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Captive Maintenance of the Lesser Electric Ray, with Observations of Feeding Behavior

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Abstract.—The lesser electric ray (*Narcine brasiliensis*) is a preferred model for research in the neurochemistry of cholinergic neurons. However, proper handling must be provided to prevent physiological deterioration prior to use. Large concrete tanks with subgravel biological filters and fine-grain sand substrates were used to house the fish. The tanks required lining with plastic or fiberglass to prevent animals from swimming into the walls. Captive animals were subject to bacterial infections and infestations of gill parasites which, if left untreated, were lethal. Weekly dipping in a mixture of the antibiotic halquinol and formalin was used to control these problems. Water temperatures below 17°C resulted in sluggish behavior, and mass mortality occurred at 14°C. Rays fed exclusively on live burrowing polychaetes; *Arenicola cristata* and *Nereis virens* were used during the study. Juvenile rays required smaller prey than adults. Feeding behavior in captivity is described.

The lesser electric ray (*Narcine brasiliensis*), along with the closely related species, *Torpedo californica* and *T. nobiliana,* is valued for research in cholinergic neurochemistry because of its massive electrogenic cells. Over 900 papers based on *Torpedo* spp. were cited in *Biological Abstracts* (BioSciences Information Service, Philadelphia) between 1980 and 1984, as well as 45 based on *Narcine* since 1968. However, *Torpedo* spp. are difficult to obtain on a reliable basis and cannot be maintained in captivity.

The lesser electric ray is widely distributed, occurs in shallow warm water, and can be readily collected throughout the year (Rudloe, in press). However, literature on the ecology or the behavior of the species is extremely limited. Anatomy, range, and a few notes on diet were reported by Bigelow and Schroeder (1953). Mathewson et al. (1958) and Bennett and Grundfest (1961) described the electric organs, and Funicelli (1975) reported the species' distribution in Mississippi Sound.

It is essential that animals held for physiological use be maintained in healthy conditions. Martini (1978) pointed out that elasmobranchs undergo significant physiological deterioration in captivity in the absence of proper care and feeding. He noted changes in serum concentrations of ions, proteins, glucose, and cholesterol, as well as muscular atrophy and changes in hematocrit and kidneys in spiny dogfish (*Squalus acanthias*). Conditions and duration of captivity are major undescribed vari-

ables in many physiological studies of both marine and nonmarine species.

The lesser electric ray, in particular, has never been reported to feed in captivity, so prior studies may have been based on semistarved animals. My study is part of an effort to make this species more accessible to neurological laboratories. Here, I report methods developed for prolonged holding of lesser electric rays prior to laboratory use.

Methods

Lesser electric rays were captured offshore from Apalachicola, Florida, during 60-min tows of a 12-m-wide, 3.8-cm-stretched-mesh otter trawl. Animals were held on deck in a covered fiberglass box (1 m × 1.5 × 1 m) with polyurethane insulation on all sides. A 110-V air pump built onto the side aerated the water via 1.27-cm-diameter polyvinyl chloride air lines. Two interior removable partitions with 15-cm-square windows of 1.27-cm-mesh screen baffled the surge produced by the vessel's rolling. The box was periodically flushed with seawater from the vessel's deck hose. This system successfully transported up to 100 lesser electric rays for trips lasting 24 h. The fish were transferred to onshore holding facilities in plastic bags with seawater and oxygen inside Styrofoam coolers or in a similar aerated baffled vat on a pickup truck. During transport to the laboratory in Panacea, Florida, the fish were held in a 1:1 mixture of laboratory seawater and water from the holding box. Travel time on the truck was approximately 1 h. At the laboratory, additional laboratory water was added to the transport box to continue the acclimation process. After 10 min, the animals were transferred in dip nets to holding tanks.

Each lesser electric ray was sexed, measured, and tagged with an FT-6 dart tag (Floy Tag and Manufacturing, Seattle, Washington). Parasitic leeches (*Branchellion ravenelii*) were removed from the gills during this process. Handling time per fish was approximately 3 min. Records were kept for each fish of date of arrival and date of shipping. If a fish died, its date of death and associated symptoms also were recorded.

Lesser electric rays were held in cement tanks with subgravel biological filters; the water received ultraviolet treatment. Densities were 50–200 fish/tank. The substrate initially was a layer of coarse unsorted quartz sand about 30 cm deep. This was eventually replaced with a 6-cm-deep layer of fine wind-sorted sand collected from the top of a sand dune. A sheet of Filterweave filter cloth was placed below the sand. Tank capacities ranged from 6,000 to 22,000 L; the tanks were connected to an 88,000-L recirculation system with biofiltration. Temperature, salinity, oxygen, nitrate, nitrite, ammonia, and pH were monitored weekly within each tank and stayed within acceptable limits. Water temperature was normally maintained between 20 and 26°C. Salinity was maintained at 25–30‰. The tank walls were lined with plastic swimming pool cover material (Centry Brand Solar Pool Cover) over a layer of foam matting.

Each week specimens were dipped for 60 min in a mixture of the antibiotic halquinol (dosage, 25 mg/L) and formalin (dosage, 0.17 mL/L). Food in the form of live polychaete worms (*Nereis virens* and *Arenicola cristata*) was stocked at a rate of approximately one worm/fish daily.

Results and Observations

Mortality in Captivity

Altogether, 2,273 lesser electric rays were used in this study between March 1985 and March 1987. Live fish were held for periods ranging from several days to 4 months before they were shipped to a neurochemical laboratory.

Injuries received during capture resulted in the death of some lesser electric rays between 1 and 3 d after capture. These animals could usually be identified by curling of the body margins and pale body coloration. Injured animals did not bury or forage and were removed from the system upon death.

During the study, adult mortalities, exclusive of deaths due to equipment failures and injuries received during trawling, were reduced from an initial level of 58% down to 6% (Figure 1). Mortality from trawling injuries was variable, ranging from insignificant after trips on calm seas to as high as 25% after trips on rough seas or to areas with high concentrations of stinging jellyfish.

Several alterations in the holding procedures were instituted in response to problems that arose. At the beginning of the project, animals swam repeatedly into the cement tank walls, resulting in massive abrasions and death within a few days of capture. After the installation of the plastic tank liners in April 1985, animals readily avoided tank walls. Later use of fiberglass liners proved equally effective.

Animals held longer than 30 d developed extensive inflammation on the ventral surface and died within an additional 2–3 weeks. Specimens

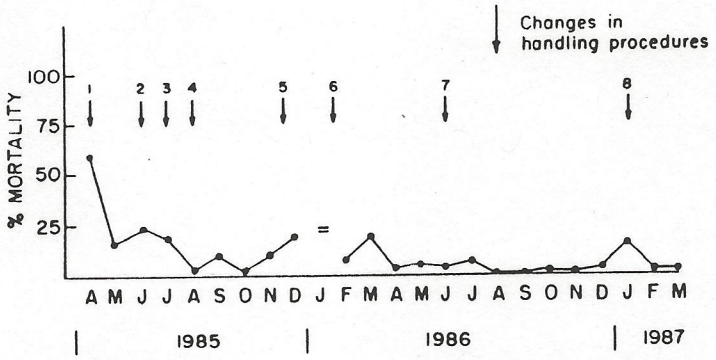

FIGURE 1.—Mortality of lesser electric rays larger than 15 cm total length in tanks, exclusive of deaths due to mechanical system failures or to injuries received during capture. Changes in holding conditions included (1) installation of tank liners, (2) appearance of disease, (3) start of regular feeding, (4) start of antibiotic treatment, (5) drop in water temperature, (6) installation of heating system, (7) provision of fine sand substrate, and (8) temperature fluctuations over a 5°C range.

submitted for pathological examination to the University of Maryland had multiple bacterial infections. Gills also became heavily infested with parasitic trematodes (*Amphibdelloides narcine*) and were destroyed if left untreated. To counteract this problem, the antibiotic and formalin dip was instituted in August 1985. Regular feeding was begun in July 1985.

After tank seawater temperatures dropped below 20°C in November and December 1985, lesser electric rays became sluggish. Anal bleeding and death occurred at approximately 14°C. Heating with electric submersion heaters was instituted in February 1986. With massive infections controlled, it became apparent that abrasions and inflammation of the gill and mouth areas remained a problem. The coarse-grained, unsorted sand that had been provided as substrate was replaced with fine-grain, wind-sorted sand collected from the top of a sand dune in July 1986; fewer cases of inflammation and lower mortality resulted. Temperature fluctuations of approximately 4–5°C/week occurred in January 1987 when a new heating system was brought on line, and mortality temporarily increased during this time (Figure 1).

Feeding

Gut contents of 51 field-collected lesser electric rays were examined at the beginning of the study. All fish had fed predominantly on burrowing polychaetes, and vermiform burrowing sipunculid worms or worm eels (family Ophichthidae) occurred in gut contents occasionally. An additional 10 newborn lesser electric rays had eaten juvenile polychaetes, anemones, nematodes, and burrowing amphipods.

Four species of polychaetes (*Diopatra cuprea, Onuphis magna, Clymenella torquata,* and *Neanthes arenacodentatia*) were tested as potential food sources for captive lesser electric rays, but they proved impractical because of their size and limited availability. Efforts to use frozen fish slices or any other dead food were totally unsuccessful. The polychaete species, *Glycera dibrachiata, Nereis virens,* and *Arenicola cristata* (bloodworms, clam worms, and lugworms, respectively), were then obtained from commercial bait dealers. *Glycera dibrachiata* was highly attractive to the lesser electric rays but proved unable to acclimate to the warm temperatures required by the fish. *Arenicola cristata* and *Nereis virens* were acceptable to adult fish. Worms were 10–20 cm long, lived in the substrate, and were readily fed upon.

Lesser electric rays nevertheless, became emaciated and died after approximately 60 d in captivity. Such fish always had empty guts and appeared to have starved despite the availability of prey species. The sand substrate above the tank filter beds was originally 30 cm deep. The sand, unlike the sea floor, remained oxygenated from top to bottom due to the action of the subgravel filters, and the prey species burrowed to the bottom of the sand layer and into the filter. When this was discovered, filter cloth was installed above the filter, and the sand depth was reduced to approximately 6 cm. This kept prey species near the surface and accessible to feeding predators.

Emaciation persisted among lesser electric rays smaller than 25 cm total length, however, because purchased worms were too large to be the sole food for these fish. Commercially available earth-

worms were tried, but these died within 5 min of being placed in salt water and were not eaten by the fish. Using the methods of D'Asaro and Chen (1976), we cultured *Arenicola cristata* and produced juvenile lugworms 2–6 cm long in 45–60 d. Smaller lesser electric rays readily fed upon worms of this size. However, when small fish were allowed to mingle freely with larger fish, they continued to starve despite the presence of small cultured lugworms in the substrate.

Small lesser electric rays and their smaller food were then isolated in 0.7-m × 1-m × 0.7-m enclosures formed of 2.5-cm × 1.2-cm-mesh screening. The enclosures contained Filterweave filter cloth and fine-grain sand substrates, and they were held within the larger fish tank. This confined the small lugworms in a small area, and they were more easily accessible to young fish. The system supported up to 10 juvenile fish (<18 cm) per cage. Prey were stocked at a rate of 2 worms/fish daily.

A small oligochaete, *Pontodrilus bermudensis*, was also tested as a food source for juvenile lesser electric rays. This saltwater-tolerant earthworm lives in and beneath dead sea grass that washes up on calm beaches in the tropics and subtropics (Lasserre and Erseus 1976) and occurs 2–20 cm below the surface. It was readily fed upon by small lesser electric rays and was available in varying amounts year-round in the study area. Small animals were estimated to each consume approximately eight small earthworms daily. *Pontodrillus bermudensis* was not practical as a sole food source for small lesser electric rays due to its small size (3 cm in length, 2 mm in diameter) and the amount of labor required to dig sufficient quantities. However, it was a useful supplement whenever small lugworms were temporarily unavailable.

Feeding Behavior

Captive lesser electric rays were active during darkness, remaining buried in daylight hours. Most feeding occurred between 2100 and 0100 hours. The fish preyed upon live, active polychaete worms, locating and attacking the worms when they were several centimeters below the surface of the substrate. The fish did not attack nonliving or nonvermiform items and often were unable to detect and attack live polychaetes that were not at least partially buried. Worms that were inactive, or moving weakly and burrowing slowly, appeared to be less attractive to lesser electric rays than vigorously moving worms.

The introduction of chemical stimuli, such as water in which worms had been held, appeared to elicit exploratory behavior and swimming. Fish then settled to the bottom and began searching. While seeking prey, they moved slowly over the substrate surface, propelling themselves with the pelvic fins. They paused periodically, positioned themselves over one particular spot, and began to flutter the posterior pectoral fin margins. A fish sometimes repositioned itself slightly and continued fluttering for periods ranging from a few seconds to 5 min. It then struck by lunging slightly forward and down, protruding its tubular mouth, and seizing the worm below the surface. Ingested sand was expelled through the spiracles. The fish then swam off the bottom and swallowed the worm in several gulps, after which it resumed searching. Individuals were observed to eat five or six large worms (8–10 cm long) in one 3-h period after being held for several weeks without food. At the termination of activity, lesser electric rays buried themselves in the sand by rocking back and forth, vigorously fluttering the body margins to dig a shallow pit. They often remained partially buried with eyes and spiracles uncovered. The behavior was fully developed in newborn captive fish, which began hunting immediately after birth.

Active lesser electric rays often interacted during feeding periods, resting on the substrate with body margins overlapping. The fish initiating the contact typically approached a stationary fish and lay its anterior margin over the lateral margin of the other fish. If the latter fish moved, which was unusual, the first often followed, actively maintaining or reestablishing the contact. Such contacts were observed between two males, two females, or male and female. They were also observed between two adults and between a juvenile and adult. In a juvenile–adult interaction, the juvenile usually sought contact with the adult. Juvenile–juvenile interactions were rarely observed.

Discussion

The techniques described allow lesser electric rays to be maintained in relatively good health in the laboratory for several months or more. The species' inshore distribution, relative ease of capture, and now susceptibility to culture should permit it to be made routinely available to neurological laboratories on a dependable basis.

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