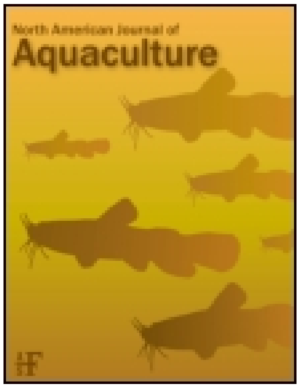

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COMMUNICATION

The Role of Activated Lignite Carbon in the Development of Head and Lateral Line Erosion in the Ocean Surgeon

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Abstract
Head and lateral line erosion (HLLE) is an acute or chronic, often progressive problem affecting captive fishes. Its etiology is enigmatic. This study examined the relationship between the use of activated carbon as a filtrant and the development of HLLE lesions in ocean surgeons *Acanthurus bahianus*. Three identical, 454-L marine aquarium systems were established. Thirty-five ocean surgeons were distributed among the three aquarium systems. Activated lignite carbon was added to one system, and pelleted carbon was added to the second system. The fish in the third system were not exposed to any carbon. All 12 fish that were exposed to lignite carbon developed severe HLLE within 3 months. The 12 fish that were exposed to pelleted carbon did not develop gross symptoms, but microscopic lesions were discovered upon histological examination. The 11 control fish did not develop any visible or microscopic lesions. Based on these results, the use of activated lignite carbon in marine aquariums that house HLLE-susceptible species is discouraged.

Head and lateral line erosion (HLLE; also known as marine head and lateral line erosion or head and lateral line erosion syndrome) is a bilaterally symmetrical dermal erosion that occurs in some captive marine fishes. Beginning as small pits around the fish's eyes, light-colored lesions then develop along the fish's lateral line system and onto wider areas of the body, sometimes involving the unpaired fins.

Although rarely fatal, HLLE does cause disfigurement, making the fish less suitable for public aquarium display. At least 20 families of fishes have been identified as having developed HLLE in captivity (Table 1). The symptoms of HLLE and the degree to which lesions develop differ among species (Hemdal 2006).

Many causes and treatments for this syndrome have been suggested, but controlled studies are lacking. An informal survey of 100 advanced aquarists in 2009 (J. Hemdal, unpublished data) identified over 25 suspected causes for HLLE in marine fishes. The majority of aquarists believed that HLLE in marine fishes, such as tangs (Acanthuridae) and angelfishes, is caused by a dietary deficiency—notably a deficiency in vitamin C or highly unsaturated fatty acids. General stress caused by captivity also was cited as a major cause of HLLE. Other survey responses included stray electrical current, the use of copper medications, lack of natural sunlight, the use of activated carbon, or the presence of a variety of chemical pollutants as possible causes of HLLE. Viral and bacterial infections have also been implicated in causing this syndrome (Hemdal 1989; Varner and Lewis 1991). In the survey results, 18 treatment methods were reported to have given complete remission of HLLE symptoms in captive fishes. Eighty-four percent of the cases of remission involved moving the fish to a new aquarium as one part of the treatment. This indicates that there are conditions in aquariums that can be changed to reduce the incidence of HLLE. When the survey results were limited to professional public aquarists, 19% of the respondents found complete reversal of HLLE by removing carbon filtration. In addition, 75% of this subgroup implicated the use of carbon in the development of HLLE in at least some instances. Pilot studies at the Toledo Zoo, Toledo, Ohio, also demonstrated that the use of activated lignite carbon caused the formation of HLLE lesions in some fishes and that moving the fish to aquariums without carbon filtration allowed the lesions to resolve without additional treatment. Carbon is commonly used in closed-system aquariums to remove some organic compounds (Noga 2000). The objective of this study was to better identify the relationship between carbon use and HLLE development so that aquarists can make more informed decisions regarding the use of carbon in their aquarium systems.

METHODS

Three 454-L marine aquarium systems were set up in identical fashion by use of new tanks and equipment. Each system consisted of two 200-L aquariums and a 54-L sump. Each tank

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TABLE 1. Fish families with members that are known to be susceptible to head and lateral line erosion (HLLE)-like syndromes when housed in aquariums (J. Hemdal, personal observation).

Family name	Common name
Acanthuridae	Surgeonfishes
Blenniidae	Combtooth blennies
Centrarchidae	Sunfishes
Ceratodontidae	Australian lungfish
Chaetodontidae	Butterflyfishes
Cichlidae	New World cichlids
Gadidae	Cods
Grammatidae	Basslets
Haemulidae	Grunts
Labridae	Wrasses
Lutjanidae	Snappers
Muraenidae	Morays
Moronidae	Temperate basses
Percidae	Perches
Plesiopidae	Roundheads
Pomacanthidae	Angelfishes
Pomacentridae	Damselfishes
Protopteridae	African lungfishes
Scorpaenidae	Scorpionfishes
Serranidae	Sea basses, groupers, and soapfishes

was filled with synthetic seawater (Instant Ocean; United Pet Group, Cincinnati, Ohio).

Light levels measured on a light meter (Model 214; GE, Cleveland, Ohio) ranged between 54 and 323 lx depending on the location above the aquarium. Voltage between the aquariums and a common ground was monitored with a voltmeter (Model SP-10a; A.W. Sperry, Hauppauge, New York) and was found to be very low, ranging between 0.2 and 1.0 V AC.

Each system was filtered by means of a biological filter comprising 20 kg of “live rock” apportioned between the sump and the two tanks in each system. Live rock is a porous, calcium-based rock that has been colonized by nitrifying bacteria and epilithic organisms. The live rock is used to oxidize the fish’s ammonia waste to nitrite and then to nitrate. Before fish were placed into the aquariums, each system was challenged with the addition of ammonium chloride (total = 9.6 g) apportioned over a 50-d period. The ammonium chloride challenge ensured that the bacterial population in the live rock would be sufficient to assimilate the ammonia produced by the fish.

The aquariums and sumps were aerated from a central compressor with air passed through a pleated high-efficiency particulate air filter. Water was pumped from each system sump to the aquariums through a Supreme 1200b water pump (Danner Manufacturing, Islandia, New York) at a flow rate of approximately 1,200 L/h into each tank. Water returned to the sump by gravity through screened overflows. Water temperature was maintained within a range of 24–28°C by use of 300-W Theo heater–thermostats (Hydor USA, Sacramento, California).

The study fish were ocean surgeons *Acanthurus bahianus* obtained from a supplier of public aquarium fish (Dynasty Marine Associates, Marathon, Florida); the fish were collected by nets and were not exposed to activated carbon before their arrival. All of the fish were treated prophylactically by the supplier with copper, nitrofurazone, and a formalin dip. The fish were not exposed to any further medications during the 4-month study. The study fish were placed into plastic bags with water and oxygen and were shipped by air cargo in standard tropical fish shipping containers. The transport from Florida to Ohio took place in April, and the transit time was less than 24 h. The 35 test fish ranged from 60 to 95 mm in total length and from 11 to 26 g in body mass. Twelve fish were placed into a system to be exposed to lignite carbon, 12 fish were placed into a system to be exposed to pelleted carbon, and 11 fish were housed in a control system that was not exposed to either type of carbon. Selection of the fish for each system was randomly made during the acclimation process.

One week after the fish arrived, 500 g of unwashed lignite activated carbon (NORIT Americas, Atlanta, Georgia) were placed into a mesh bag and added to the filter sump of the first system. At the same time, 500 g of extruded pellet carbon (Allied Techno Chemical, Klang, Selangor, Malaysia) were placed into a mesh bag and added to the filter sump of the second system. No carbon was added to the control system. Two months later, the carbon in the first and second systems was supplemented with an additional 500 g of each type. This corresponds to the typical use of carbon in aquariums and to the recommendations made by Spotte (1979) for the addition of carbon at 1 g/L of water and for replacement of the carbon every 2 months. The two carbon types used in this study were chosen because they are commonly used as filtration media in aquariums.

The fish were initially fed live brine shrimp *Artemia* spp. as they were gradually adapted to prepared diets. Once adapted to prepared food, the fish were fed twice each day. Half of the fish in each system were fed a pelleted food (Spectrum Marine Fish Formula; New Life International, Homestead, Florida), which was shown in a previous study not to induce HLLE in comparison with fish that received a flake food (Tilghman et al. 2003). The other half of the fish in each system were fed a standard general flake food diet (Prime Tropical Flake; Zeigler Brothers, Gardners, Pennsylvania).

Water quality characteristics (temperature, specific gravity, pH, and dissolved oxygen) in each system were measured weekly with a Model HI 9828 multiparameter probe (Hanna Instruments, Woonsocket, Rhode Island). Because of an operational problem with the calibration of the probe, accurate dissolved oxygen readings were obtained for only the final month of the study. Ammonia, nitrite, and nitrate measurements were taken monthly with a Hach Model DR2000 spectrophotometer (Hach Co., Loveland, Colorado). Water changes were performed for each system at a rate of 45% per month. Small amounts of water (~3 mL) were transferred between all three systems on a weekly basis; the intent of these transfers was to demonstrate that there were no easily communicable fish diseases present in one system and not another. Because a reovirus has been associated with HLLE lesions in marine angelfishes (Varner and Lewis 1991), it was important not to restrict the potential transport of virus particles between the test systems.

After 4 months, two fish from the lignite carbon system, one fish from the pelleted carbon system, and one fish from the control system were humanely euthanized with tricaine methanesulfonate (MS-222). The whole bodies of the fish were preserved in a 10% solution of neutral buffered formalin and were submitted to Northwest ZooPath, Monroe, Washington, for analysis. Tissues were decalcified and then sectioned for comprehensive histological examination.

RESULTS

The water quality was maintained within acceptable parameters for tropical marine aquarium fish as outlined by Spotte (1979). The water quality data for the three test systems as well as the acceptable ranges are listed in Table 2.

Minor HLLE lesions were noted on two fish 20 d after 500 g of activated lignite carbon was added to the sump of their system. The lesions then began to develop on the additional fish in the lignite carbon system and grew in size; after 4 months, all of the fish in that system showed severe lesions ($N = 12$). These results were statistically significant in comparison with the control fish, none of which developed lesions ($N = 11$; chi-square analysis: $P < 0.001$). After 3 months, one ocean surgeon from the lignite carbon system died. This fish had severe HLLE, but the immediate cause of death was attributed to aggression from tankmates. No other fish died during the course of the study.

The areas of the visible lesions in the two sacrificed fish from the lignite carbon system were measured from digital images by using ImageTool Software (Health Science Center, University of Texas, San Antonio). The lesions involved an estimated 44% and 33% of the respective total body surface areas of these fish.

The histopathology report for the two fish from the lignite carbon system stated that their lateral lines had multifocal, bilateral (but asymmetrical), mild mixed inflammation within the mucosa. The inflammation predominantly involved mononuclear cells and rodlet cells with fewer granulocytes. There were also some associated exfoliative changes in the mucosal epithelium of the lateral line. The skin of the two fish from the lignite carbon system showed extensive foci of exfoliation, erosion, ulceration, and necrosis associated with an infiltrate of moderate to large numbers of mixed mononuclear, granulocytic, and histiocytic cells in the underlying dermis. The lesions were especially severe on the head and on the dorsal aspects of the body involving the fins. In these areas, inflammation extended into the underlying bone and was associated with bone resorption and necrosis. In the caudal region, ulceration was also associated with focal mycotic and bacterial colonization of the ulcerated surface. These were assumed to be secondary infections by opportunistic organisms.

The specimen from the pelleted carbon system showed the same lateral line inflammation microscopically as the fish in the lignite carbon system, but there were no grossly visible lesions on that individual or on any of its cohorts. Sections of a crustacean parasite were found in the lumen of the lateral line, but this also produced no grossly visible symptoms and was considered to be an incidental finding.

The histology of the control specimen indicated no inflammation of the lateral line, and there were no grossly visible lesions on the control specimen or its cohorts. Fish from all three systems showed microscopic evidence of myxozoanosis and intestinal trematodiasis, but these findings were not considered to be a factor in the HLLE lesion development because they were present in the asymptomatic control fish as well as the HLLE-affected fish.

The fish were held in their respective systems for an additional 4 months after the conclusion of the study. During this time, two fish from the pelleted carbon system and one fish from the lignite carbon system died from undetermined causes. Two fish in the pelleted carbon system developed small HLLE lesions around their eyes. None of the fish in the control system died, and none developed any visible lesions. After the conclusion of the study, the HLLE lesions on the fish in the lignite carbon system remained severe despite the removal of the lignite carbon and four 45% water changes.

TABLE 2. Range of water quality conditions in each of three systems during the study of head and lateral line erosion (HLLE) in ocean surgeons (DO = dissolved oxygen). The suitable range of each variable (from Spotte 1979) is also presented.

Variable	Lignite carbon system	Pelleted carbon system	Control system	Suitable range
Temperature (°C)	24–28	25–28	24–28	Not given
Specific gravity	1.022–1.024	1.022–1.025	1.022–1.024	1.024
pH	8.0–8.3	8.0–8.3	8.0–8.3	8.0–8.3
DO (% saturation)	94–97	94–97	94–96	>85
NH_3-N (mg/L)	0.00–0.03	0.00–0.04	0.00–0.10	0.00–0.10
NO_2^--N (mg/L)	0.01–0.04	0.003–0.04	0.02–0.04	0.00–0.10
NO_3^--N (mg/L)	3.4–7.9	3.0–7.6	3.6–9.2	0.0–20.0

FIGURE 1. Ocean surgeon exhibiting severe head and lateral line erosion 81 d after exposure to activated lignite carbon.

DISCUSSION

The hypothesis for this study was that lignite carbon causes HLLE in fishes. Originally, it was presumed that carbon dust (known as fines) was the causative agent. This premise was based on anecdotal observations in which carbon was removed from an aquarium and the water was changed, yet HLLE symptoms were still produced by adding susceptible fish to the aquarium, indicating that there was some unknown residual action by the carbon. Carbon fines were frequently discovered in the filter sumps and substrate of these tanks. Changing all of the aquarium's water, furnishings, and substrate would then render the aquarium safe for housing susceptible fishes. Furthermore, it has been reported anecdotally that for aquariums equipped with foam fractionators, carbon use is not associated with a high prevalence of HLLE (J. Hemdal, personal observation). It was thought that these systems did not induce HLLE as readily because the foam fractionators export particulate organic carbon (including carbon fines) from the water. In addition, the hard, pelleted carbon used in this study did not cause severe HLLE, whereas the soft, dusty lignite carbon did. However, no carbon fines were seen during histological examination of the lesions of study fish; therefore, the effect may be transitory, the fines may be too small to visualize during histology, or there is some other factor associated with carbon use that causes HLLE in susceptible fishes. Figure 1 depicts an ocean sturgeon exhibiting severe head and lateral line erosion.

Lateral line depigmentation, which is similar to HLLE, has been reported in channel catfish *Ictalurus punctatus* that were fasted for 12 months (Corrales et al. 2009). In that study, there was no mention of whether the channel catfish were exposed to carbon filtration. Another similar condition, termed chronic erosive dermatopathy, has been seen in aquacultured Murray cod *Maccullochella peelii peelii* (Baily et al. 2005). The use of borehole (well) water was identified as the causative factor, and moving the affected fish to river water alleviated the symptoms (Baily et al. 2005). Activated carbon was not used to treat the borehole water in that study (J. F. Turnball, University of Stirling Institute of Aquaculture, personal communication). Therefore, it seems that either there are different syndromes with similar etiology or there are multiple causes for this syndrome in captive fishes.

Based on the trenchant effect of carbon on the ocean surgeons assessed in this study, we discourage the use of activated lignite carbon in marine aquariums that house HLLE-susceptible species. Other means of water quality management should first be explored, including noncarbon chemical filtration, foam fractionation, and water exchanges. Extruded carbon pellets may be more suitable, especially if used sparingly.

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