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Robert A. Fisher
Virginia Institute of Marine Science

Eric M. Stroud
SharkDefense LLC

Michael M. Herrmann
SharkDefense LLC

Patrick H. Rice

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An Evaluation of the Behavioral Responses of Cownose Rays (*Rhinoptera bonasus)* to Permanent Magnets and Electropositive Alloys

Completed by

Robert A. Fisher
Virginia Institute of Marine Science
College of William and Mary

Eric M. Stroud and Michael M. Herrmann
SharkDefense LLC

Patrick H. Rice
Consulting Marine Biologist

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Introduction

The cownose ray, *Rhinoptera bonasus* (Family: Myliobatidae), is a demersal elasmobranch inhabiting the coastal waters of the Eastern Atlantic from New England throughout the Gulf of Mexico and as far south as northern Brazil (Robins et al. 1986). They are most easily identified by the blunt, squarish snout with a median indentation (Figure 1). Cownose rays feed on a variety of crustaceans and mollusks, including oysters and large schools have been reported migrating into brackish waters where they decimate commercial oyster beds (Merriner and Smith 1979).

The objective of this experiment was to determine if shark repellent technologies could be exploited to control cownose ray behavior. The potential use of these selective non-lethal repellent technologies involves the reduction of cownose Ray interactions with oyster beds in the Chesapeake Bay.

Fig. 1 - The cownose ray *(photo by R.A. Fisher)*

Overview of Repellent Technologies

Magnetics

Several species of elasmobranchs have demonstrated the ability to sense magnetic fields (Kalmijn and Holland, 1978; Bloch and Ryan 1980; Klimley 1993; Klimley et al 2002). The Ampullae of Lorenzini organ within elasmobranchs is used to detect weak electrical fields at short ranges. The detection range of this organ is effective only within inches, as sharks sense bioelectrical fields in the final stages of prey capture. The flux distance of a permanent Neodymium-Iron-Boride (NdFeB) magnet corresponds closely with the detection range of the Ampullae of Lorenzini. The magnetic field generated by these specific magnets decreases at the inverse cube of the distance from the magnet (Figures 2 and 3). Therefore, at distances of a few meters from the magnet, the field exerted is less than the Earth's magnetic field.

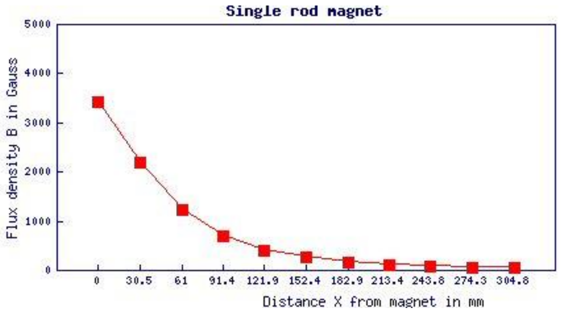

Fig. 2 - Flux density decreases with the inverse cube of the distance from the magnet surface. *Courtesy of International Magnetic Solutions.*

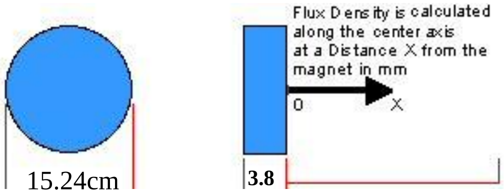

Fig. 3 - Flux density geometry for a 15.25cm diameter grade N38 permanent magnet. *Courtesy of International Magnetic Solutions.*

“Rare” earth magnets have been demonstrated to terminate tonic immobility (i.e. a comatose state in sharks induced by turning them upside-down; TI) in several shark species including lemon sharks (*Negaprion brevirostris*) and nurse sharks (*Ginglymostoma cirratum*). Once the comatose or “tonic” state in sharks is achieved, it is difficult to interrupt. Therefore, it is a technique often employed during surgical procedures on sharks (e.g. internal tag implantation). Our primary assumption for using TI bioassays to test repellent effects is that a treatment that interrupts TI suggests that it may be a good repellent.

Twenty sharks were evaluated with the tonic immobility protocol using large cylindrical permanent “rare earth” magnets (N48 grade; 4"diameter x 1.5" height). All sharks displayed “rousing” behavior (Figure 4) - fourteen sharks terminated tonic immobility with violent thrashing behavior, and six sharks exhibited a physical bending away from the magnet. To avoid responses from visual cues, shark eyes were shielded as the magnets were moved towards the snout (i.e. primary location of Ampullae of Lorenzini, Figure 6).. The effective range was 0.01-0.30 meters, causing the shark to experience a magnetic field of 50 Gauss or greater. For comparison, strong (30 Amp) and weak (6 Amp) electromagnets had no effect during tonic immobility on the same test subjects at the same ranges (Figure 6).

Fig. 4 – Violent rousing behavior of a juvenile *N. brevirostris* in response to a rare-Earth magnet.

Fig. 5 - The tonic immobility assay using a blinder, rule, and immobilized shark. The magnet is moved relative to the stationary shark, or vice versa.

Fig. 6- An energized 12VDC 30A electromagnet having no effect on an immobilized lemon shark.

An acrylic Y-maze was constructed to establish a preference test for several species of captive sharks. Sharks were allowed to enter and exit the maze without negative reinforcement if the correct path was chosen. For each trial, raw shrimp was used as the reward, and a 4"x1.5" cylindrical “rare” earth magnet (grade = N48) was used as negative reinforcement. In a series of trials reported at the July 2005 American Elasmobranch Society (AES) meeting in Tampa, Florida, SharkDefense reports that juvenile nurse sharks entering the trap of the maze (i.e. branch with the magnet) became very distressed and would not take any reward baits. Two nurse sharks clearly learned to avoid the magnet trap and take the reward baits during a period of 6 trials. Results were similar when the trap was placed in the other branch of the Y maze. During one of the replicate trials, a third nurse shark entered the trap and became highly distressed and needed to be rescued from the maze (Fig. 7). This shark never entered the maze again in subsequent trials. A juvenile lemon shark became highly distressed when approaching the junction towards the magnet, and did not take any rewards.

Fig. 7 - Two nurse sharks in the Y-Maze, with one nurse shark highly distressed in the magnetic trap. The other nurse shark has successfully navigated the maze.

Electropositive Alloys

During May 2006 trials at South Bimini, Bahamas, it was discovered that electropositive metals, particularly early-Lanthanide metals, also terminated tonic immobility and produced violent rousing behavior in tonic immobilized *N. brevirostris* and *G. cirratum*. This phenomenon was initially observed when a 113g ingot of 99.5% Samarium metal was presented to immobilized sharks.

Magnetic and induced electric current effects were suspected, however no magnetic field increase was detected around a Samarium ingot submerged in sea water within 0.1mG using a two-axis magnetometer and a separate calibrated milligauss meter. Additionally, a picoammeter and submerged 10μH coil failed to detect any induced currents within 10μA when the ingot was passed through the coil's diameter in seawater.

Many transition metals, Lanthanide metals, and metalloids were screened for repellency using tonic immobility bioassays. Attention was given to oxidation state, but no correlation was found. For example, 99% or greater purity ingots of Zirconium (oxidation state 4), Niobium (oxidation states 5, 3) Chromium (oxidation states 6, 4, 3, 2), Tungsten (oxidation states 6, 5, 4, 3, 2), and Rhenium (oxidation states 7, 6, 4, 2) all failed to illicit the desired rousing behavior during TI tests.

Certain Lanthanide metals (oxidation state 3) did produce violent responses. The strength of the avoidance behavior (i.e. repellent response) appeared to roughly correlate to the position of the metal in the Lanthanide series, with elements 57-64 showing more reactivity than elements 75-71.

During June 2006, a series of trials were conducted using juvenile *N. brevirostris* and *G. cirratum* wherein behavioral responses were assigned a score from 0 to 4, with 0 being no response and 4 being a violent rousing behavior (a score of 5 was used to describe extremely violent responses to 99.5% Nd metal). All metal ingots were secured to a 0.6m-long acrylic pole, which electrically insulated the tester from the metal ingot. Metal ingots were submerged and moved in a slow motion to a tonic immobilized juvenile shark's left sagittal region, starting at a distance of 1.0 m and moving toward to shark. Magnetic materials (a Barium Ferrite magnet ("ceramic") and a Samarium-Cobalt magnet ("SmCo")) were also evaluated. Diamagnetic pyrolytic graphite was used a control material. Results of all trials are shown in Fig. 8 and Fig. 9.

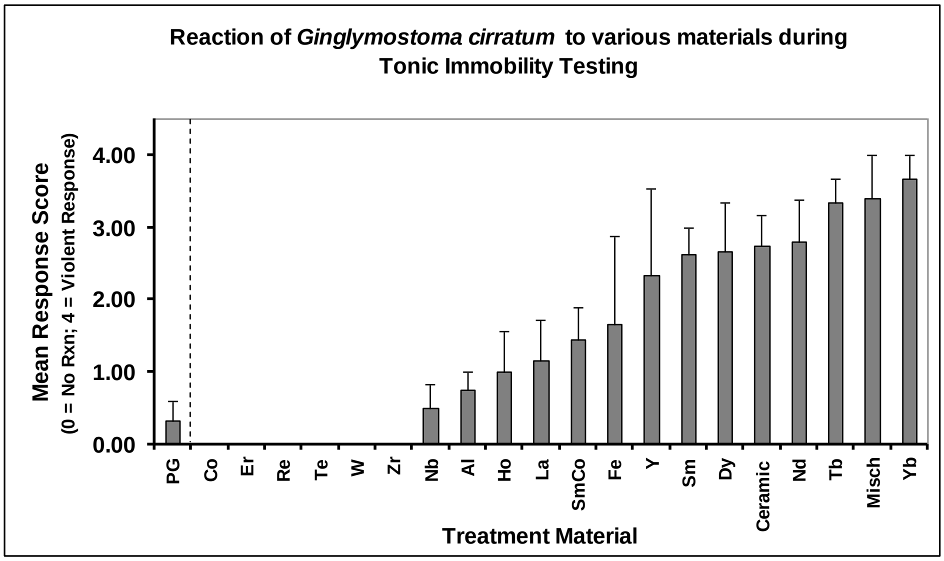

Fig. 8. Reaction of nurse sharks, *Ginglymostoma cirratum*, when exposed to various test materials (chemical element symbols) during tonic immobility. PG = pyrolytic graphite, Co = cobalt, Er = erbium, Re = rhenium, Te = tellurium, W = tungsten, Zr = zirconium, Nb = niobium, Al = aluminum, Ho = holmium, La = lanthanum, SmCo = samarium cobalt, Fe = iron, Y = yttrium, Sm = samarium, Dy = dysprosium, Ceramic = barium-ferrite ceramic magnet, Nd = neodymium, Tb = terbium, Misch = cerium misch metal (lanthanide alloy), Yb = ytterbium.

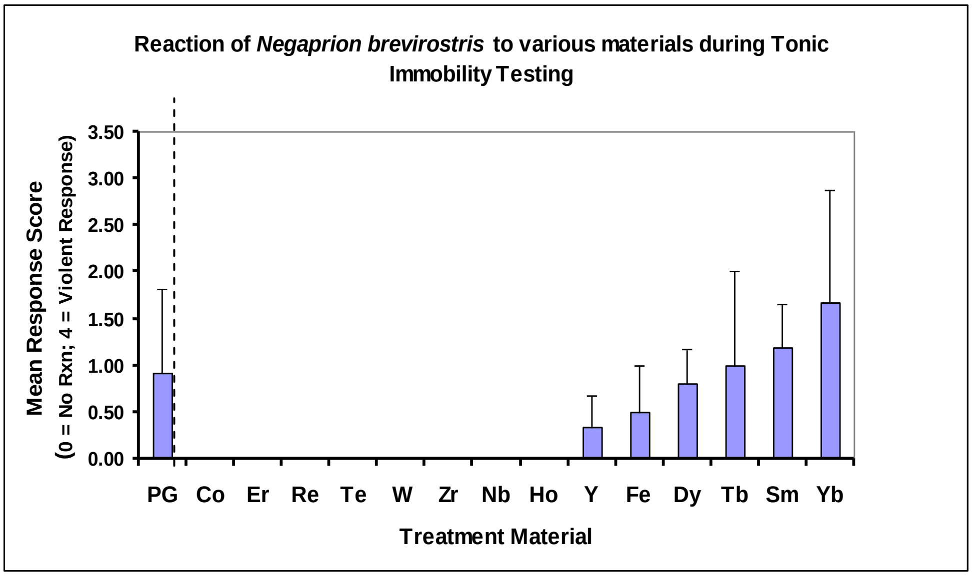

Fig. 9. Reaction of lemon sharks, *Negaprion brevirostris*, when exposed to various test materials (chemical element symbols) during tonic immobility. PG = pyrolytic graphite, Co = cobalt, Er = erbium, Re = rhenium, Te = tellurium, W = tungsten, Zr = zirconium, Nb = niobium, Ho = holmium, Y = yttrium, Fe = iron, Dy = dysprosium, Tb = terbium, Sm = samarium, Yb = ytterbium.

Based on these results, SharkDefense hypothesizes that electronegativity directly correlates to the repellent response. Electronegativity describes a measure of the ability of an atom or molecule to attract electrons in the context of a chemical bond. The Pauling scale is used to describe electronegativity, where Fluorine, the most electronegative element is 3.98, and Francium, the least electronegative element, is 0.7. We propose that metals such as Ytterbium (Pauling=1.1) and Neodymium (Pauling=1.14) will produce a greater response than Dysprosium (Pauling=1.22) and Samarium (Pauling=1.17). Cerium Mischmetal has an average Pauling electronegativity of 1.1-1.15, depending on alloy composition. These results must be considered with caution due to a small sample size.

The electronegativity hypothesis may produce a synergistic repellent effect in rare-Earth magnets. The primary repellent action is caused by electromagnetic induction creating an electric field which stimulates the Ampullae of Lorenzeni as the shark approaches an increasingly powerful permanent magnetic field surrounding the magnet. Within the same range, the highly electropositive Neodymium metal present in the magnet's sintered core adds to the repellent effect, but only minimally – The sinter's exposure to seawater is limited by a thin nickel coating on the magnet.

Preliminary experiments indicate that higher electropositive metals, such as Calcium (Pauling=1.00) and Strontium (Pauling=0.95) are even more potent repellents, but these metals have a limited life (1-2 hours) in seawater. Metals with higher electropositivity than Strontium are too reactive for use in seawater and safe handling.

An alternative to high priced pure lanthanide metals are alloys of early-lanthanide metals, particularly Neodymium-Praseodymium alloy, which offer benefits of high electropositivity, machinability, and somewhat stronger corrosion resistance than the pure lanthanide metal component in seawater.

Methods

An evaluation of magnetic and electropositive repellent technologies on captive cownose ray (*Rhinoptera bonasus)* behavior was conducted at the Virginia Institute of Marine Science, Gloucester Point, VA, on October 23, 2006, under the direction of Mr. Robert Fisher. Mr. Eric M. Stroud from SharkDefense LLC supplied the experimental repellent devices and assisted in the execution of the experiment.

Experimental Design

An outdoor above-ground fiberglass oblong captive specimen tank with sand filter recirculation was used to support a population of four cownose rays, two spiny butterfly rays (*Gymnura altavela),* and one clearnose skate (*Raja eglanteria).* The cownose ray population consisted of one adult female, and three juveniles. The butterfly rays and the clearnose skate were not utilized in this experiment. Water temperature prior to experimentation was 27.7°C and the salinity was 19 ppt. A wide-angle color video camera was secured above the tank in for the remote monitoring of specimen interactions with bait cages. Bait cages were constructed by securing two 15cm X 20cm panels of black, polyethylene plastic 12.7mm square aquaculture mesh with plastic zip-ties along 3 of the four sides, resulting in cages resembling envelopes with the un-tied side allowing bait and/or magnet/metal alloy to be inserted.

The interaction of the adult cownose ray with a bait cage containing a large permanent rare-Earth magnet (no bait) was observed. A 15.24 cm diameter, 3.81cm thick grade N38 cylindrical Neodymium-Iron-Boride permanent magnet with a Nickel exterior coat was utilized as the first repellent treatment. This magnet exhibits a flux density (residual induction, B_r) of 38 MGOe. The surface of this magnet produces a flux per unit area (magnetic induction, B) of 0.9T (9,000G), as measured with a calibrated teslameter (F. W. Bell). The flux is reduced exponentially (inverse cube) with distance from the magnet's surface, thus, an approaching ray may experience greater than 100G at a distance of less than 0.1m from the magnet's surface.

Additionally, the interaction of the adult cownose ray with a bait cage containing electropositive metal alloys (no bait) was observed. A 400g ingot of Cerium-Lanthanum Mischmetal (Pauling Electronegativity =1.12) and a 500g ingot of Neodymium-Praseodymium Mischmetal (Pauling Electronegativity=1.135) were used simultaneously. We hypothesize that the sensory detection range is the same for electropositive metal alloys as they are for permanent magnets.

Repellency was quantified by counting the number of protected baits and unprotected baits eaten by the adult female cownose ray, and the amount of time spent dwelling at a bait cage. The adult female ray was selected because it previously exhibited the most aggressive feeding behavior in the population. For all comparative trials, a single live blue crab (*Callinectes sapidus*) was cut in equal halves (anterior-posterior) with one half used with control cage, the other with treatment cage. Trials were conducted by simultaneously placing control and treatment cages into ray holding tank 1.5 meters apart (Figure 10, 11).

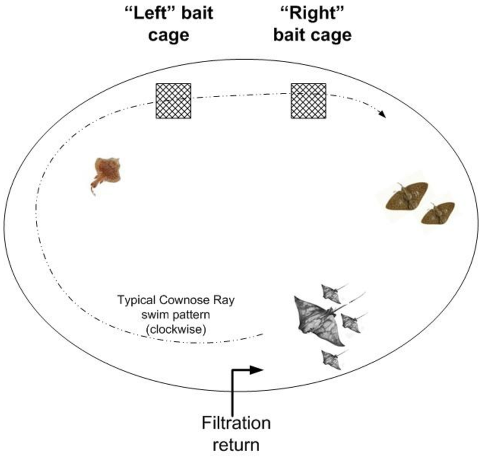

Fig. 10 - Aerial view of the specimen tank, illustrating the position of the bait cages.

Fig. 11 - Robert Fisher (right) and Eric Stroud with two submerged bait cages, visible in the left foreground.

Temporal variations were reduced by simultaneously presenting both bait cages into the tank, and conducting all trials within 6 hours in the same day. A trial consisted of placing two bait cages in the specimen tank, and allowing enough time for an interaction to occur. At the conclusion of a trial, both cages were simultaneously removed, and new, fresh bait was replaced for next trial. External interference by the researchers was minimized by remaining away from the tank edge during trials. To reduce complications arising from learned avoidance behavior, the position of the treatment and the control cages was alternated at least once.

The primary interaction of the adult female cownose ray with empty bait cages was observed in Trial 1. Two empty bait cages were simultaneously lowered into the tank and the time at which the ray passed over each cage, as well as the direction of swimming, was recorded.

The interaction of the female adult cownose ray with baited cages was observed in Trial 2. Both cages, baited with blue crab, were simultaneously lowered into the tank, and the swim pattern and dwell time at each cage was recorded. The dwell time was calculated as the difference in time the adult ray's head was over the cage, to the time the adult ray's head was no longer in proximity to the cage.

In Trial 3, the permanent magnet was secured inside of the left cage along with fresh blue crab. The right cage received only blue crab and served as the experimental control. Both cages were simultaneously lowered into the tank. Trial 4 involved utilizing two controls was initiated immediately after the magnet treatment trial concluded. Two cages were simultaneously lowered into tank, each containing fresh blue crab. In Trial 5, as in the third trial, we introduced the permanent magnet in the left cage along with blue crab in both cages. In Trial 6, two controls were introduced immediately after trial 5 concluded. Two cages were simultaneously lowered into tank, each containing fresh blue crab. In Trial 7, we replaced the permanent magnet in the left cage. Baiting of cages was changed for this trial to allow for predation success. Fresh blue crab halves were secured to the outside top panel of cage by short tether (10cm) using monofilament fishing line.

We placed a 500g ingot of highly electropositive Neodymium-Praseodymium alloy along with a 400g ingot of highly electropositive Cerium Mischmetal in the left cage for Trials 8 and 9. Again, one half of a fresh blue crab was tethered to each bait cage to allow for predation success.

Results

Trial 1 – Control Swim Behavior

It was previously observed that the rays swam close to the tank wall, therefore, bait cages were deliberately placed near the tank wall in order to increase the likeliness of an interaction.

For a trial lasting 48 minutes 37 seconds, we report that the adult ray passed equally over both bait cages a total forty-five (45) times, in a clockwise direction.

The mean round trip cycle from the right cage back to the left cage along the interior tank wall was 66 ± 0.2 seconds. Only one (1) counterclockwise swimming event was observed in this trial, but the ray resumed clockwise swim behavior within 34 seconds. The three juvenile rays displayed schooling behavior with the adult ray for the entire trial. We therefore assign "normal" behavior as a clockwise swimming pattern, completing one cycle along the interior tank wall at an average of 1.1 minutes with schooling behavior.

Trial 2 – Controls

For a trial lasting 19 minutes and 17 seconds, we report sixteen (16) clockwise passes over both bait cages and one (1) single pass only over the left cage occurred. One (1) counterclockwise swim pattern occurred after passing over the left cage just prior to passing over the right cage. The juvenile rays again displayed schooling behavior with the adult ray for the entire duration of this trial.

In exactly half of all passes over both cages, the adult ray was observed to descend upon and remain at a bait cage, attempting to access the bait. This investigatory behavior was quantified by an increase in dwell time at the cage. At the left cage, investigatory behavior resulted in a mean dwell time of 19.3 ± 7.6 seconds (n=3) versus a casual pass over the cage with a mean dwell time of 3.9 ± 1.1 seconds (n=14). At the right cage, investigatory behavior resulted in a mean dwell time of 18.4 ± 10.1 seconds (n=5) versus as usual pass over the cage with a mean dwell time of 4.2 ± 1.6 seconds (n=11). We therefore assigned a dwell time of 10 seconds or greater as a discriminator for passes versus investigations in subsequent trials (Figure 12).

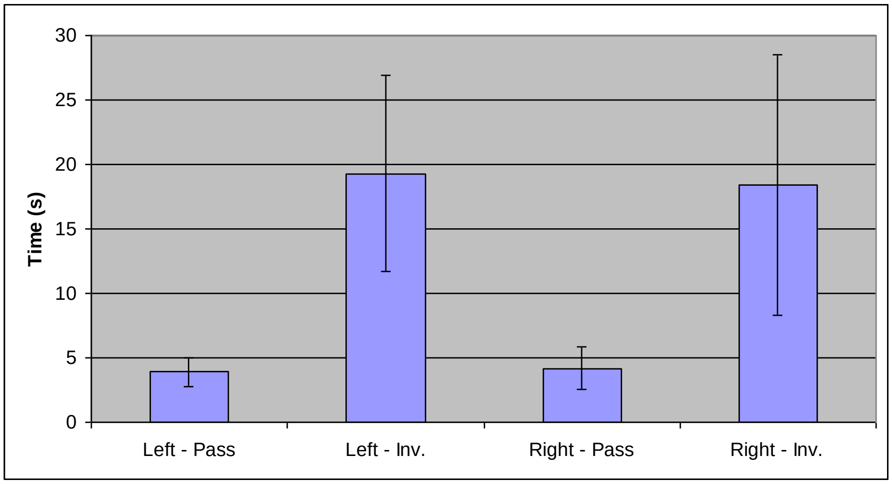

Fig. 12 – Dwell time at two baited cages without repellent treatments (left and right) distinguishing passes ("Pass)" and bait investigations ("Inv.") by an adult female *R. bonasus.*

Trial 3 – Magnetic Treatment

Upon the first pass over the left cage, an alteration in swim behavior was readily apparent. The adult ray broke its circular swim pattern along the tank wall and changed direction to the opposite side of the tank. In this 13 minute 52 second trial, only five (5) passes over both cages was observed. Nine (9) counterclockwise swim patterns occurred during this trial, and schooling behavior was noticeably disrupted for much of the trial. No investigations occurred at either cage. The right cage was approached three (3) times by the adult ray without first passing over the treatment cage. The mean dwell time at the left (treatment) cage was 4.3 ± 1.4 seconds, and the mean dwell time at the right (control) cage was 3.0 ± 1.1 seconds, well below our established 10 second threshold for a bait investigation.

Trial 4 – Controls

Clockwise swim patterns and schooling behavior resumed quickly, and bait investigations resumed on the first pass. For a duration of 16 minutes 37 seconds, we report thirteen (13) passes over the left cage, with eleven (11) of these subsequently including a pass over the right cage. There were four (4) investigations at the left cage, with a mean dwell time of 12 ± 0.8 seconds, and two (2) investigations at the right cage, with a mean dwell time of 17 ± 2.8 seconds.

Trial 5 – Magnetic Treatment

For duration of 14 minutes and 19 seconds, we report fifteen (15) passes over the left cage and twelve (12) of these also included a pass over the right cage. Two (2) investigations occurred on the right (control) cage, with mean dwell time of 15 ± 5.7 seconds. No investigations occurred at the left (treatment) cage.

In contrast to the Third Trial, schooling behavior was only momentarily disrupted in this trial. Two (2) counterclockwise swim patterns were observed. Increases in altitude were observed as the adult ray swam near the magnetic followed by a gradual decrease in altitude as the control was approached (Fig. 13). Further studies and quantification of this observed effect employing underwater cameras are warranted.

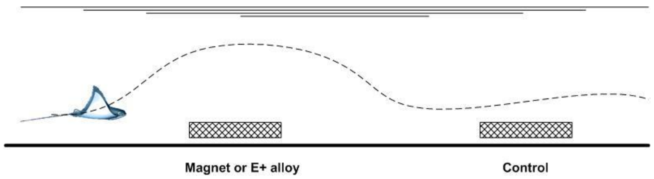

Fig. 13 – Change of swim depth (altitude) observed in *R. bonasus* in response to a permanent rare-Earth magnet or an electropositive alloy. Further study is needed to quantify this behavior.

Trial 6 – Controls

For duration of 13 minutes and 10 seconds, we report four (4) passes over the left cage only, one (1) pass over the right cage only, and nine (9) passes over both cages. One (1) investigation occurred at the right cage, lasting 10 seconds. No investigations occurred at the left cage. Two (2) counterclockwise swim patterns were observed during this trial, and schooling behavior was consistent.

Trial 7 – Tethered Bait with Magnetic Treatment

For duration of 9 minutes and 53 seconds, we report seven passes over the treatment cage (left). One investigation occurred at the right cage on the first pass and the bait was readily consumed, giving a dwell time of 63 seconds. The tethered bait on the treatment cage remained undisturbed for the duration of the trial. Schooling behavior remained intact during this trial, and no counterclockwise swim patterns were observed.

Trial 8 – Tethered Bait with Electropositive Alloy Treatment

For duration of 14 minutes, we report seven (7) passes over the left (treatment) cage. One (1) investigation occurred at the right cage on the second pass and the bait was readily consumed, giving a dwell time of 72 seconds. The tethered bait on the left cage remained undisturbed for the duration of the trial. Schooling behavior remained intact during this trial, and no counterclockwise swim patterns were observed.

Trial 9 – Tethered Bait with Electropositive Alloy Treatment, Reversed Position

The ninth trial was conducted in a similar manner as Trial 8, except that the position of the control and the treatment was reversed. For duration of 5 minutes and 21 seconds, one investigation occurred at the left cage on the first pass, lasting 34 seconds. The tethered bait on the right cage remained undisturbed for the duration of the trial. Schooling behavior remained intact during this trial, and no counterclockwise swim patterns were observed.

Graphs

Figures 14, 15, 16, and 17 summarize the total investigation times for experimental trials.

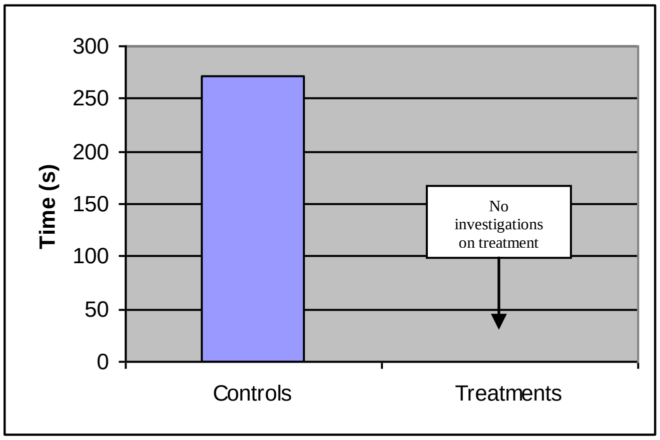

Fig. 14 – (Trials 2, 3, 4, 5, and 6) Total time spent investigating *Callinectes sapidus* bait secured inside of a bait cage by an adult female *R. bonasus* across five trials. An investigation is defined as a period of 10 or more seconds spent at a bait cage by the ray. Treatment consisted of a Neodymium-Iron-Boride grade N38 cylindrical magnet.

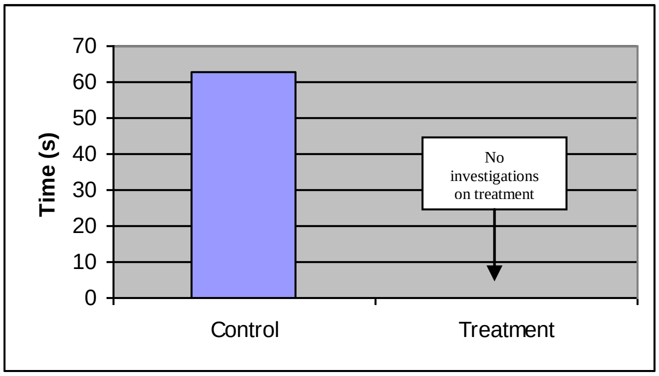

Fig. 15 – (Trial 7) Total time spent investigating *C. sapidus* bait tethered outside of a bait cage by an adult female *R. bonasus* for one trial. An investigation is defined as a period of 10 or more seconds spent at a bait cage by the ray. Treatment consisted of a Neodymium-Iron-Boride grade N38 cylindrical magnet secured inside of one bait cage.

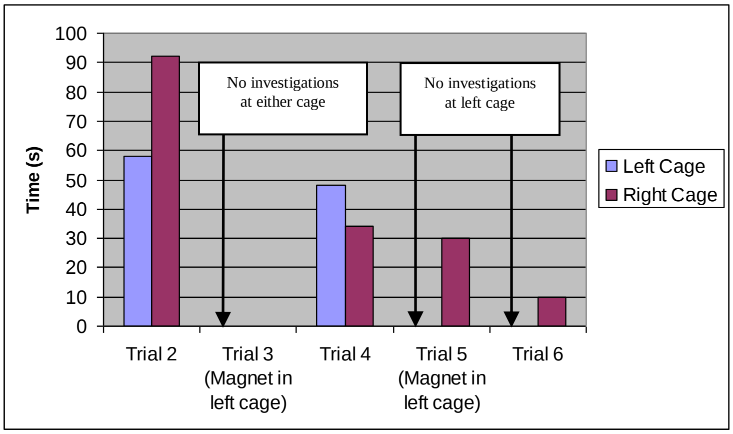

Fig. 16 – Total investigation time spent at the left cage and right cage for *C. sapidus* bait secured inside of a bait cage by an adult female *R. bonasus* across five trials. An investigation is defined as a period of 10 or more seconds spent at a bait cage by the ray. Treatment consisted of a Neodymium-Iron-Boride grade N38 cylindrical magnet secured inside of one bait cage.

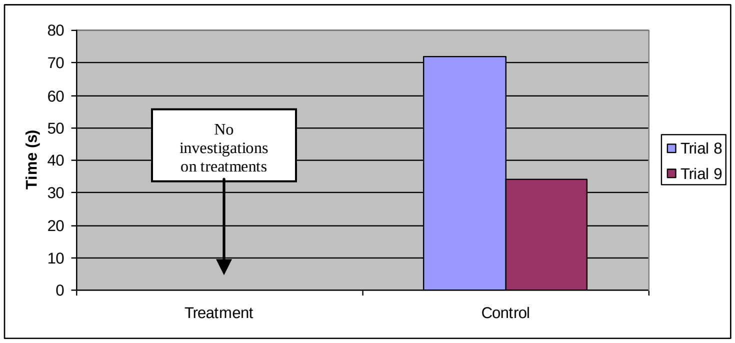

Fig. 17 - (Trials 8 and 9) Total investigation time spent at the control and treatment cages using *C. sapidus* bait tethered outside of a bait cage by an adult female *R. bonasus* across two trials. An investigation is defined as a period of 10 or more seconds spent at a bait cage by the ray. Treatment consisted of a 400g ingot of Cerium -Lanthanum Mischmetal and a 500g ingot of Neodymium-Praseodymium (Pr) alloy secured inside of one bait cage.

Discussion of Results

Preliminary data suggests that the number of bait investigations by the adult female cownose ray is appreciably reduced when a permanent magnet or an electropositive alloy was utilized near the bait.

Tethering the bait outside of the cage provided a positive reward for the ray. Whereas trials 2, 3, 4, 5 and 6 did not allow the ray to consume the bait, trials 7, 8, and 9 allowed this and represented a more stringent test.

The amount of time spent at the each cage (left and right) also suggests that interactions are reduced when a permanent rare-Earth magnet is in proximity to the bait (see Fig. 16). Trials 2, 4, and 6 did not utilize the permanent magnet, thus, interactions with both the left and right cages was expected. In trial 6 however, no investigations occurred at the left cage. This may evidence negative conditioning of the ray at that location. Trials 3 and 5, which utilized the permanent rare-Earth magnet in proximity to the bait, demonstrated no investigations, as expected.

The trial 6 produced somewhat unexpected results, as only one investigation occurred at the right cage despite the lack of any repellent treatments in the tank. At this point in the overall experiment, no positive reward had been given to the rays. All bait was secured inside of the cage, which prevented the ray from eating it. We decided that subsequent trials would use baits tethered outside of the cages as a positive reward.

Electropositive alloys appear to produce the same desirable repellent effect as the permanent magnet. The use of the electropositive alloys appreciably reduced the total interaction time (Fig.17).

Conclusions

Preliminary data suggests that an adult female *R. bonasus* displays a strong preference to bait cages that do not employ a permanent magnet or electropositive alloys in close proximity to the bait. Further experimental trials using larger populations of adult specimens are encouraged to allow for descriptive statistics.

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