The Elasmobranch Husbandry Manual:

Captive Care of Sharks, Rays and their Relatives

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Chapter 26
An introduction to Viral, Bacterial, and Fungal Diseases of Elasmobranchs

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Abstract: Compared to some other aspects of elasmobranch biology, the study of infectious diseases in this group is in its infancy. Diagnostic sampling of sick or dead animals should be considered an essential aspect of any husbandry program. Viral diseases are characterized by rapid onset, disease in multiple individuals, and often a stressful event that causes immunosuppression. Viral diseases are among the most difficult diseases to diagnose. There are currently no recognized treatment or control methods specific to viral diseases. General recommendations include supportive care and reducing stress. Outbreaks of bacterial disease are relatively uncommon in elasmobranchs. Reports of bacterial disease caused by *Vibrio* spp. are the most common. In contrast to viral diseases, most bacterial diseases can be diagnosed by bacterial culture of blood or diseased tissues such as the brain, liver, kidney, or spleen. Bacterial culture and antibiotic sensitivity testing may help guide antibiotic choice for elasmobranchs. Fungal diseases are typically characterized by disease in an individual animal rather than explosive outbreaks affecting multiple animals. Fungal agents can cause disease in all age groups of animals (in contrast to bacteria and viruses that commonly infect young or old individuals). Treatment of fungi is difficult, and prevention is the best tool for management. The occurrence of fungal disease may be reduced by maintaining susceptible sharks in appropriate environments (i.e., appropriate water quality and temperature regimes) and by reducing stress associated with capture or handling. Skin wounds are a potential portal of entry for fungi, so skin wounds should be avoided or closely monitored when they do occur.

Compared to some other aspects of elasmobranch biology, the study of infectious diseases in this group is in its infancy. Reports of viral, bacterial, and fungal diseases are few in number and often involve case reports of disease in single animals. It is doubtful that this lack of data on infectious diseases of elasmobranchs reflects the true situation, but rather suggests a lack of data collection and diagnostic sampling by people studying the animals, and institutions holding or displaying them. Much of the information in this chapter is taken from the author's personal experience, communication with colleagues, and most importantly from the book *Fish Medicine* by Stoskopf (1993). In addition, an attempt has been made by the author to sample the scientific literature available on this topic for background on various organisms and the diseases they cause.

This chapter will provide information on basic disease biology, brief descriptions of various organisms reported from elasmobranch species, descriptions of disease syndromes, and methods of diagnosis and sampling for animals suspected to have viral, bacterial, or fungal disease.

Diagnostic sampling of sick or dead animals should be considered an essential aspect of any husbandry program. It is only by attempts to cultivate and identify microorganisms that medical and biology professionals can gather baseline data. Furthermore, I encourage professionals studying elasmobranchs to publish findings concerning infectious disease in peer reviewed scientific journals so that information can be disseminated. There exists an abundance of experience and knowledge that cannot be shared without publication.

A more complete discussion of medications successfully used with elasmobranchs, and the diseases treated, is provided in Chapter 29 of this manual.

VIRUSES AND VIRAL DISEASES

Viruses are among the smallest infectious agents and often are responsible for explosive outbreaks of disease in both human and animal populations, including fishes. Several viral diseases of teleosts are well characterized as causes of significant disease outbreaks. In general, most viral diseases are characterized by rapid onset, disease in multiple individuals, and often a stressful event that causes immunosuppression. Viral diseases are among the most difficult diseases to diagnose. Cultivation and identification of viruses require specialized techniques and equipment. In general, viral diseases are not treatable with conventional medications or antibiotics and it is up to the immune system of the infected individual to "fight" the virus so that healing and recovery may begin.

Viral diseases

Two different viral diseases have been reported in elasmobranchs, with most reports coming from the dusky smooth-hound (*Mustelus canis*). These diseases, although uncommon to rare and poorly studied, exemplify some of the common characteristics of viral disease.

A viral skin disease has been reported in the dusky smooth-hound, affecting a small percentage of animals. The disease is characterized by areas (~1-10 mm diameter) of white to gray skin discoloration (McAllister and Stoskopf, 1993; Leibovitz and Leibovitz, 1985). There are no recognized systemic effects in infected animals. A herpes virus has been identified in association with this disease (Leibovitz and Leibovitz, 1985). The disease typically manifests itself following a stressful event and poses no significant risk to the victim. The disease will spontaneously resolve itself after a variable period of time. Diagnosis of this disease was made by microscopic and ultra-structural (electron microscope) exams of abnormal skin tissue (Leibovitz and Leibovitz, 1985).

The other viral disease reported from elasmobranchs is a disease reported in dusky smooth-hound and leopard sharks (*Triakis semifasciata*) (Johnston, 1975; Kahn and Newman, 1982). The disease is called viral erthryocytic necrosis; it is a viral infection of red blood cells, the oxygen carrying cells of the blood. Animals infected with this virus may become ill or die, with evidence of destroyed red blood cells (hemolysis) and post-mortem evidence of pale organs (McAllister and Stoskopf, 1993). Viral erthryocytic necrosis typically affects young animals with no immunity to the virus. The disease is caused by an iridovirus. The virus specifically targets and attacks red blood cells and can be diagnosed by finding characteristic cellular changes in a blood smear (McAllister and Stoskopf, 1993). Infected cells often contain large intra-cytoplasmic (within the cytoplasm, not the nucleus) "inclusions" in routinely stained blood smears.

Husbandry recommendations

It is easily appreciated that the extent of knowledge concerning viral diseases of elasmobranchs is extremely limited. The study and diagnosis of viral diseases is difficult. However, this fact should not discourage people and organizations housing elasmobranchs from attempting diagnosis of these diseases.

Certain characteristics of a disease or disease outbreak may suggest a viral cause:

1. Disease or death occurs in a young immunologically naive animal or animals.
2. Disease or death occurs in multiple animals over a short period of time.
3. Disease or death occurs shortly after introducing a new specimen or species to the collection.
4. Disease or death occurs following a stressful event (e.g., capture and transport, environmental or water quality changes, unusual social interactions, etc.).
5. Outbreak of disease fails to respond to an appropriate antibiotic therapy or treatment.

If you suspect an outbreak of viral disease in an elasmobranch collection, certain practices may help improve your chances of diagnosis. Clinical samples such as blood or tissue biopsies should be taken from sick animals and sent to a laboratory that specializes in diseases of fishes. Some state or commercial diagnostic laboratories may be comfortable with fish submissions. Collect and freeze tissue, whole blood, and serum samples at -70 °C for viral culture attempts.

A complete post-mortem examination should be performed on animals that die. Tissues such as brain, liver, kidney, heart, or any diseased tissue should be frozen at -70 °C for viral culture attempts. In addition, tissues should be collected into 10% neutral buffered formalin for histopathology. Samples can be small (<1 cm^3) and still be valuable.

Treatment and control

There are currently no recognized treatment or control methods specific to viral diseases. General recommendations include supportive care and minimizing stress.

BACTERIA AND BACTERIAL DISEASES

Bacteria are single-celled organisms that are responsible for disease in a variety of animal species including teleosts and elasmobranchs. Bacterial diseases of elasmobranchs are the most thoroughly studied of the diseases discussed in this chapter. In contrast to other species, elasmobranchs (sharks specifically) may have a unique relationship with bacteria. While it is quite common in all species to find "commensal" bacteria living on the skin surface or within the gastrointestinal tract, sharks are unique in that bacteria are commonly identified within internal organs such as liver and muscle with no evidence of disease (Knight et al., 1987). In other words, bacteria can grow on tissues such as the livers, muscles, and kidneys of absolutely healthy animals. It is theorized that many of these bacteria use or aid in metabolism of urea produced as a normal by-product of shark metabolism.

Another unique aspect of bacteria and elasmobranchs is the relationship between shark bites and bacterial infection in humans. Bacterial infection commonly follows shark bite injury in humans and this secondary infection can often be quite serious. All shark bites should be considered contaminated wounds with high potential for "infection." These wounds, regardless of severity, should be treated by appropriate medical personnel.

Bacterial disease and elasmobranchs

Outbreaks of bacterial disease are relatively uncommon in elasmobranchs. Reports of bacterial disease caused by *Vibrio* spp. are the most common.

Aeromonas salmonicida, a common teleost pathogen, has been documented as a cause of disease in a blacktip reef shark (*Carcharhinus melanopterus*) (Briones et al., 1998).

Flavobacterium sp. was recently isolated by the author from bonnethead shark (*Sphyrna tiburo*) pups showing evidence of neurologic disease. Disease caused by *Flavobacterium* spp. in teleosts results from a toxin produced by the bacteria (Stoskopf, 1993).

Several species of *Vibrio* spp. have been implicated as disease agents in sharks. The most common *Vibrio* sp. isolated from sharks is *Vibrio carchariae* (Grimes et al., 1984; Stoskopf, 1993). *Vibrio carchariae* has been repeatedly implicated as the cause of meningitis (inflammation of the outer covering of the brain) in sand tiger (*Carcharias taurus*), lemon (*Negaprion brevirostris*), and sandbar (*Carcharhinus plumbeus*) sharks, and the spiny dogfish (*Squalus acanthias*) (Stoskopf, 1993). The trematode, *Dermophthirius* sp., has been implicated as a vector of *Vibrio carchariae*, transmitting the disease from shark to shark (Grimes et al., 1984). *Vibrio carchariae* has been isolated from sandbar sharks in association with chronic skin ulcers. A variety of *Vibrio* spp. was isolated from Port Jackson (*Heterodontus portusjacksoni*) and epaulette (*Hemiscyllium ocellatum*) sharks, and southern fiddler rays (*Trygonorrhina fasciata*), that died following a change in salinity (Callinan, 1988).

Husbandry recommendations

In contrast to viral diseases, most bacterial diseases can be diagnosed by bacterial culture of blood or diseased tissues such as the brain, liver, kidney, or spleen. In the author's experience, bacterial culture of cerebrospinal fluid has been a useful diagnostic tool in sharks showing evidence of neurologic disease. *Vibrio* spp. require specialized techniques for isolation, so a diagnostic lab should be contacted prior to submission of samples.

It is important to remember the unique relationship between elasmobranchs and bacteria when interpreting bacterial culture results from sick fishes (i.e., it is always possible to isolate certain species of bacteria from the tissues of healthy sharks). Culture results should be interpreted in combination with a clinical history of illness or death, and evidence of disease in ante- or post-mortem tissues.

Certain characteristics of a disease or disease outbreak may suggest a bacterial cause:

1. Bacterial diseases can be sudden in onset.
2. Disease may be seen in individual animals or whole populations.
3. Disease may be initiated by a stressful event (e.g., an environment or water quality change, movement to a new environment, other diseases, etc.).
4. Lesions, such as "boils", abscesses, or skin hemorrhages, may be observed.
5. Sick animals may respond to appropriate antibiotic treatment.

If you suspect an outbreak of bacterial disease in an elasmobranch collection, certain practices may help improve your chances of diagnosis. Clinical samples, such as blood or biopsies of skin lesions should be taken from sick animals and sent to a laboratory that specializes in diseases of fishes. Contact the diagnostic lab prior to submission of samples to be sure it is equipped with specialized media and techniques to isolate *Vibrio* spp. Culture of blood or abnormal tissues (e.g., abscesses, "boils," ulcers, etc.) should be made. Blood collected ante- or post-mortem can be submitted for bacterial culture.

A complete post-mortem examination should be performed on animals that die. Small samples of tissues should be collected aseptically during the post-mortem exam for bacterial culture. Additional tissue samples should be fixed in 10% neutral buffered formalin for histopathology. Isolation of the brain and culture of the cerebrospinal fluid may be valuable. This practice is essential if the shark had evidence of neurologic disease prior to death.

Treatment and control

Few people have experience with treatment of bacterial diseases in elasmobranchs. Some pharmacokinetic studies have been done with antibiotics and sharks to determine appropriate drug dosing and treatment intervals (Stoskopf *et al.*, 1986). Bacterial culture and antibiotic sensitivity testing may help guide antibiotic choice in elasmobranchs.

The reader is directed to Chapter 29 of this manual for more information about the treatment of bacterial diseases.

FUNGI AND FUNGAL DISEASES

Fungi are common pathogens of a wide variety of species including fishes. Fungal diseases are typically characterized by disease in an individual animal rather than explosive outbreaks affecting multiple animals. Fungi are transmitted via environmental exposure (i.e., contact with contaminated soil in the case of mammals or contaminated water in the case of fishes). The most common fungal pathogen of teleost fishes is *Saprolegnia* spp., an opportunistic environmental pathogen that typically colonizes damaged skin, gill, or fin tissue. However, these oomycetes cannot tolerate saltwater. Fungal agents can cause disease in all age groups of animals (in contrast to bacteria and viruses that commonly infect young or old individuals).

Fungal diseases of elasmobranchs

A specific fungal disease is well characterized in the bonnethead shark. Known as "bonnethead shark disease" this syndrome is the result of infection by the fungus *Fusarium solani*. In addition to bonnethead sharks, this disease has been described in scalloped hammerhead sharks (*Sphyrna lewini*). Affected sharks develop white pustules (or pimples) along their lateral line as well as other sites on the skin and cephalofoil ("bonnet"). In some cases skin erosions or ulcers may be seen. There may be hemorrhages or swelling of the skin as well as hemorrhage into deep muscle or cartilage. Disease in sharks typically follows some environmental change or stressor. In the author's experience, disease caused by *Fusarium* spp. is seen when shallow, warm-water sharks have been kept in deeper, cold water aquarium systems. The disease is progressive, and attempts to treat affected sharks with anti-fungal medication have been unsuccessful to date. Affected sharks die with evidence of deep invasion of the fungus into skin, muscle, cartilage and, occasionally, internal organs.

Once a fungal disease is suspected in an animal, the diagnosis can be made by examination of fluid collected from pustules or from dermal pores adjacent to affected areas of skin. In the author's experience, a bloody red fluid can often be squeezed, with a light pressure, from dermal pores around the head or cephalofoil. This fluid contains fungal elements that are visible by light microscopy. The fluid may be submitted for fungal culture at an appropriate laboratory.

Husbandry recommendations

Certain characteristics of a disease or disease outbreak may suggest a fungal cause:

1. Disease occurs in an animal at any age.
2. Disease occurs in a single animal.
3. White pustules, pimples, or ulcers are seen on the skin.
4. Disease occurs following handling or other stressful event (e.g., reduction in water temperature, injury to the skin, movement to new environment, etc.)
5. Disease fails to respond to appropriate antibiotic therapy or treatment.

If you suspect an outbreak of fungal disease in an elasmobranch collection, certain practices may help improve your chances of diagnosis. Clinical samples, such as fluids from pustules and pimples, or tissue biopsies, should be taken from sick animals and sent to a laboratory that specializes in diseases of fishes. These samples should be cultured for fungi. Some labs may have to send fungal cultures to specialty labs for identification. A rapid initial diagnosis may be achieved by examining fluid collected from pustules or pimples under a microscope.

Treatment and control

As mentioned previously, treatment of fungi is difficult. Some anecdotal success has been reported for *Fusarium* sp. infections in bonnethead sharks by increasing water temperatures. Because of the difficulty of treatment of this disease, prevention is the best tool for management. The occurrence of this disease may be reduced by maintaining susceptible sharks in appropriate environments (i.e., appropriate water quality and temperature regimes) and by reducing stress associated with capture or handling. Skin wounds are a potential portal of entry for fungi, so skin wounds should be avoided or closely monitored when they do occur.

The reader is directed to Chapter 29 of this manual for more information about the treatment of fungal diseases.

QUARANTINE

Quarantine is an important tool for disease prevention when dealing with infectious viral, bacterial, fungal, or parasitic diseases. Quarantine should be imposed any time a new animal is transferred from the wild, another institution, or one independent system to another in the same institution.

A quarantine period allows for detection of disease agents possibly introduced from a newly acquired animal to animals within the existing collection. Quarantine promotes a recovery period for animals following transport, acclimatization of animals into a new environment, and recovery of the immune system, thus increasing resistance to novel pathogens. A minimum 30-day quarantine period is recommended for elasmobranchs. The extent of diagnostic testing (e.g., blood sampling, bacterial culture, fecal examination, etc.) performed during quarantine should be determined at each individual institution. The reader is directed to Chapter 10 of this manual for more information about elasmobranch quarantine regimes.

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