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Research report

Electrosensory interference in naturally occurring aggregates of a species of weakly electric fish, *Eigenmannia virescens*

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Abstract

The detection and identification of behaviorally relevant signals in the presence of competing signals in the environment is a major challenge of animal sensory systems. In weakly electric fish such as *Eigenmannia virescens*, the interactions between the autogenous electric field and the electric fields of nearby conspecifics can have profound effects on the perception of other behaviorally relevant electrosensory information. To better understand the natural signals that the nervous system of *Eigenmannia* experiences during the processing of electrosensory information, we examined the electrosensory milieu of *Eigenmannia* in the wild and in the laboratory. Recordings of the electric fields of *Eigenmannia* were made in 'black' and 'white' waters near the Napo River in eastern Ecuador. Fourier analysis revealed that *Eigenmannia* typically experience the electric fields of three to five conspecifics during the day and night in each habitat. The median difference in electric organ discharge frequencies between nearby *Eigenmannia* during the day was 23 Hz in black water habitats, 41 Hz in white water, and 37 Hz at night in both habitats: these signals are known to activate tuberous electroreceptors and downstream CNS circuits. There was no correlation between the number of individual *Eigenmannia* detected at recording sites and electric organ discharge frequencies. Further, *Eigenmannia* apparently do not maximize the frequency differences between conspecifics. In laboratory studies fish were preferentially observed in aggregates of two fish or more. Aggregate sizes observed in the laboratory were similar to those in the wild.

Keywords: Shoaling; Jamming avoidance response; Amazon Basin; Signal detection

1. Introduction

Animal sensory systems are subject to a wide range of interfering and competing signals that can have profound effects on the function of sensory systems [3,7,9,20]. Animal behavior is often adapted to reflect the challenges of signal detection in noisy environments [1,23,26,31,32]. The wave-type weakly electric fish *Eigenmannia virescens* is a particularly good model system to study problems related to sensory interference as these fish experience different electrosensory conditions when they are alone versus when they are in close proximity to one another [4,18,39]. These electrosensory conditions can dramatically modulate the activity of CNS neurons, altering information available for biologically relevant computations [11,30,33].

Eigenmannia produce a continuous, nearly sinusoidal electric field at frequencies between 200 and 700 Hz. This electric field is generated by a specialized electric organ; the field is known as the electric organ discharge, or EOD. When individual *Eigenmannia* are in close proximity to conspecifics, the electric fields interact and produce an emergent pattern of 'beats' that occur at rates equal to the frequency difference between the fish. The frequency of

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beats is known as the 'beat rate'. Electrosensory function is maximally impaired in *Eigenmannia* when the fields of nearby conspecifics are within 3–8 Hz [4,17,30]. In contrast, beat rates of more than 15 Hz do not significantly impair electrosensory function [4].

Eigenmannia exhibit a specialized behavior, the 'jamming avoidance response' (JAR), to avoid detrimental beat rates. In the JAR, the fish with the higher EOD frequency increases its frequency while the other fish decreases its EOD frequency [39]. The difference in EOD frequencies between the fish is thereby increased, resulting in beat rates that do not impair either fish's ability to electrolocate [4]. The net result of the JAR behavior is that both fish will continue to experience ongoing beat rates above 15 Hz for as long as the fish remain in proximity of each other.

Interestingly, these ongoing post-JAR beat rates appear to have an ongoing impact on electrosensory processing. For example, beat rates of 15 Hz or more have been shown to modulate short-term synaptic plasticity in midbrain neurons in the torus semicircularis (electrosensory midbrain) [11,36]. Short-term synaptic plasticity is used in the generating of low-pass filtering properties [10,11], and may be critically involved in the generation of direction selectivity [6]. Therefore, it is likely that the processing of salient electrosensory information in these midbrain neurons is different when *Eigenmannia* are alone versus in groups [34]. Additionally, neurons in the electrosensory lateral line lobe (ELL) of *Apteronotus*, a closely related genus, exhibit high-pass temporal filtering properties for 'global' stimuli, like those that result from the JAR. In effect, the response properties of ELL neurons appear to preferentially pass information resulting from the JAR behavior to higher stations in the brain [5].

The potential effects of the JAR behavior on electrosensory processing are particularly interesting in light of the availability of alternative behavioral solutions to the jamming problem. For instance, the jamming problem can be avoided in *Eigenmannia* by simply moving away from conspecific fish. Which strategy does *Eigenmannia* commonly employ for jamming avoidance? Despite intensive study of the JAR and other electrosensory behaviors in *Eigenmannia*, there are only anecdotal reports concerning the strategies for jamming avoidance that are used by these fish under natural conditions [21,24,40].

The present study examines a specific aspect of the social behavior of weakly electric fish that has direct implications for electrosensory processing in the ELL and in the midbrain: how many conspecific electric fields, if any, do *Eigenmannia* commonly experience? That is, how many individuals are located in close enough proximity to affect and to be affected by conspecific electric fields? We examined the distribution of 'electrosensory' aggregates or shoals of *Eigenmannia* in natural habitats and in naturalistic experiments in the laboratory. The goal is to determine the ongoing electrosensory conditions that fish experience in the natural environments to better understand how the CNS processes behaviourally relevant electrosensory information.

2. Materials and methods

2.1. Study sites

Fish were studied in both "black water" and "white water" habitats near the Napo River in eastern Ecuador. Black water habitats are tannin-rich, clear water from local rain runoff. White water is characterized by low visibility and high silt loads from mountain runoff. Observations were made over a 3-year period: January of 2003, 2004, 2005, and August of 2004. Black water recordings of EODs were made in and around Lake Pilchicocha, Orchidea creek, and other streams in the privately held Sacha Lodge reserve. White water recordings of EODs were made in the Tiputini River within the Yasuní National Park near the Estación Ciéntifica Yasuní (PUCE). Conductivity of water at each habitat was between 10 and 50 μS/cm. *Eigenmannia* were commonly found in reed-grasses and in the root systems of many floating plants. They were also found in leaf litter and large debris, such as fallen trees, in moving streams. Water depths ranged from 0.5 m in the shallow streams to over 5 m depth in larger rivers and lakes. *Eigenmannia* were not commonly found in igapó (flood plains in black water areas) and varesa (flood plains in white water areas) habitats, although other gymnotiform species were encountered (particularly pulse-type species of *Gymnotus* and *Brachyhypopomus*).

These studies were conducted with approval of the Ministerio del Ambiente, the owners of Sacha Lodge, and the Pontificia Universidad Católica del Ecuador. All procedures were approved by the Institutional Animal Care and Use Committee of the Johns Hopkins University, and follow international standards for animal use by the Society for Neuroscience. Samples of fish examined in this study have been added to the collections of the Museo de Zoología de la Pontificia Universidad Católica del Ecuador.

2.2. Amplification and recording of EODs in Ecuador

Recordings of electrical activity were made using two systems: portable and stationary. For the portable system, differential recordings were obtained from two wire leads, 10 cm apart, on wood, plastic, or fiberglass rods. Probes were submerged 10–50 cm into the water for each recording. Electric signals were amplified using either a BMA-200 (Charles Ward Equipment, Inc., Ardmore, PA) or a mini audio amplifier #277-1008 (Radio Shack Corp.). Signals were captured using consumer MP3 encoders. These included a TASCAM Pocketstudio V, PogoProducts RipFlash+, and Creative MuVo N200. Each of these devices was used in its highest recording quality and fastest encoding rate. Extensive testing in the laboratory was conducted to ensure that the MP3 encoding process, which is a "lossy" process, did not impact the data in any substantive way. We found, as is the case for highest-quality JPEG images, that the effects of the transform were not easily discernable.

Recording sites were chosen by the presence of EODs in the range of frequencies produced by *Eigenmannia* (see below). EODs could be detected up to about 1.5 m from individual *Eigemannia*. Sites were typically more than 2 m apart, although a few samples were taken at approximately 1 m distance. These distances were chosen to avoid resampling of individual fish between adjacent recording sites. Recordings used in this study were from sites of no more than 2 m depth. Although we did not attempt an exhaustive search of deeper habitats, no weakly electric fish signals were encountered when we made observations from greater depths of up to 4 m below the surface.

Samples were a minimum of 20 s duration. Samples were taken in wide variety of locations in all habitats where the fish were encountered. During the day, sites were identified using the electric field amplifier system alone, as the fish were not visible. At night, either the amplifier system or flashlights, particularly in black water areas, were used to find fish.

For stationary recordings, wireless systems were used to transmit amplified signals from recording sites to a computer for signal capture. Four recording sites were monitored simultaneously. The amplifier system was identical to the portable system: a 15 cm probe was placed in the water and the signals amplified using the RadioShack device. The probes were either hung from overhanging trees, placed along the shore, or on small floats. The amplified signals were transmitted and received using Nady systems WGT-15 or WLT-15 wireless transmitter packages. Data were captured using custom software via an Iotech DaqBoard 2000 (16 bit resolution, 20 K sample rate per channel). The computer was powered via a 12 V car battery and power inverter. Twenty-second duration samples were taken every 10 min for periods up to 10 h.

No recordings were used from sites that did not contain EOD signals in the range of frequencies produced by *Eigenmannia*. We made hundreds of additional recordings of pulse-type species in locations that did not include *Eigenmannia*—these samples were not included here.

2.3. Aggregates of *Eigenmannia* in the laboratory

Adult *Eigenmannia virescens* were purchased from various commercial vendors and maintained in tanks at 20–23 °C in groups of 2–30 fish. The experimental arena was a large round plastic tub (diameter = 1.5 m, depth = 0.5 m) filled halfway with water at about 250 μS/cm conductivity (Fig. 1). This conductivity is within the normal range, 50–600 μS/cm, that is used in the laboratory, and reduces the effective size of each fish's electric field thereby increasing the electrical isolation between refuges. Four refuges were placed on the perimeter of the tub. The refuges were 20 cm square plastic sheets that rested on the bottom of the tub with an array of 2 cm spaced, 10 cm tall black plastic rods (∼3 mm diameter). These refuges were a form of artificial reed grass habitat. Each refuge was also equipped with a bubbler and two pairs of electrodes. Signals were amplified using custom-built amplifiers and recorded using a custom computer system identical to that used in the stationary recording system in Ecuador. The quadrant in which each fish was located during a sample was, in the vast majority of cases, easily determined by relative amplitudes of EODs recorded using this system. The combination of conductivity and the calibration of the recording system allowed us to minimize any overlap of signals of fish in each of the four quadrants of the tub (Fig. 1).

Individual fish were taken from various tanks in the laboratory. The selection of fish was random. Two densities of fish were used: one fish per refuge (four fish total) and two fish per refuge (eight fish total). Each experiment was conducted for a minimum of 2 weeks, 12L:12D light cycle. Fish were allowed to acclimate to the new environment for the first 3 days of each experiment. Five-second duration recordings of EOD activity at each refuge were taken every 30 min throughout each experiment after the initial acclimation period. Visual observations were routinely made and compared with the automatically collected behavioral data. Once observations were made for the 2-week period, the fish were removed and returned to holding tanks in the facility.

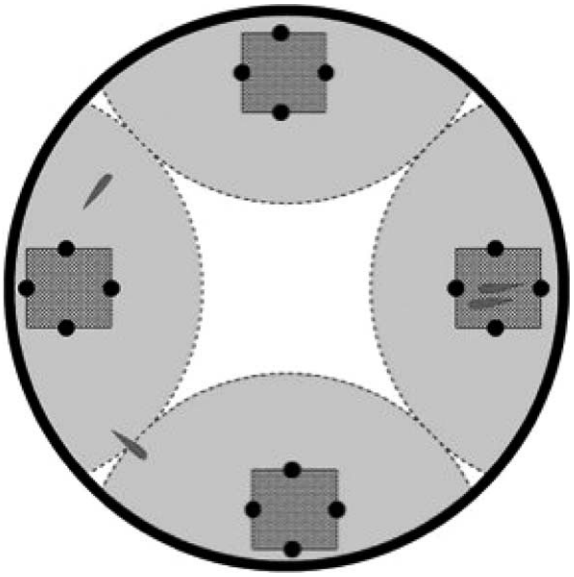

Fig. 1. Schematic of the experimental arena. A large plastic tank was equipped with four refuges (dark squares): figure is not drawn to scale. Recordings were obtained from one pair of electrodes (black dots) at each refuge. The shaded region surrounding each refuge represents the region in which the EODs of *Eigenmannia* could be recorded by the electrodes at that refuge. During the day fish were observed within the refuges (right). During the night the fish were active. The two fish at left illustrate the limitations of the system. These two fish would have been scored as both in the left quadrant. The EOD amplitude of the fish at the border of two quadrants would have been greatest in the left quadrant because the tail, where the electric organ resides, was closest to that refuge. Nevertheless it is likely that the fish could indeed perceive each other's EODs from these positions. Fish were rarely observed in the center region.

2.4. Data analysis

The EODs of *Eigenmannia* are nearly sinusoidal, and fish maintain regular EOD frequencies. It is therefore possible to identify individuals based on the frequencies of each fish's EOD alone. The JAR behavior ensures that each fish in a location will have a distinct EOD frequency. Each sample was plotted as a sonogram using a custom software package that allows very long sample windows (16384+) and window overlap (95%) (Fig. 2). The minimum difference between two EOD frequencies that could be unambiguously resolved was 3 Hz. EOD frequencies differences of less than 3 Hz could be detected by amplitude modulations of the individual EODs but not quantified: this situation was rarely observed. Care was taken to avoid accidental counting of harmonic frequencies as additional fish: power at harmonic frequencies was sufficiently low that setting the dB range of sonograms easily excluded harmonics.

EOD frequencies were determined using the peak power at each EOD frequency. The number of EOD frequencies was counted in each sample. The numbers of samples of each group size were summed. This value was then multiplied by the number of EODs: for example the number of samples with two fish was multiplied by 2 and the number of samples with eight fish was multiplied by 8. In short, the number of samples (unscaled) indicates the distribution of group sizes, whereas the scaled value (group size multiplied by

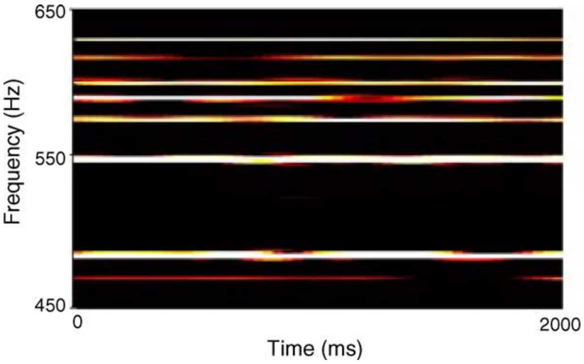

Fig. 2. Sonogram of a sample taken in black water during the day. Each horizontal line represents the EOD of a single *Eigenmannia*. The EOD frequencies were readily distinguished and counted. There are eight fish in this sonogram. Bands that appear then disappear represent fish that swam in and out of the recording area.

number of samples) indicates the number of fish within each group size. To present the scaled and unscaled data on the same axes, we normalized each. These values are the 'percent' data shown in the figures. For example, imagine 10 fish distributed in a group of seven fish, two fish, and a solitary fish. The unscaled value for the group of seven fish would be 33.3% (one out of three groups) and the scaled value would be 70% (seven out of 10 fish observed).

Differences in EOD frequencies were always measured in relation to the nearest signals. For example, for three EOD frequencies we counted two differences in frequencies: the difference between the middle and highest and the middle and lowest EODs. However, the difference in frequency between the highest and lowest EODs was not counted. This method counts the frequency differences for each fish that result in the lowest frequency beat rates, which are most likely to elicit the strongest JARs and electrophysiological responses at the level of the electroreceptors and higher stations in the brain.

Statistical analyses were performed using Statistica 5.5 (StatSoft, Tulsa).

3. Results

3.1. Black water habitats at Sacha Lodge

Recordings were made at 146 sites during daylight hours using the portable recording system (Fig. 3). Sites included the edges of lakes and streams with a wide variety of vegetation including reed grasses, leaf litter, fallen trees, and other floating plants. A total of 827 individual EODs were identified in the daytime data. The median number of EODs detected per sample was 5 and the mean was 5.7 (S.D. = 3.5). A total of 338 recordings were made at six sites using the stationary recording system at night. A total of 1034 EODs were identified among the night samples. There was evidence that fish were far more active at night, as EOD bands could be seen entering and leaving the recordings during individual samples. It is likely that many fish were recorded in more than one sample during the night. Nevertheless, during any single sample the number of EOD frequencies is accurate, and therefore describes the electrosensory conditions of those fish. The

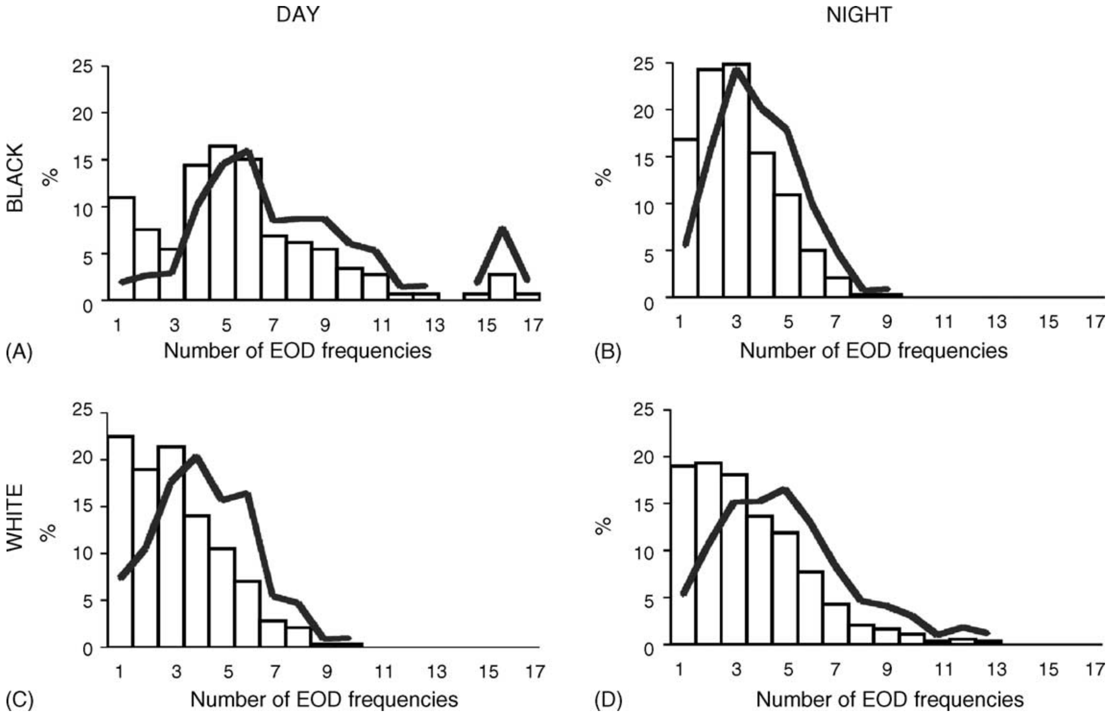

Fig. 3. Distributions of numbers of *Eigenmannia* in samples taken in Ecuador. Bars indicate the percent of samples that contained each number of EOD frequencies ('unscaled', see Section 2). The line indicates the percent of fish for each group size ('scaled', see Section 2). (A) Black water habitat, day; (B) black water habitat, night; (C) white water habitat day and (D) white water habitat, night.

median and mean numbers of EODs observed at night were 3 and 3.1 (S.D. = 1.6), respectively, per sample.

These data were corroborated by visual observations of the fish at night. *Eigenmannia* and other species were commonly observed using flashlights in some of the smaller streams and especially in Orchidea creek. Pulse-type gymnotiforms, including *Brachyhypopomus* sp. and *Gymnotus* sp., were commonly observed—these animals were rarely seen in pairs or larger groups. In contrast, *Eigenmannia* were routinely seen in pairs or groups of three. Although the visual observations support our conclusions, these are weak evidence as debris and leaf litter obscured the view of fish.

The greatest number of EODs observed in a single sample in black water was 29. The largest group sizes, between 24 and 29 EODs, were observed in only 10 samples collected during the day. The sites where larger groups were encountered were different from the more common groups of 17 or fewer fish (Fig. 3). These large groups were located around root systems of trees in igapó habitats where *Eigenmannia* were rarely observed. The differences in EOD frequencies in largest groups were often less than 15 Hz, and would likely result in significant impairment of electrosensory function in many of the fish. Unfortunately it was not possible to capture or otherwise visualize these very large groups.

The frequency of each EOD was measured in each sample. There were no significant differences in the distributions of EOD frequencies (linear regression, $R^2 = 0.006$) in relation to the numbers of EOD frequencies detected in single samples (Fig. 4). Subsequently the differences in EOD frequencies of fish in aggregations were calculated for each sample. The mean difference in closest EOD frequencies (see Section 2) during the day was 35.3 Hz (S.D. = 17.3) and 54.6 Hz (S.D. = 29.6) at night (Fig. 5). These differences are smaller than the maximum frequency differences that the fish could presumably produce (Fig. 5), and are well within the range of detection and encoding by tuberous electroreceptors.

3.2. White water habitats in the Tiputini

Eigenmannia were more difficult to locate along the Tiputini than in black water habitats. Whether this reflects differences in fish densities or differences in the distributions of fish in these habitats remains unclear. Recordings in white water were all achieved using the stationary recording system. During the daylight hours, 285 samples from 12 different locations were recorded: 1022 individual EODs were identified in these recordings. The numbers of EODs observed in samples during the day were smaller than those observed in black water habitats. The largest number of EODs in a sample taken in white water habitats was 10 (Fig. 3). The median number of EODs was 3 and the mean was 3.2 (S.D. = 1.9).

Night samples were collected at the same locations as the daytime samples. A total of 3289 EODs were recorded in 916 samples. The maximum number of EODs observed in a single sample was 13 (Fig. 3). The numbers of EODs observed at night were generally greater than those found in the day. The median number of EODs was 3 and the mean was 3.6 (S.D. = 2.3).

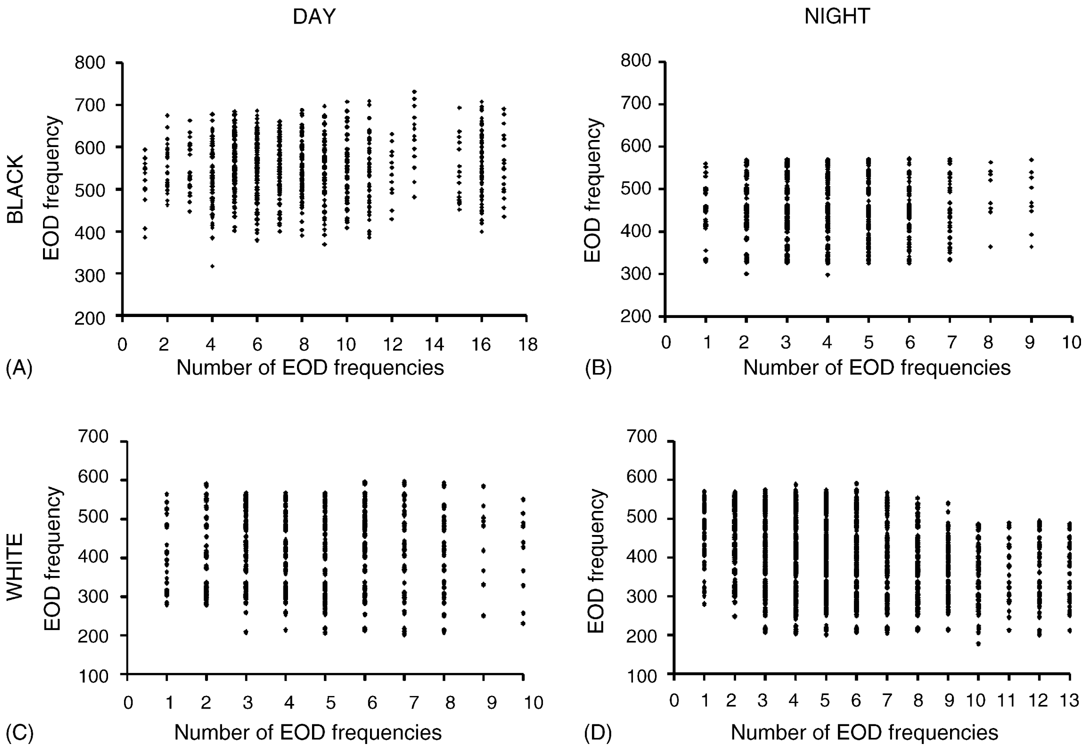

Fig. 4. Distributions of EOD frequencies. Points on the graphs indicate each individual EOD observed in each group size. (A) Black water habitat, day; (B) black water habitat, night; (C) white water habitat day and (D) white water habitat, night.

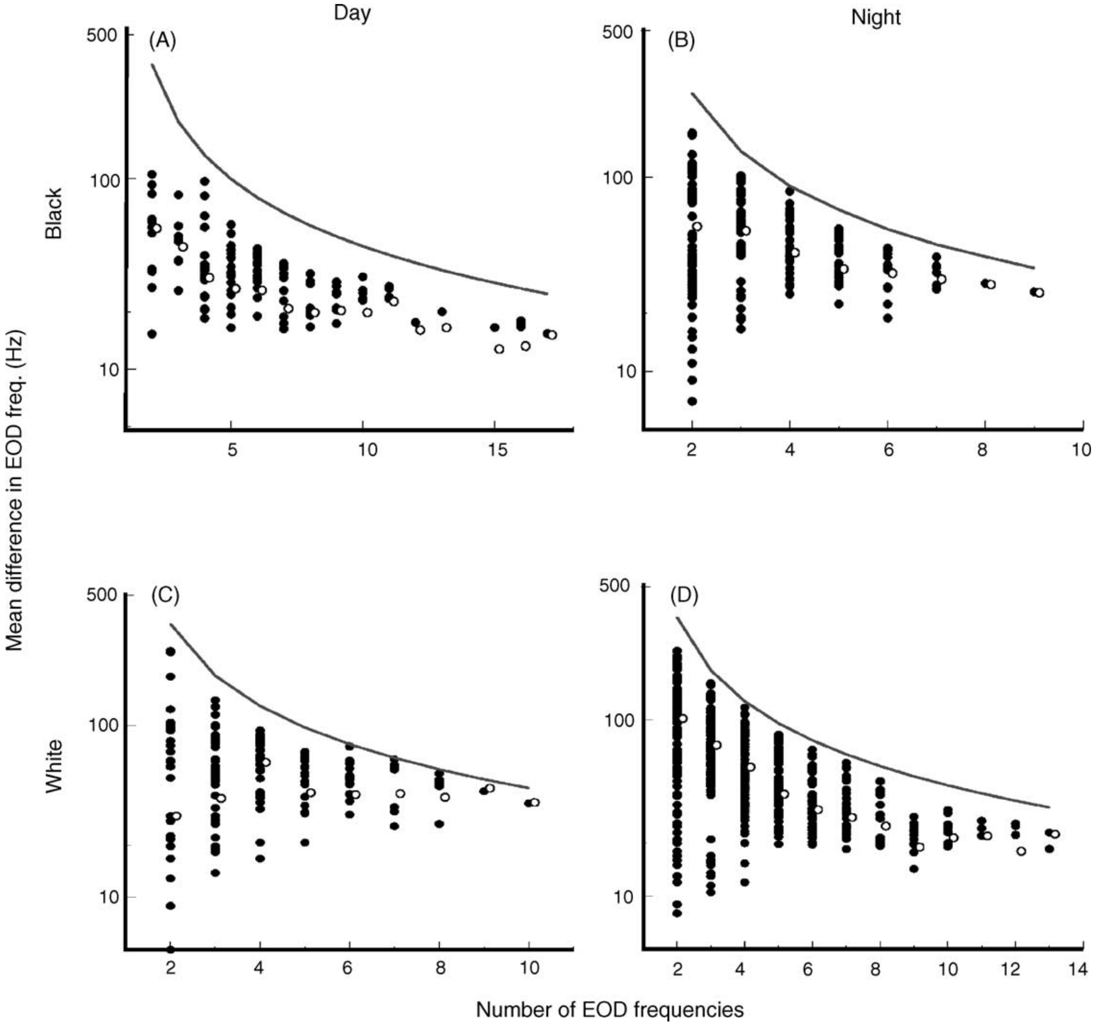

Fig. 5. Distributions of differences between EOD frequencies. Points on the graphs indicate the mean difference in frequencies between all fish in each sample taken. The line represents the calculated maximum average frequency difference possible for each group size: the difference between minimum and maximum frequency observed from the entire data set divided by the one less than the group size. Open circles indicate median frequency differences. Differences in EOD frequencies (*Y*-axis) have been plotted on a log scale. (A) Black water habitat, day; (B) black water habitat, night; (C) white water habitat day and (D) white water habitat, night.

The distribution of EOD frequencies was similar to those observed in black water habitats. There was no significant relation between group size and EOD frequencies (linear regression, $R^2 = 0.06$; Fig. 4). The mean difference in EOD frequencies during the day was 54.1 Hz (S.D. = 35.5) and 65.8 Hz (S.D. = 42.2) at night. The median differences between EOD frequencies were relatively constant across group sizes (Fig. 5). Two trends appear in these data: the median difference in EOD frequencies drops slightly as group size increases in three of the four conditions, and larger group sizes have fewer low EOD-frequency differences.

3.3. Aggregates of *Eigenmannia* in the laboratory

Two different densities of fish were used in this experiment. Seven different groups of fish were tested at a density of one fish per refuge (i.e. four fish in the arena with four refuges). More than 300 samples were analyzed for each group in both the day and the night. The distributions of observed numbers of fish in each quadrant were statistically different from random in each of the seven experiments (Chi Squared test, observed versus random, each $p < 0.001$) (see Fig. 6): fish were observed more often in groups than expected in the random distribution. Even though there were sufficient refuges so that each fish could occupy its own refuge all of the time, fish were nonetheless found in groups of two fish or more in 65% of samples during the day and 69% at night. Fish did not exhibit preferences for particular refuges within and between experiments. There were no discernable trends in the frequency relations of fish over the course of each experiment.

Six experiments were conducted at a density of two fish per refuge (thus there were eight fish in the arena in each group). This higher density of fish allowed for greater degrees of free-

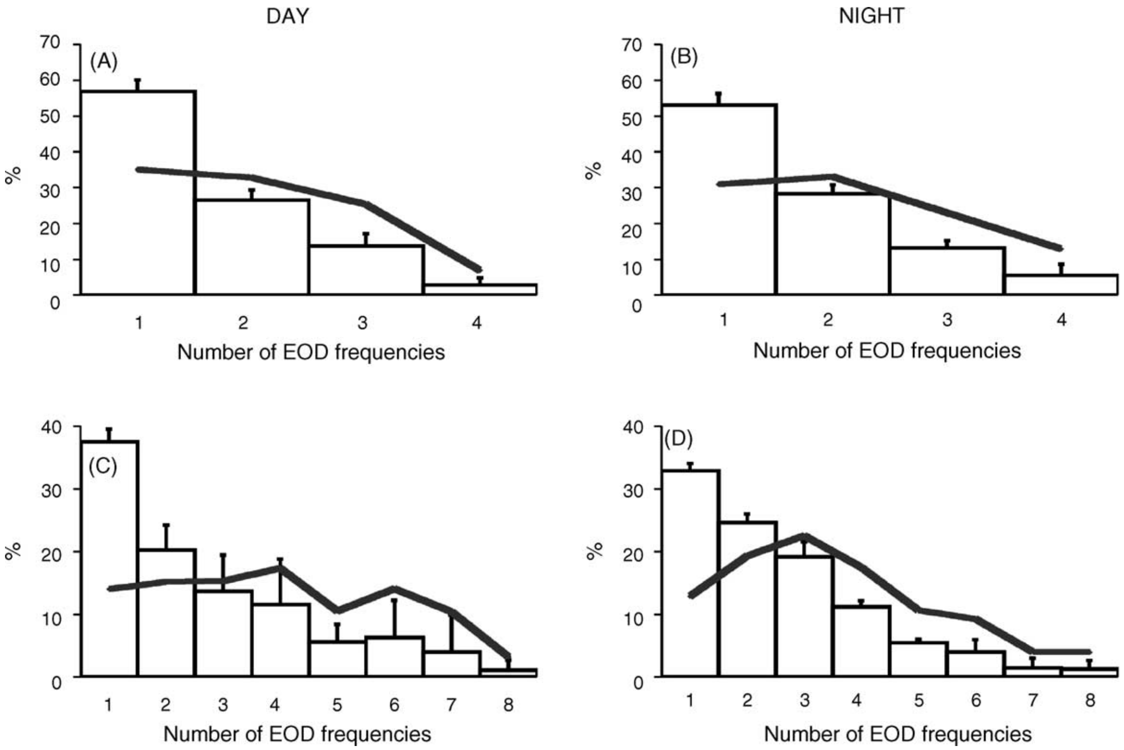

Fig. 6. Distribution of group sizes in refuges in the laboratory. As in Fig. 3, bars indicate the percent of samples that contained each number of EOD frequencies. The line indicates the percent of fish for each group size. Error bars indicate the standard deviation. (A and B) one fish per refuge, day and night, respectively. (C and D) two fish per refuge, day and night, respectively.

dom and larger group sizes than the lower density conditions. More than 300 samples were analyzed for each group in both the day and the night (Fig. 6). The median group size during the day was 4, and night was 3. These group sizes are within the range observed at our field sites. Again, there were no discernable trends in the frequency relations of fish over the course of each experiment.

4. Discussion

This study described the electrosensory interference produced by nearby conspecifics that is routinely experienced by individual *Eigenmannia* both in the wild and in the laboratory. In both black water and white water habitats near the Napo river in the upper Amazon, *Eigenmannia* were most commonly found in electrosensory groups of three to five fish throughout the day and night. The differences in EOD frequencies between fish, the key measure relative to the activation of electroreceptors and electrosensory perception, were most commonly between 23 and 41 Hz. This range of differences in EOD frequencies are detected and encoded by tuberous electroreceptors. In the laboratory, *Eigenmannia* were preferentially found in groups, even when there were sufficient refuges so that individual fish could each be isolated. Group sizes were similar in the naturalistic laboratory setting to those observed in Ecuador.

The studies conducted in Ecuador have three important limitations. First, it was not possible to observe the fish during most recording samples. As a result, it is impossible to unambiguously determine what species of fish produced the signals at each recording site or assess the actual spatial distribution of fish. However, when we traveled in the waterways during the night, fish could be observed by flashlight, and the EODs recorded. These observations strongly suggest that most if not all of the recordings identified as *Eigenmannia* were from that species. Second, it was not possible to assess the effects of population density on aspects of social behavior. One possibility is that the fish are abundant and the numbers of refuges are limited. If so, fish would be found at high densities at all refuges. Although it was not possible to directly assess the quality of refuges available to the fish, it appeared that there were many potential refuges that were not occupied, and in those that were occupied the fish were in close proximity to each other, perhaps under 20 cm. Alternatively, it is possible that the years when these studies were conducted experienced a far lower density of fish than normal, in which case the numbers of nearby conspecifics would have been underestimated relative to normal years. Finally, all of the recordings were made at depths less than 2 m. We did not encounter weakly electric fish except near the edges of streams and lakes. It is possible that fish are abundant in areas that were not examined in this study.

In contrast to the studies in Ecuador, in the laboratory the numbers/densities of fish relative to the availability of refuges could be controlled. We tested two densities of fish. At the lowest densities tested, each fish could remain electrically isolated from each other. This is important because there is a categorical difference between fish that are alone versus fish that are in any size group. Fish in groups experience emergent ongoing 'global' oscillations that are not experienced by fish that are alone. At these low densities, fish could potentially remain alone but most commonly did not. The limitation of the experiments that used the low density of fish is that the maximum group size could only be four fish. In the wild, group sizes were typically between three and five fish, with a maximum of 29 fish. At the higher density of fish, two fish per refuge, a maximum of eight fish in a group was possible. Under these conditions, median group sizes observed in the laboratory were similar to those observed in Ecuador. Visual observations confirmed that fish were most often found within centimeters of each other. Fish appeared to prefer being near other conspecifics rather than maximizing use of the available hiding areas.

Higher densities of *Eigenmannia* are commonly maintained in laboratory tanks. For example, we routinely keep 25 *Eigenmannia* in 50 gal tanks. In these conditions, it is unlikely that individuals can escape the EODs of other fish throughout the tank. At these densities, *Eigenmannia* are observed in shoals [29]. The relations of these dense laboratory conditions to those that occur in the wild are unclear. We did observe a few unusual sites with very high densities of fish, up to 29 fish, within the range of detection of our probe. Previous reports describe larger shoals of *Eigenmannia* in the wild [24].

These data are consistent with previous reports using *Eigenmannia* and other species. Early reports by Lissmann (1961) noted that the different species of gymnotiform fishes exhibited different social interactions: *Eigenmannia* were always found in groups whereas other species were not [19,24]. Indeed, most other genera of gymnotiform fishes appear to be less social. *Apteronotus*, which exhibits a "weaker" JAR than *Eigenmannia*, and *Sternopygus*, which does not have a JAR behavior, are found to be more territorial in laboratory settings (unpublished observations). *Apteronotus leptorhynchus* behave differently at high densities in laboratory tanks than *Eigenmannia*. *Apteronotus* preferentially hide in refuges rather than shoal [19]. Finally, the independently evolved wave-type mormyrid species *Gymnarchus niloticus* have been observed in natural settings in of groups of two or more fish with differences in EOD frequencies of at least 4 Hz [25].

4.1. What is the meaning of the EOD frequency?

Field studies of the gymnotiform species *Sternopygus macrurus* have demonstrated that EOD frequency is sexually dimorphic [41]. In this wave-type species, the males have EODs frequencies between 50 and 90 Hz while the female EODs occur at rates of 110–200 Hz. Sexual dimorphism in EOD frequency has also been reported in *Eigenmannia virescens* [16,19]. Further, Hagedorn and Heiligenberg (1985) observed that dominant male *Eigenmannia* typically possess the lowest EOD frequencies in groups while dominant females had the highest EOD frequencies [16]. In contrast, Dunlap et al. (2002) observed that there might not be a correlation between dominance and EOD frequency in *Apteronotus leptorhynchus* [8]. Pairs of *Apteronotus* produced more chirps and aggressive behaviors than when alone, but no difference in EOD frequencies of paired versus solitary fish were observed [8].

The data presented here do not support the hypothesis that there is a relationship between EOD frequency and dominance in *Eigenmannia*. Dominant individuals are most likely to secure and protect the most desirable refuges. Thus, dominant fish might be expected to be observed in smaller groups. If EOD frequency was correlated with dominance, the smaller group sizes should be characterized by having either higher or lower EOD frequencies. No such relationship was observed. These data are not, however, a direct test of this hypothesis, as dominance relations were not studied and fish could not be seen by eye in most cases. How groups are generated and maintained also remains unclear.

An important question remains—how do fish achieve the wide range of EOD frequencies observed in the wild? In *Apteronotus*, sustained jamming for minutes to hours can lead to a long-term frequency elevation [28]. This elevation is mediated by NMDA receptors at the pacemaker nucleus. A similar mechanism has recently been demonstrated in *Eigenmannia* for both EOD frequency increases and decreases [29]. Preliminary data from our laboratory suggest that sustained jamming can lead to changes in EOD frequencies of 100 Hz or more, and can change resting EOD frequency of *Eigenmannia* [12].

4.2. What is the function of aggregation in *Eigenmannia*?

There are potentially many reasons why *Eigenmannia* aggregate or shoal. Aggregation can reduce the risk of predation [2,15,35,37,38]. Fish in groups decrease the per capita mortality rate because a predator is less likely to be able to capture an entire group than to capture a solitary fish. The larger group may also serve to confuse predators, as they are unable to isolate specific targets [22].

Using modeling techniques, Grunbaum (1998) suggested that schooling was a strategy for improving behavioral performance [14]. Isolated individuals were less able to accurately perform gradient-climbing and alignment behaviors in noisy environments than individuals within groups [14]. Grouping may also enhance sensory perception [13,27]. Aggregations can allow for the interaction of sensory systems, which may result in an increase in the ability to obtain and perceive information about the environment.

It is possible that the emergent electrosensory signals in aggregates or shoals of *Eigenmannia* enhance electrosensory perception of salient stimuli. *Eigenmannia* in aggregates most commonly experience ongoing 20–40 Hz global interference patterns. These signals have been shown to elicit short-term synaptic depression in midbrain neurons [11,36]. The level of depression in synapses, therefore, differs when the fish are solitary versus when the fish are in close proximity. Short-term synaptic depression may be a mechanism for the processing of moving electrosensory images [6]. Recent evidence suggests that motion processing, in particular the discrimination of direction of movement, is improved in midbrain neurons with the addition of global interference patterns like those found in aggregates of *Eigenmannia* [34].

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References

[1] Atema J. Chemical signals in the marine environment: dispersal, detection, and temporal signal analysis. Proc Natl Acad Sci 1995;92(1):62–6.

[2] Bertram BCR. Behavioral Ecology: An Evolutionary Approach. Sunderland, Massachusetts: Sinauer; 1978.

[3] Bregman AS. Auditory Scene Analysis. Cambridge, MA: MIT Press; 1990.

[4] Bullock TH, Hamstra RH, Scheich H. The jamming avoidance response of high-frequency electric fish, I and II. J Comp Physiol 1972;77:1–48.

[5] Chacron MJ, Doiron B, Maler L, Longtin A, Bastian J. Non-classical receptive field mediates switch in sensory neuron's frequency tuning. Nature 2003;425:77–81.

[6] Chance FS, Nelson SB, Abbott LF. Synaptic Depression and the temporal response characteristics of V1 cells. J Neurosci 1998;18:4785–99.

[7] Cherry EC. Some experiments on the recognition of speech, with one and two ears. J Acoust Soc Am 1953;25:975–9.

[8] Dunlap KD, Oliveri LM. Retreat site selection and social organization in captive electric fish, *Apteronotus leptorhynchus*. J Comp A Neurothol Sens Neural Behav Physiol 2002;188:469–77.

[9] Feng AS, Ratnam R. Neural basis of hearing in real-world situations. Annu Rev Psychol 2000;51:699–725.

[10] Fortune ES, Rose GJ. Short-term synaptic plasticity as a temporal filter. TINS 2001;24:381–5.

[11] Fortune ES, Rose GJ. Short-term synaptic plasticity contributes to the temporal filtering of electrosensory information. J Neurosci 2000;20:7122–30.

[12] Fortune ES, Tan EW, Nizar J. Relation of group size to the control of electric organ discharges in gymnotiform fish. Soc Neurosci Abstr 2003:627.9.

[13] Grunbaum D. Animal Groups in Three Dimensions. London: Cambridge University Press; 1997. pp. 257–300.

[14] Grunbaum D. Schooling as a strategy for taxis in a noisy environment. Evol Ecol 1998;12:503–22.

[15] Hagedorn M. Chapter seventeen: the ecology, courtship, and mating of gymnotiform electric fish. In: Bullock TH, Heiligenberg WF, editors. Electroreception. New York, NY: John Wiley and Sons, Inc.; 1986. p. 613–49.

[16] Hagedorn M, Heiligenberg WF. Court and spark: electric signals in the courtship and mating of gymnotoid electric fish. Anim Behav 1985;33:254–65.

[17] Heiligenberg WF, Baker C, Matsubara J. The jamming avoidance response in *Eigenmannia* revisited: the structure of a neural democracy. J Comp Physiol 1978;127:267–86.

[18] Heiligenberg WF. Neural Nets in Electric Fish. The MIT Press: Cambridge, Massachusetts; 1991.

[19] Hopkins CD. Electric communication: functions in the social behavior of *Eigenmannia virescens*. Behavior 1974;50:270–305.

[20] Julesz B, Hirsh IJ. Visual and auditory perception: an essay in comparison. In: David EE, Denes P, editors. Human Communication: A Unified View. New York: McGraw-Hill; 1972. p. 283–335.

[21] Kramer B, Kirschbaum F, Markl H. Species specificity of electric organ discharges in a sympatric group of gymnotid fishes from Manaus (Amazonas). In: Sensory Physiology of Aquatic Lower Vertebrates. Budapest: Akademiai Kiado, Pergamon; 1981. pp. 195–219.

[22] Landeau L, Terborgh J. Oddity and the 'confusion' effect in predation. Anim Behav 1986;34:1372–80.

[23] Lardner B, bin Lakim M. Animal communication: tree hole frogs exploit resonance effects. Nature 2002;420(6915):475.

[24] Lissmann HW. Ecological studies on gymnotids. In: Bioelectrogenesis. Amsterdam: Elsevier; 1961. pp. 215–226.

[25] Moller P, Serrier J, Belbenoit P. Electric organ discharges of the weakly electric fish *Gymnarchus niloticus* (Mormyriformes) in its natural habitat. Experientia 1976;32:1007–8.

[26] Nelson BS. Reliability of sound attenuation in Florida scrub habitat and behavioral implications. J Acoust Soc Am 2003;113(5):2901–11.

[27] Norris KS, Schilt C. Cooperative societies in three-dimensional space: on the origins of aggregations, flocks, and schools, with special reference to dolphins and fish. Ethol Sociobiol 1988;9:149–79.

[28] Oestreich J, Zakon HH. The long-term resetting of a brainstem pacemaker nucleus by synaptic input: a model for sensorimotor adaptation. J Neurosci 2002;22:8287–96.

[29] Oestreich J, Zakon HH. Species-specific differences in sensorimotor adaptation are correlated with differences in social structure. J Comp Physiol A, 2005(July 9):1–12.

[30] Partridge BL, Heiligenberg W, Matsubara J. The neural basis for a sensory filter in the jamming avoidance response: no grandmother cells in sight. J Comp Physiol 1981;145:153–68.

[31] Perla BS, Slobodchikoff CN. Habitat structure and alarm call dialects in Gunnison's prairie dog (*Cynomys gunnisoni*). Behav Ecol 2002;13(6):844–50.

[32] Pytte CL, Rusch KM, Ficken MS. Regulation of vocal amplitude by the blue-throated hummingbird, *Lampornis clemenciae*. Anim Behav 2003;66:703–10.

[33] Ramcharitar JU, Tan EW, Fortune ES. Effects of broad-field electrosensory signals on motion processing in the torus semicircularis of *Eigenmannia*. J Comp Physiol., 2005(July 6):1–8.

[34] Ramcharitar JU, Tan EW, Fortune ES. Role of short-term synaptic plasticity in the generation of direction selectivity for moving electrosensory images. Soc Neurosci Abstr 2004:58.7.

[35] Riipi M, Alatalo RV, Lindstrom L, Mappes J. Multiple benefits of gregariousness cover detectability costs in aposematic aggregations. Nature 2001;413:512–4.

[36] Rose GJ, Fortune ES. Frequency-dependent PSP depression contributes to low-pass temporal filtering in *Eigenmannia*. J Neurosci 1999;19:7629–39.

[37] Treisman M. Predation and the evolution of gregariousness. I. Models for concealment and evasion. Anim Behav 1975;23:779–800.

[38] Turner GF, Pitcher TJ. Attack abatement: a model for group protection by combined avoidance and dilution. Am Nat 1986;132:228–40.

[39] Watanabe A, Takeda K. The change of discharge frequency by A.C. stimulus in a weakly electric fish. J Exp Biol 1963;40:57–66.

[40] Westby GWM. The ecology discharge diversity and predatory behavior of gymnotiform electric fish in the costal streams of French Guiana. Behav Ecol Sociobiol 1988;22:341–54.

[41] Zakon HH, Thomas P, Yan HY. Electric organ discharge frequency and plasma sex steroid levels during gonadal recrudescence in a natural population of the weakly electric fish *Sternopygus macrurus*. J Comp Physiol A 1991;169:493–9.