bladder, lymph nodes, and brain) as well as from any suspect tissue. Samples should measure about 2 cm by 2 cm by 0.5 cm thick and should be preserved in 10% neutral buffered formalin. The volume ratio of tissue to preservative should be approximately 1:10. Note tissue color, size, shape, consistency, and smell. Be cautious when interpreting color and wetness of tissues since lividity can create artifactual changes by redistributing fluids. Search for, and collect in ≥70% ethanol, ectoparasites and epibionts (barnacles, copepods, algae, etc.) as well as endoparasites found in the airways, stomach, and intestines[60]. Collect a small piece of skin for genetic analyses and preserve it in a tissue buffered DMSO solution or ≥70% ethanol[61]. Practice proper sanitary hygiene and safety at all times throughout the examination[62].

Conclusions

Understanding the cause and effect of mortality processes is a first step in better understanding the population dynamics of the studied species. Mortality assessment programs have been important aspects of acquiring basic life-history information on the species and useful for monitoring conservation efforts in Florida, Puerto Rico, Mexico, Belize, Brazil, and Australia. Mitigation efforts have resulted in substantial reductions in sirenian mortality over the last few decades in certain portions of their range. In some areas, these conservation efforts to protect sirenians—such as net bans, boat access and speed restrictions, changes in flood gate operations, habitat protection, and establishment of sanctuaries—have already shown measurable success. The continuation of these diligent safeguards, coupled with monitoring of mortality to identify and detect potential threats to populations of sirenians, will help ensure persistence and population viability.