

The Elasmobranch Husbandry Manual: Captive Care of Sharks, Rays and their Relatives

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Chapter 10

Quarantine and Prophylaxis for Elasmobranchs

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Abstract: As applied by aquarium facilities quarantine refers to the process of isolating a new or sick specimen for the purpose of treatment and observation, while prophylaxis refers to the process of applying preventative treatments to existing healthy specimens. Treatment protocols frequently overlap between quarantine and prophylaxis. Exemplary water quality is essential to animals surviving the rigors of chemotherapeutic treatments. Both the duration of a protocol and the allowable density of animals may be influenced by the spatial needs of specific species. Throughout quarantine, sterile techniques should be used to ensure that pathogens are not transferred between aquariums or animals. A thorough understanding of the proper handling and use of chemotherapeutics is essential. Veterinary and pathology laboratory services should be retained, to aid in both diagnoses and treatments. The ability to correctly identify pathogenic organisms (e.g., monogeneans, cestodes, nematodes, crustaceans, protozoans, bacteria, etc.), in combination with an understanding of their life history, will lead to informed diagnoses and more effective treatments.

This chapter briefly reviews issues to consider when formulating quarantine and prophylactic protocols for elasmobranchs, including: water treatment considerations, spatial considerations, sterile techniques, an understanding of pathogens, and modes of medication. A summary of typical chemotherapeutics used during quarantine and prophylaxis, based on a review of 22 public aquariums, is presented.

As applied by aquarium facilities quarantine refers to the process of isolating a new or sick specimen for the purpose of treatment and observation, while prophylaxis refers to the process of applying preventative treatments to existing healthy specimens. Treatment protocols frequently overlap between quarantine and prophylaxis.

Where practical, all new animals should go through quarantine for a minimum of 30 days prior to their introduction to a display or experimental aquarium. This time line is based upon our current understanding of the pathogens that affect elasmobranchs.

Prophylaxis is frequently applied while specimens are in display aquariums and is often based on a

schedule of expectation (e.g., the appearance of monogeneans at the same time each year). In this situation, the effect of a treatment protocol on other species within the display aquarium needs to be carefully reviewed.

Treatment protocols should be based upon a thorough evaluation of specimens, including an assessment of the following: condition, length and weight, behavior, appetite, skin scrapes, and blood profiles, and, where possible, biopsies, lavages, radiography, and ultrasonography.

TREATMENT PROTOCOLS

Water treatment considerations

Quarantine facilities require the same thorough approach to life support system (LSS) design as display aquariums. Biological, mechanical, and chemical filtration each play an important role. Exemplary water quality is essential to animals surviving the rigors of chemotherapeutic treatments.

The use of ultra-violet sterilizers (UV) and/or ozone and activated carbon, as part of the LSS,

should be considered standard. Total counts of some pathogens can be reduced by the use of UV and/or ozone. In addition, UV and ozone can reduce many treatment chemicals to harmless by-products, which subsequently may be removed by activated carbon.

UV units are typically sized to achieve a specific level of irradiation, expressed as microwatts per second per centimeter squared ($\mu\text{W sec}^{-1} \text{cm}^{-2}$). UV unit manufacturers will supply a chart with suggested irradiation levels required to achieve a given kill ratio of specific pathogens. Suggested exposure rates can be as low as $6,000 \mu\text{W sec}^{-1} \text{cm}^{-2}$, while others can be as high as $400,000 \mu\text{W sec}^{-1} \text{cm}^{-2}$. When using UV, a number of issues need to be considered. UV increases the heat load applied to a system. If temperatures are already close to the upper tolerance for elasmobranchs, an increase in chilling capacity may be required. Teflon or quartz sleeves are typically used to separate UV bulbs from the water. Frequent cleaning of these sleeves maintains an effective kill rate. Some UV units do not work well with coldwater aquariums, where condensation can diminish the UV unit's effectiveness. Additionally, organically-rich, turbid water can reduce kill rates. Application of UV is usually via a bypass that allows a portion of aquarium water to be treated. LSS design should be configured to maximize the passage of pathogen-laden water through the UV unit.

Ozone is typically introduced via a venturi to either foam fractionators or contact chambers. When used as a sterilizing agent, ozone is applied at high concentrations to oxidize water-borne parasitic organisms. During this process a number of residual oxidants are produced when used in salt water. Residual oxidants take part in the sterilizing process within the respective reaction chamber. However, if these chemicals persist and are carried into an exhibit, they can present a serious health risk to elasmobranchs. Residual oxidants may be monitored through DPD total oxidant tests, Oxidation Redox Potential, and animal behavior.

Spatial considerations

Elasmobranchs vary widely in their spatial requirements. Serious consideration should be given to these demands when developing quarantine and/or prophylactic protocols. Both the duration of a protocol and the allowable density of animals may be influenced by the spatial needs

of a specific species. In some situations it may not be practical to quarantine a particular species of elasmobranch at all.

Pools of inadequate size or shape can be as devastating as pathogens, causing serious health issues to potentially valuable specimens. If pools are not the appropriate dimension and shape, contact lesions can occur on the caudal fin, ventral surface, and rostrum. Reduction in stocking density, modifying swimming patterns (e.g., introducing visual or physical obstacles), changing the lighting, and the addition of a sand substrate can reduce the occurrence of some of these injuries.

In some instances it may be necessary to maintain elasmobranchs in confined conditions (e.g., if protocols require repeated injections, tube feeding, wound care, etc.). In some specific cases, it may be less stressful to use a smaller pool, where it is easier to catch and restrain the elasmobranch, than a larger pool where an animal can swim freely.

Sterile techniques

Throughout quarantine, sterile technique (e.g., sterilization of nets between uses, etc.) needs to be instituted to ensure that pathogens are not transferred between aquariums or animals. With restricted quarantine spaces, sterile technique includes eliminating aerosol transmission via aeration, bio-towers, etc., and splashing caused by elasmobranchs.

The life history of many parasites includes a dormant stage. Thorough cleaning and sterilization of pools and LSSs, after each quarantine cycle, reduce the chance of transferring problems from one quarantine cycle to another. This process entails reseeding of the biological filters before the start of each quarantine cycle.

Safety and Record keeping

Before adopting quarantine and prophylactic protocols, it is important to review any local guidelines and regulations for the use of selected drugs and chemicals. A thorough understanding of the proper handling and use of any product is essential. Material safety data sheets should be studied and appropriate personal protective equipment (PPE) used.

Veterinary and laboratory (i.e., clinical and pathological) services should be retained, to aid in both diagnoses and treatment. In some locations it may be a legal requirement, or a stipulation by professional zoological associations, to retain these services.

Thorough records should be maintained throughout quarantine and/or prophylaxis. These records will help in assessing the efficacy of treatments. All elasmobranch mortalities should be followed by a complete necropsy, and resulting records maintained for future reference.

Pathogen diagnosis

The ability to correctly identify pathogenic organisms (e.g., monogeneans, cestodes, nematodes, crustaceans, protozoans, bacteria, etc.), in combination with an understanding of their life history, will lead workers to make informed diagnoses and implement more effective treatments. In particular, it is important to understand primary and secondary health concerns. For example, it may be determined that an outbreak of monogeneans has been exacerbated by the presence of environmental stressors (e.g., poor water quality, high population density, etc.). Once monogeneans have infested a population of elasmobranchs, a secondary bacterial infection may ensue and ultimately result in specimen mortality. Any treatment regime should thus address the primary infection (i.e., monogeneans), the secondary infection (i.e., bacteria), and importantly, any conditions that have aided the disease process (i.e., poor water quality and/or high population density), for the regime to be effective.

Monogeneans represent the greatest challenge to newly-arrived elasmobranchs. These organisms are difficult to eradicate because of their ability to remain viable, without a host, for extended periods of time. Control of these pathogens, through quarantine, is recommended. If quarantine is impractical, serious consideration should be given to the application of a medicated bath (e.g., praziquantel) before elasmobranchs are moved into their destination aquarium.

If an elasmobranch is suspected to have a specific pathogen, but is asymptomatic and presents no risk to other animals (e.g., in the case of species-specific parasites), it may be deemed appropriate to leave the animal untreated (i.e., forgo prophylaxis). For parasites (e.g., trematodes,

cestodes, etc.) requiring an intermediate host that is not present within the system, it is advisable to let the parasite perish naturally. Wherever possible, it is preferred to keep treatments to a minimum. Although chemotherapeutic treatments are obviously intended to aid elasmobranchs, medication will always present an associated stress that could do more damage to the host animal than the intended target pathogen.

Mode of medication

Immersion (bath)

When preparing medicated baths it is critical to accurately assess the volume of treatment water before adding the medication. Water volume can be determined by using a calibrated flow meter, a calibrated container, or by a calculation of vessel volume. For aquariums with irregular dimensions, volume can be calculated by adding a known weight of salt and measuring the change in salinity. Dividing the weight of added salt (grams) by the change in salinity ($\text{g l}^{-1} = \text{‰} = \text{ppt}$) provides the vessel volume in liters. Once the volume of the treatment vessel is known, it is important to accurately calculate the amount of drug or chemical to add to the vessel to achieve the desired dosage. It is highly recommended to have two people perform the calculations independently to ensure accuracy.

An important consideration, when applying medicated baths, is an understanding of the chemical's reaction to LSS components (e.g., some chemicals are destroyed by ozone), and indeed their impact on LSS components (e.g., some antibiotics can damage the beneficial bacteria inside biological filters). Another important consideration is the possibility of synergistic effects—e.g., the presence of nickel at just $2.0 \mu\text{g l}^{-1}$ will double the effect of a copper treatment (Sorensen, 1991). Thus, a 2.0 mg l^{-1} antiparasitic treatment of copper effectively becomes a 4.0 mg l^{-1} lethal dosage of copper, in the presence of $2.0 \mu\text{g l}^{-1}$ nickel. In some cases synergy can be used to advantage (e.g., a lower concentration of two treatments—copper and organophosphates—can be used to effectively treat ectoparasites).

Once a bath is complete, medicated water must be safely disposed in accord with domestic and international regulations. This precaution is important not only for the products themselves

(i.e., antibiotics, heavy metals, organophosphates, etc.), but also filter media (e.g., activated carbon) used to remove products from the water.

Oral

When administering oral medications it is important to have an accurate measurement of specimen weight, before calculating dosages. The smaller the animal the more critical it is to have an accurate and precise measurement.

Some oral medications may be rejected by an elasmobranch because of their unusual taste. To disguise the taste, it may be necessary to secrete gel caps, filled with the medication, within a food item.

Parenteral (injectable)

As per oral medications, it is important to have an accurate measurement of specimen weight before calculating the dosage of injectable medications. Most parenteral treatments are administered intramuscularly (IM). Do not sterilize the injection site with alcohol prior to administration as alcohol can damage elasmobranch skin. Intramuscular medications are typically administered via a large muscle mass (e.g., the dorsal saddle) and in some cases multiple injection sites may be required if a large volume of medication is to be administered. Massaging the injection site, during and after administration, can reduce the risks of medications leaking out of the intended site.

Protocol formulation

In addition to the removal of hooks and tags, the treatment of gross lesions and abrasions, and the potential treatment of inappetence, a quarantine protocol for elasmobranchs should address the following problematic organisms: external parasites (i.e., monogeneans, crustaceans, and protozoans), internal parasites (i.e., cestodes, nematodes, and protozoans), and potential secondary bacterial infections.

Table 10.1 presents a summary of some typical chemotherapeutics successfully used during the quarantine and prophylaxis of elasmobranchs.

The information contained in Table 10.1, and the discussion that follows, represents a summary of a survey conducted during 2001 of 22 public aquariums. In general, two medications should not be applied simultaneously, although some oral treatments may be given during long-term medicated baths. Extreme caution should be exercised when interpreting these data as they represent very small sample sizes, in some cases only a single individual, and do not have the support of pharmacokinetic studies.

CHEMOTHERAPEUTICS

Amikacin

Amikacin sulphate is a broad-spectrum antibiotic. Amikacin has been administered via IM injection at a dosage of 3.0-5.0 mg kg⁻¹ (5.0 mg kg⁻¹ in the case of the ocellate river stingray, *Potamotrygon motoro*) every 72 hours for five consecutive treatments.

Ceftazadime

Ceftazadime pentahydrate (Fortaz®, Glaxo-SmithKline Inc., USA) is a broad-spectrum antibiotic. Ceftazadime has been administered via IM injection at a dosage of 30.0 mg kg⁻¹ every 72 hours (8 hours in the case of the spotted eagle ray, *Aetobatus narinari*) for five consecutive treatments.

Copper

Copper (citrated and non-citrated) is used as a treatment for external parasites, especially monogeneans, crustaceans, and protozoans. Copper has been administered as a bath at a dosage of 0.15 mg l⁻¹ for up to three months (3-4 months in the case of the bat eagle ray, *Myliobatis californica*) and at 0.20 mg l⁻¹ for a period of 30 days (0.15 mg l⁻¹ for a period of 30 days in the case of the following species: sand tiger shark, *Carcharias taurus*; whitespotted bambooshark, *Chiloscyllium plagiosum*; brownbanded bambooshark, *Chiloscyllium punctatum*; nurse shark, *Ginglymostoma cirratum*; epaulette shark, *Hemiscyllium ocellatum*; and the smalltooth sawfish, *Pristis pectinata*). When applying copper baths, activated carbon filtration should be discontinued. Never use copper in the presence of formalin, praziquantel, or trichlorfon.

Table 10.1. Chemotherapeutics used in 22 public aquariums when applying prophylaxis during quarantine (Q) and prophylaxis in exhibit (P). Please refer to body text for details of dosages and treatment conditions for each medication.

Species name	Common name	Amikacin	Ceftazidime	Copper	Enrofloxacin (IM)	Enrofloxacin (PO)	Fenbendazole	Formalin	Hydrogen Peroxide	Ivermectin	Metronidazole	Furazace	Praziquantel (bath)	Praziquantel (PO)	Salinity	Trichlorfon
<i>Aetobatus narinari</i>	spotted eagle ray	P	P		P								Q			Q
<i>Atelomycterus marmoratus</i>	coral catshark						P	P								
<i>Carcharhinus acronotus</i>	blacknose shark	P			P		P				P		P	P		Q+P
<i>Carcharhinus amblyrhynchos</i>	grey reef shark															Q
<i>Carcharhinus leucas</i>	bull shark	P			P	P		P								Q+P
<i>Carcharhinus limbatus</i>	blacktip shark	P			P								Q+P			Q+P
<i>Carcharhinus plumbeus</i>	sandbar shark	P			P								Q+P			Q+P
<i>Carcharias taurus</i>	sand tiger shark	P		Q+P	P	P	P				P		Q+P	P	Q	Q+P
<i>Cephaloscyllium ventriosum</i>	swellshark							P								
<i>Chiloscyllium plagiosum</i>	whitespotted bamboo shark	P		Q+P	P			Q				P	Q+P			Q+P
<i>Chiloscyllium punctatum</i>	brownbanded bamboo shark	P		Q+P	P			Q				P	Q+P			Q+P
<i>Dasyatis americana</i>	southern stingray	P		Q+P	P		P				P	P	Q+P	P		Q+P
<i>Dasyatis brevis</i>	whiptail stingray							Q	Q			Q				
<i>Dasyatis sabina</i>	Atlantic stingray	P		Q+P	P		P				P	P	Q+P	P		Q+P
<i>Ginglymostoma cirratum</i>	nurse shark	P		Q+P	P	P	P	Q			P	P	Q+P	P		Q+P
<i>Hemiscyllium ocellatum</i>	epaulette shark	P		Q+P	P			Q				P	Q+P			Q+P
<i>Heterodontus francisci</i>	horn shark	P						Q				Q	Q			Q
<i>Heterodontus portusjacksoni</i>	Port Jackson shark	P	Q										Q			
<i>Himantura fai</i>	pink whipray				P											
<i>Myliobatis californica</i>	bat eagle ray			P				Q	Q			Q				
<i>Negaprion brevirostris</i>	lemon shark	P		Q+P		P	P				P		Q+P	P	Q	Q+P

Table 10.1 (continued). Chemotherapeutics used in 22 public aquariums when applying prophylaxis during quarantine (Q) and prophylaxis in exhibit (P). Please refer to body text for details of dosages and treatment conditions for each medication.

Species name	Common name	Amikacin	Ceftazidime	Copper	Enrofloxacin (IM)	Enrofloxacin (PO)	Fenbendazole	Formalin	Hydrogen Peroxide	Ivermectin	Metronidazole	Furazace	Praziquantel (bath)	Praziquantel (PO)	Salinity	Trichlorfon
<i>Orectolobus japonicus</i>	Japanese wobbegong	P			P			Q					Q+P			Q+P
<i>Orectolobus ornatus</i>	ornate wobbegong	P			P			Q					Q+P			Q+P
<i>Platyrrhinoidis triseriata</i>	thornback guitarfish							P								
<i>Potamotrygon motoro</i>	ocellate river stingray	P						P								
<i>Pristis pectinata</i>	smalltooth sawfish	P		Q+P	P		P				P		Q+P	P	Q	Q+P
<i>Pteroplatytrygon violacea</i>	pelagic stingray							Q				Q				
<i>Raja inornata</i>	California ray							P								
<i>Rhinobatos productus</i>	shovelnose guitarfish	P		Q+P	P		P	Q			P	P	Q+P	P	Q	Q+P
<i>Rhinoptera bonasus</i>	cownose ray	P	P	Q+P	P								Q+P			Q+P
<i>Scyllorhinus canicula</i>	smallspotted catshark									Q						Q+P
<i>Scyllorhinus stellaris</i>	nursehound									Q						Q+P
<i>Sphyrna zygaena</i>	smooth hammerhead											P				
<i>Squalus acanthias</i>	spiny dogfish							P								
<i>Squatina californica</i>	Pacific angelshark							P								
<i>Stegostoma fasciatum</i>	zebra shark	P			P		P				P		Q+P	P	Q	Q+P
<i>Trienodon obesus</i>	whitetip reef shark	P	P		P								Q+P		Q	Q+P
<i>Triakis semifasciata</i>	leopard shark	P		P	P			Q	Q				Q+P			Q+P
<i>Urobatis jamaicensis</i>	yellow stingray	P		Q+P	P		P				P	P	Q+P	P		Q+P
<i>Zapteryx exasperata</i>	banded guitarfish							P								

Enrofloxacin

Enrofloxacin (Baytril®, Bayer Corp., USA) is a broad-spectrum antibiotic. Enrofloxacin has been administered both orally and via IM injection at a dosage of 10.0 mg kg⁻¹ every 5-7 days (2 days in the case of the pink whipray, *Himantura fai*, and 3.5 or 7 days in the case of the following species: blacknose shark, *Carcharhinus acronotus*; bull shark, *Carcharhinus leucas*; blacktip shark, *Carcharhinus limbatus*; and the sandbar shark, *Carcharhinus plumbeus*) for three to five consecutive treatments.

Fenbendazole

Fenbendazole (Panacur®, Intervet Inc., USA) is an antihelminthic used for the treatment of internal parasites. Fenbendazole has been used in elasmobranchs to treat nematodes at an oral dosage of 25.0 mg kg body weight⁻¹ for 3x each week, over three consecutive weeks of treatment.

Formalin

Formalin is an antibiotic, antihelminthic, crustacide, and protozoacide. Formalin has been applied as a bath at a dosage of 250 mg l⁻¹ for a period of one hour. Formalin has been used in conjunction with hydrogen peroxide when treating the leopard shark (*Triakis semifasciata*), the bat eagle ray, and the whiptail stingray (*Dasyatis brevipes*).

Hydrogen peroxide

Hydrogen peroxide is an antibiotic, antihelminthic, crustacide, and protozoacide. Hydrogen peroxide has been applied as a bath at a dosage of 150.0 mg l⁻¹ for a period of one hour.

Ivermectin

Ivermectin (Ivomec®, Merial Inc., USA) is an antihelminthic used for the treatment of internal parasites. Ivermectin has been used in elasmobranchs to treat nematodes and cestodes administered via IM injection at a dosage rate of 200 mg kg⁻¹ every 15 days for two treatments.

Metronidazole

Metronidazole (Flagyl®, Rhone-Poulenc Rorer Pharmaceuticals Inc., USA) is a protozoacide and anaerobe antibiotic. Metronidazole has been used in elasmobranchs at an oral dosage of 25.0 mg kg body weight⁻¹ for 3 days a week, over three consecutive weeks of treatment.

Furanace

Furanace (Nitrofurazone, Novalek Inc., USA) is broad-spectrum antimicrobial. Furanace has been applied as a bath at a dosage of 20.0 mg.l⁻¹ for 2 hours each day of five consecutive days of treatment (10.0 mg l⁻¹ for 10 hours each day of five consecutive days of treatment in the case of the smooth hammerhead shark, *Sphyrna zygaena*; 10.0 mg l⁻¹ for 8 hours each day of seven consecutive days of treatment in the case of the bat eagle ray; and 10.0 mg l⁻¹ for 8 hours each day of five consecutive days of treatment in the case of the following species: the whitespotted bambooshark; the brownbanded bambooshark; the nurse shark; the horn shark, *Heterodontus francisci*; and the epaulette shark). When applying furanace baths, activated carbon filtration, ozone dosing, and UV irradiation should be discontinued.

Praziquantel

Praziquantel (Praziquantel 100%, Professional Pharmacy Services Inc., USA) is an antihelminthic used for the treatment of both internal and external plathyhelminthes. Praziquantel has been applied as a bath to treat monogeneans at a dosage of 10.0 mg l⁻¹ for a period of two hours and at 2.0 mg l⁻¹ for a period of 48 hours (2-20 days in the case of the sandbar shark). When applying praziquantel baths, activated carbon filtration, ozone dosing, and UV irradiation should be discontinued. Never use praziquantel in the presence of copper or trichlorfon. Praziquantel has been used in elasmobranchs to treat trematodes and cestodes at an oral dosage of 50.0 mg kg body weight⁻¹ for 3 days a week, over three consecutive weeks of treatment.

Salinity

Reduced salinity can be used as an antihelminthic, crustacide, and protozoacide. A reduced salinity of 15.0 ‰, maintained for a period

of 14 days, has been used to treat elasmobranchs for external parasites, both as a stand-alone treatment or as a complement to other immersion medications. A 30-minute bath of freshwater has been used to treat lemon sharks (*Negaprion brevirostris*) for external parasites, as has a reduced salinity of 10.0-15.0 ‰ maintained for a period of four weeks.

Trichlorfon

Trichlorfon (Dylox® 80, Bayer Corp., USA) is an antihelminthic and crustacicide. Trichlorfon has been applied as a bath to treat monogeneans and parasitic crustaceans at a dosage of 0.5 mg l⁻¹ (0.3 mg l⁻¹ in the case of the grey reef shark, *Carcharhinus amblyrhynchos*) for a period of 24 hours, once a week, for a total of four treatments (0.25 mg l⁻¹ for a period of 24 hours, once every 10 days, for a total of five treatments in the case of the smallspotted catshark, *Scyliorhinus canicula*, and the nursehound, *Scyliorhinus stellaris*). When applying trichlorfon baths, activated carbon filtration, ozone dosing, and UV irradiation should be discontinued. Never use trichlorfon in the presence of copper, formalin or praziquantel.

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