ICN Satellite meeting

Electric fishes: Neural systems, behavior and evolution

FU Berlin, 27 - 28 July 2024

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Abstract booklet

at at a a a a a

Electric fishes: Neural systems, behavior and evolution

FU Berlin, 27 - 28 July 2024



Organizing committee:

- Laura Quintana (Montevideo, Uruguay)
- Rüdiger Krahe (HU Berlin, Germany)
- Jan Benda (Tübingen, Germany)

Abstract booklet

Talks	1
Poster	35
Author index	55

Document navigation

To aid navigation, links marked in **red** have been inserted:

- You can click on the contribution categories listed above to get there.
- In the program you can click on the author, \ldots
- and in the talk and poster lists at the beginning of each category, you can click on the abstract titles to get to the corresponding abstracts.
- In the abstract's references you can click on the DOIs, to get to the publications.
- In the author index, you can click on the page numbers to get to the corresponding abstracts.

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Production

Abstract submission was handled by the conference abstracts and submission web-interface provided by the German Neuroinformatics node (https://abstracts.g-node.org/). Abstracts were downloaded, filtered and sorted using the open-source GCA-Python tools. The document was then typeset with LATEX.

Cover

The Brandeburg Gate at the heart of Berlin (photo by Thomas Wolf, www.foto-tw.de / Wikimedia Commons / CC BY-SA 3.0) and two of the *Electrophorus voltai* living in the Natural History Museum (Museum für Naturkunde), Berlin (photos by Peter Bartsch).

Venue



EFish Satellite Arnimallee 22, 14195 Berlin-Dahlem

Alter Krug Königin-Luise-Straße 52, 14195 Berlin-Dahlem

ICN main meeting Henry Ford Building, Garystraße 35, 14195 Berlin-Dahlem

Public transport fare zones



for rides from the conference venues to the city center for rides from the conference venues to Potsdam A B C city center, conference venues, Potsdam, and airport (BER)

Travel awards

We were able to financially support seven participants with a travel award. This was made possible by the generous support of the priority program SPP 2205 of the DFG: "Evolutionary optimization of neuronal processing". We are very grateful to this.



SPP 2205 Evolutionary optimization of neuronal processing

Saturday, July 27th, 2024: morning

8:15	Registration & Badges	
8:50	Welcome	
9:00 - 10:30	Session: Behavior in the wild: ecological and physiological aspects	
Ana Silva (Mo	ntevideo, Uruguay): Spatial patterns and electric behavior of an undisturbed population of the pulse-type weakly electric fish, <i>Gymnotus omarorum</i> , in the wild.	
Jorge Molina	Bogota, Colombia): Spatial aggregation and resting places of <i>Apteronotus galvisi</i> (Apteronotidae: Gymnotiformes) individuals in a stream in the Orinoquia region (Meta: Colombia).	
Cecilia Jalabe	rt (Montevideo, Uruguay): Seasonality of neuroestrogens and their relationship with aggression: insights from the electric fish model <i>Gymnotus omarorum</i> .	
Lok Poon (Orla	ando, Florida): Eye size predicts Neotropical electric fish response to moonlight: the role of visual acuity and light intensity in determining foraging trade-offs.	
Peter Bartsch	(Berlin, Germany): From the Amazon to the Museum für Naturkunde: electric eels in captivity.	
10:30 - 11:00	Coffee	
11:00 – 11:55	Session: Physiology of the electric sense	
Jan Grewe (Tübingen, Germany): Bursts boost nonlinear encoding in electroreceptor afferents.		
Amin Akhshi (Montreal, Canada):	
	Burst firing generates invariant coding of natural electrocommunication stimuli.	
Liz Weerdmee	Burst firing generates invariant coding of natural electrocommunication stimuli. ster (Berlin, Germany): Electrogenic Na ⁺ /K ⁺ -ATPases constrain electrocyte activity and pose additional evolutionary pressure.	
Liz Weerdmee 11:55 – 12:30	Burst firing generates invariant coding of natural electrocommunication stimuli. ster (Berlin, Germany): Electrogenic Na ⁺ /K ⁺ -ATPases constrain electrocyte activity and pose additional evolutionary pressure. Session: Electric signaling	
Liz Weerdmee 11:55 – 12:30 Günther Zupa	Burst firing generates invariant coding of natural electrocommunication stimuli. ster (Berlin, Germany): Electrogenic Na ⁺ /K ⁺ -ATPases constrain electrocyte activity and pose additional evolutionary pressure. Session: Electric signaling nc (Boston, Massachusetts): The effects of urethane, MS-222, and eugenol anesthesiaon the electric organ discharge of the weakly electric fish <i>Apteronotus leptorhynchus</i> .	
Liz Weerdmee 11:55 – 12:30 Günther Zupa Vielka Salazar	Burst firing generates invariant coding of natural electrocommunication stimuli. ster (Berlin, Germany): Electrogenic Na ⁺ /K ⁺ -ATPases constrain electrocyte activity and pose additional evolutionary pressure. Session: Electric signaling nc (Boston, Massachusetts): The effects of urethane, MS-222, and eugenol anesthesiaon the electric organ discharge of the weakly electric fish <i>Apteronotus leptorhynchus</i> . (Sydney, Canada): Circadian regulation of gene expression in the electric organ of the gymnotiform fish <i>Brachyhypopomus gauderio</i> .	

Saturday, July 27th, 2024: afternoon

14:00 – 16:30	Joint session with satellite meeting: From Archerfish to Zebrafish: How teleost brains interact with the world
Clare Baker (C	ambridge, UK): Electroreceptor development and evolution: insights from comparative molecular approaches.
Adelaide Sibea	aux (Oxford, UK): Can fish take shortcuts? A study of vector-based navigation in fish.
Eric Fortune (N	Newark, New Jersey): Neurophysiological control of refuge tracking.
Avner Wallach	(Haifa, Israel): (Title to be confirmed).
16:00 – 17:30	Coffee & posters
17:30 – 18:00	Pioneer talk
Angel Caputi (Montevideo, Uruguay): A synthetized view of the electrosensory lobe of pulse Gymnotiformes.
18:00 – 19:00	Session: Electric signaling
William Cramp	ton (Orlando, Florida): Reproductive signals and signal evolution in wild electric fishes: perspectives from <i>Brachyhypopomus</i> .
Ralph Tiedema	ann (Potsdam, Germany): Evolution of Electric Organ Discharge (EOD) in African weakly electric fishes: genomics and behavioral ecology of a magic trait.
Francesco Jan	Izen (Toronto, Canada): A molecular phylogenetic tree and DNA barcode database for Gymnotiformes.
19:30	Dinner at Alter Krug

Sunday, July 28th, 2024: morning

9:00 - 10:30	Session: Physiology of the electric sense
Harold Zakon	(Austin, Texas): Dual mechanisms contribute to enhanced voltage dependence of an electric fish potassium channel.
Gary Marsat (N	Morgantown, West Virginia): Spatial coding of conspecific signals in the ELL of A. leptorhynchus.
Masashi Kawa	saki (Charlottesville, Virginia): Action potentials in the pacemaker nucleus of <i>Apteronotus leptorhynchus</i> may lack resting potential.
Megan Freiler	(Bloomington, Indiana): Neuromodulator receptor gene expression in electrosensory brain regions varies across species of electric knifefishes.
Martin Jarzyna	a (St. Louis, Missouri): Shared neural substrates for seasonal and evolutionary shifts in sensorimotor integration.
10:30 - 11:00	Coffee & posters
11:00 – 11:55	Session: Behavior in the wild: ecological and physiological aspects
Stefan Mucha	(Berlin, Germany): From hypoxia to hyperoxia: distribution and activity patterns of mormyrids across extreme spatiotemporal environmental gradients.
Nonato Gome	s Mendes-Junior (Macapá, Brasil): Relationship between length and electrical organ discharges (EOD) in the electric eel <i>Electrophorus voltai</i> (Gymnotiformes: Gymnotidae).
Devika Narain	(Leeuwarden, Netherlands): Habitat, natural activity and ethno-cultural use of electric eels in Suriname.
11:55 – 12:30	Session: Electric signaling
Livio Oboti (Be	erlin, Germany): Specialized signals for conspecific electrolocation in weakly electric fish.
Patrick Weygo	Idt (Tübingen, Germany): Detection and assignment of electrocommunication signals with deep learning.

Sunday, July 28th, 2024: afternoon

13:00 – 18:00 Excursions to electric fish in Berlin

18:00 Openening ceremony of the ICN main meeting

Talks

T1	Migliaro : Spatial patterns and electric behavior of an undisturbed population of the pulse-type weakly electric fish, <i>Gymnotus omarorum</i> , in the wild	3
T2	Osorio-Ospina : Spatial aggregation and resting places of <i>Apteronotus galvisi</i> (Apteronotidae: Gymnotiformes) individuals in a stream in the Orinoquia region (Meta: Colombia)	4
Т3	Jalabert : Seasonality of neuroestrogens and their relationship with aggression: insights from the electric fish model <i>Gymnotus omarorum</i>	5
T4	Poon : Eye size predicts Neotropical electric fish response to moonlight: the role of visual acuity and light intensity in determining foraging trade-offs	6
T5	Bartsch : From the Amazon to the Museum für Naturkunde: electric eels in captivity	7
T6	Grewe: Bursts boost nonlinear encoding in electroreceptor afferents	9
Τ7	Akhshi : Burst firing generates invariant coding of natural electrocommunication stimuli	11
Т8	Weerdmeester: Electrogenic Na^+/K^+ -ATPases constrain electrocyte activity and pose additional evolutionary pressure	13
Т9	Zupanc : The effects of urethane, MS-222, and eugenol anesthesia on the electric organ discharge of the weakly electric fish <i>Apteronotus leptorhynchus</i>	15
T10	Cox : Circadian regulation of gene expression in the electric organ of the gymnotiform fish <i>Brachyhypopomus gauderio</i>	17
T11	Baker : Electroreceptor development and evolution: insights from compara- tive molecular approaches	18
T12	Fortune: Neurophysiological control of refuge tracking	19
T13	Caputi: A synthetized view of the electrosensory lobe of pulse Gymnotiformes	20
T14	Crampton : Reproductive signals and signal evolution in wild electric fishes: perspectives from Brachyhypopomus	21
T15	Tiedemann : Evolution of Electric Organ Discharge (EOD) in African weakly electric fishes: genomics and behavioral ecology of a magic trait	22
T16	Janzen : A molecular phylogenetic tree and DNA barcode database for Gymnotiformes	23

T17	Zakon : Dual mechanisms contribute to enhanced voltage dependence of an electric fish potassium channel	24
T18	Marsat: Spatial coding of conspecific signals in the ELL of A. leptorhynchus	25
T19	Kawasaki: Action potentials in the pacemaker nucleus of <i>Apteronotus leptorhynchus</i> may lack resting potential	26
T20	Freiler : Neuromodulator receptor gene expression in electrosensory brain regions varies across species of electric knifefishes	27
T21	Jarzyna : Shared neural substrates for seasonal and evolutionary shifts in sensorimotor integration	28
T22	Mucha : From hypoxia to hyperoxia: distribution and activity patterns of mormyrids across extreme spatiotemporal environmental gradients	29
T23	Mendes-Junior : Relationship between length and electrical organ discharges (EOD) in the electric eel <i>Electrophorus voltai</i> (Gymnotiformes: Gymnotidae)	30
T24	Narain: Habitat, natural activity and ethno-cultural use of electric eels in Suriname	32
T25	Oboti : Specialized signals for conspecific electrolocation in weakly electric fish	33
T26	Weygoldt : Detection and assignment of electrocommunication signals with deep learning	34

Spatial patterns and electric behavior of an undisturbed population **T1** of the pulse-type weakly electric fish, *Gymnotus omarorum*, in the wild

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- 4. Neuroethology, University of Tubingen, Tubingen, Germany

Assessing animals' locomotor and activity-rest patterns in natural populations is challenging. It requires individual identification and tracking of its behavior in sometimes complex and inaccessible environments. Weakly electric fish are advantageous models for remote monitoring due to their continuous emission of electric signals (EODs). *Gymnotus omarorum* is a South American freshwater pulse-type weakly electric fish. Previous manual recordings of restrained individuals in the wild showed a spatial distribution compatible with territoriality and a nocturnal increase in EOD rate interpreted as arousal. This interdisciplinary study presents the development of low-cost amplifiers for remote EOD recordings and the refinement of tracking algorithms that provide individual recognition of *Gymnotus omarorum* in the wild. We describe natural daily spacing patterns of undisturbed individuals that are compatible with territoriality, although heterogeneous across sampling sites, and confirm that all resident fish showed a robust nocturnal increase of EOD rate likely associated with daily variations of water temperature.

Spatial aggregation and resting places of *Apteronotus galvisi* (Apteronotidae: Gymnotiformes) individuals in a stream in the Orinoquia region (Meta: Colombia)

Juan Camilo Osorio-Ospina¹, Jorge Molina¹

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South American weak electric fish (Gymnotiformes) navigate and communicate using their electric organ discharges (EOD). The emission of EODs depends on internal and external factors and is used to study the biology and ecology of these organisms. Recent studies have evaluated the distribution and territoriality of some Apteronotus species under laboratory conditions, but more studies are needed to better understand their ecology and behavior under natural conditions. Taking this into account, we used fish-finder amplifiers to locate individuals of Apteronotus galvisi at rest and a GPS to geoposition their resting places in a 96 m stretch of a stream located in San Martín (Meta, Colombia) from September to November 2022. For some individuals we also conducted 24-hour recordings of their EODs to ensure that this monitoring method accurately identifies fish regardless of water temperature. We found on average 49 individuals resting in the sampled area, with a ratio of 1:6 mature males:females/immatures. Furthermore, we found a clustered distribution of the fish in their resting places, and most males retained their resting places during the months sampled. The effect of water temperature on the EOD frequency of A. galvisi showed an average temperature coefficient (Q10) of 1.67 under natural conditions. We were unable to track females/immature individuals at the study site due to overlap in their EOD frequency ranges in the clustered areas of the stream. In conclusion, through non-continuous studies of A. qalvisi in its natural habitat we found an asymmetric sex ratio, a clustered pattern of spatial distribution in roosting sites, and males with fixed territories surrounded by females/mmatures.

Acknowledgements

We are thankful to Miguel Ángel Suárez Russi and "El Caduceo Natural Reserve" for allowing us to carry out this research in the stream. JM wants to thanks the Faculty of Sciences–Universidad de los Andes for funding through the research Program INV-2023-176-2970.

Seasonality of neuroestrogens and their relationship with aggression: insights from the electric fish model *Gymnotus omarorum*

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Steroids play a crucial role in modulating brain and behavior. While traditionally considered recipients of sex steroids produced in endocrine glands, the brain itself produces steroids. Neurosteroids, synthesized in brain regions regulating social behaviors, play a local regulatory role. While mainly studied in mammals and birds, insights from fish models shed light on neurosteroid synthesis mechanisms and their role on social behavior in vertebrates. Our model species, the weakly electric fish *Gymnotus omarorum*, is an excellent model to study the seasonality and role of neurosteroids as estrogens affect social behavior differently based on season. In this study, we characterized seasonal changes in estrogen (estrone and 17β -estradiol) levels in plasma and forebrain of wild fish using liquid chromatography-tandem mass spectrometry (LC-MS/MS). Examining the relationship between peripheral and central measurements reveals the brain's sexspecific role in local estrogen synthesis. In males, during both seasons estrogens were detectable in the brain but not in the circulation. In contrast, in breeding females, estrogen levels were similar in the brain and circulation, while in non-breeding females estrogens were detectable only in the brain. These data suggest that neuroestrogens are synthesized year-round in males, whereas in females, they are predominantly synthesized in the non-breeding season. These findings align with previous studies of G. omarorum, indicating that non-breeding aggression relies on estrogen signaling, consistent with observations in bird and mammal models. Overall, our results provide a foundation for understanding the role of neurosteroids, the interplay between central and peripheral steroids, and potential sex differences in regulating social behaviors. Lastly, G. *omarorum*, is a teleost fish that offers a unique opportunity to identify common strategies in the role of neurosteroids on social behaviors that may have emerged across vertebrates.

Acknowledgements

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Eye size predicts Neotropical electric fish response to moonlight: **T4** the role of visual acuity and light intensity in determining foraging trade-offs

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Foraging requires a trade-off between acquiring food and the risks posed by predation. At night, the low intensity of natural nighttime light often limits visual acuity. Given the importance of vision in foraging and predator detection, nighttime light mediates the perception of predation risk. Moonlight varies in a complex manner due to the waxing and waning of the moon through the monthly lunar cycle, as well as its nightly rise and set. These variations in light result in substantial variations in predation risk. Along with light availability, the perception of predation risk depends on the animal's visual adaptation. While some nocturnal animals have highly sensitive eyes that aid in foraging and predator detection in dim light, others rely on non-visual sensory modalities. Therefore, within a nocturnal predator-prey dynamic, we expect the prey's response to light to depend on the relative sensory modalities and dim light visual adaptation of the animals. The 'visual acuity hypothesis' predicts that species with low visual acuity should suppress activity during brightly moonlit periods more. This is because while moonlight is associated with predation risk, prey with better acuity could also benefit from moonlight in improving visual detection of the predators. We tested this visual acuity hypothesis in four species of nocturnally active gymnotiform from shallow rainforest streams. The species possess substantial interspecific variation in body mass-corrected eye size, implying variations in their visual acuity. Additionally, the species we focused on forage in the open water sand bottom and are thus exposed to moonlight. Gymnotiform fishes rely on active electrolocation for navigation and foraging in the dark, employing an electric organ discharge (EOD) to detect distortions in the electric field. This allowed us to non-invasively monitor foraging activity by recording EODs at the stream via custom loggers connected to submerged electrodes. By combining the foraging activity over a full lunar cycle with precise measurements of moonlight illuminance, we were able to document the moonlight responses of gymnotiforms with high temporal and activitylevel resolution. Our results were consistent with the visual acuity hypothesis. We found that species with small eyes were highly responsive to moonlight level, showing significant suppression in foraging activity during brightly moonlit periods, whereas a species with larger eyes did not respond to moonlight.

Acknowledgements

We are thankful to Jorge Molina and Universidad de Los Andes for hosting fieldwork in Colombia. Funding to LP was provided by the American Philosophical Society Lewis and Clark Fund and the National Science Foundation Graduate Research Fellowship Program.

From the Amazon to the Museum für Naturkunde: electric eels in **T5** captivity

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A group of *Electrophorus voltai* rescued from a backwater pool at Rio Xingu dam is kept in pondlike facilities at Museum für Naturkunde in Berlin. The purpose is to integrate field and captive observations and develop new technologies to quantify, formulate, and test hypotheses of social behavior in electric eels (e.g., Bastos et al. 2021). In particular, prey capture during collective hunting, and reproduction of these strongly electric pulse-type fish. Several observations and EOD recordings of coordinated pack hunting of electric eels by Bastos & de Santana suggest coordination and communication. Superimposed strong discharges of several individuals are used to catch and prev upon swarms of highly mobile characid fish. Generally, a more advanced level of interaction is expected from these animals equipped with a rich assortment of sensorial and electrogenic systems: it is confirmed that in electric eels different EOD pulse amplitudes and frequencies are employed for stunning prey, and for electrolocation. Whether and how electric eels use their EODs for communication, in particular during courtship and pack-hunting is not known yet. We observed male antagonistic as well as initial courtship behavior. One essential factor is the state of nutrition and ovary ripening of the female, together with a potential external trigger of a sequence of change in relative conductivity, as follows Kirschbaum's approach of cyclical reproduction. The proper nesting site rather seems the problem under conditions in captivity. Since 8 months we are successfully recording EODs with a 16 channel microcontrollerbased EOD logger with stainless steel electrodes as well as corresponding videos. Twenty-fourhour recordings provide insight into a diurnal rhythm of electric activity, unexpectedly high all daytime, almost diminishing during long phases at night. The accumulated data is used to develop new algorithms for detecting and separating EOD pulses and for tracking their behavior that then can be used to interpret and analyze electrocommunication behavior of electric eels in the field. This study demonstrates how integrated field and museum studies can drive new technology for understanding the natural world.



A 10s snippet of a 16 channel recording of two eels. The electrodes were arranged along a line and were spaced by 25cm.

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Thanks are due to the fishermen of Altamira, and Leandro de Sousa, Raimundo Nonato Mendes Junior, Douglas Bastos, and Janne Ekstrom, taking care of the captured electric eels and safe transport and to Patrick Weygoldt who crafted the stainless steel electrodes.

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1 Douglas A. Bastos, Jansen Zuano, Lúcia Rapp Py-Daniel, Carlos David de Santana 2021. Social predation in electric eels. DOI: 10.1002/ece3.7121

Т6

Bursts boost nonlinear encoding in electroreceptor afferents

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Nonlinear mechanisms are at the heart of neuronal information processing, e.g. to fire an action potential, the membrane voltage must exceed a threshold nonlinearity. Even though linear encoding schemes are commonly used and often successfully describe large parts of sensory encoding, nonlinear mechanisms such as thresholds and saturation are well known to be crucial to encode behaviorally relevant features in the stimulus space. Such nonlinear encoding is not sufficiently captured by linear methods.

Here we analyze the role of bursts in p-type electroreceptor afferents (P-units) in the weakly electric fish *Apterontus leptorhynchus*. It is long known that subpopulations of these cells fire bursts of action potentials while others do not¹. Previous research suggests, that the non-bursting cells are better at encoding the stimulus time-course while bursting neurons are better suited to encode special features in the stimulus². Surprisingly, the bursting P-units fire bursts even at rest, when there are no features to encode. Also, their ability to increase bursting during stimulation is limited. What does that mean for the linear and nonlinear encoding performance of these neurons?

We show based on the analyses of experimental data and modelling that bursts affect the linear as well as the nonlinear encoding. Theoretical work³ predicts that in simple leaky integrateand-fire model neurons two stimulus frequencies interact nonlinearly when the sum of the two frequencies matches the neuron's baseline firing rate, as quantified by the second-order susceptibility. Indeed, we find such nonlinear responses also in non-bursting P-units when stimulated by two beats simultaneously, but only in those cells that exhibit very low levels of intrinsic noise. Bursts, on the other hand, strongly enhance these nonlinear responses which may play a critical role at the detection of weak intruder signals in the presence of a strong female signal, i.e. an electrosensory cocktail-party⁴.



Burst spikes enhance second-order susceptibility. The more spikes a burst contains (A), the stronger and wider are the ranges of nonlinerally boosted responses.

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Τ7

Burst firing generates invariant coding of natural electrocommunication stimuli

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Understanding how neurons process sensory information in order to successfully interact with their environment remains a central problem in neuroscience. In particular, animals need to corretly recognize various representations of sensory inputs from the same "object" under different conditions. This is thought to be achieved in the brain by having neurons responding in a similar manner (i.e., invariantly) to sensory input through identity-preserving transformations of a given "object". While such invariant representations have been observed across systems and species, the mechanisms by which they are achieved in the brain remains poorly understood to this day.

In our study, we explored how burst firing contributes to the invariant representation of electrocommunication signals ("chirps") in the electrosensory system of weakly electric fish species Apteronotus leptorhynchus. We used Neuropixels probes for multi-unit recordings from ELL pyramidal cell responses to stimulus waveforms associated with a given chirp. We found that, at the population level, bursts of action potentials tended to be elicited more reliably and similarly by different stimulus waveforms, leading to a more invariant representation than that obtained by considering the entire spiking activity. Interestingly, isolated spikes provided the least invariant representation. Additionally, we developed a biophysical model of ELL pyramidal cells to explore the intrinsic mechanisms leading to such invariant findings, highlighting the crucial role of somato-dendritic interactions in generating invariant representations. Finally, we assessed how downstream torus neurons in the hierarchy of the electrosensory circuit decode these invariant representations by developing and training a deep neural network (DNN) optimized for invariance. Overall, our results show a novel function for burst firing in establishing invariant representations by ELL pyramidal cell populations of natural electrosensory communication stimuli and suggest that such representations are decoded by downstream TS neurons to further optimize invariance.



Invariant encoding of chirp communication signals

Acknowledgements

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Electrogenic Na⁺/K⁺-ATPases constrain electrocyte activity and **T8** pose additional evolutionary pressure

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The evolution of P-type ATPases in ancestral methanogenic archaea also lay the foundation for the energetics of energy-intensive signaling tissues like the metazoan nervous systems billions of years later [1]. In particular, one of the ATPases, the Na⁺/K⁺-pump, plays a significant role in charging the batteries required to operate the nervous systems the motile life of *metazoa* is so reliant on. The Na⁺/K⁺-pump exchanges intracellular sodium for extracellular potassium ions in a 3:2 ratio, and thereby generates a net outward current. This electrogenic property of the pump appears not only as a useful exaptation for osmoregulation in eukaryotes, but also invokes activity-dependent changes in cell excitability, which lead to posttetanic hyperpolarization and firing rate adaptation [2]. These mechanisms are in turn exploited for certain neuronal encoding paradigms, cell-intrinsic bursting dynamics, and accelerated ion homeostasis [3–5]. In the weakly electric fish electrocyte however, which rapidly fires action potential trains for long stretches of time, the electrogenicy of the pump has further, less explored consequences that require special adaptations of the molecular machinery that underlies this sustained electrical activity.

We demonstrate in a computational model of the weakly electric fish electrocyte [6] extended with Na^+/K^+ -pumps and ion concentration dynamics that tonic and pacemaker-modulated high frequency firing can only be achieved through a combination of co-expression of ion channels and pumps, a very specific voltage-dependence and ion efficacy of the Na^+/K^+ -pump, or strict homeostatic regulation of extracellular space. This suggests that even though Na^+/K^+ -ATPase was jury-rigged to support the generation of action potentials in excitable cells, the fact that their original function required them to be electrogenic inevitably calls for countermeasures when the firing of these action potentials is tonic or pacemaker-driven, increasing the metabolic cost of signaling. Furthermore, it suggests that besides the evolution of ion channels that gave rise to the diversity of electrocyte signaling, a corresponding evolution related to the significant pumpinduced alterations in cell excitability due to their high energetic costs is just as important.



The pump current resulting from the electrogenic Na+/K+-ATPase (B) affects electrocyte excitability (C), and thereby its entrainment by the pacemaker (A,D). To ensure proper entrainment, these effects should be mitigated.

Acknowledgements

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Т9

The effects of urethane, MS-222, and eugenol anesthesia on the electric organ discharge of the weakly electric fish *Apteronotus leptorhynchus*

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Urethane, MS-222, and eugenol are agents widely employed for general anesthesia in fish. Yet, virtually nothing is known about their effects on CNS functions, and thus about potential interference with neurophysiological experimentation. To address this issue, we developed an in vivo assay using the readily accessible electric organ discharge (EOD) of the weakly electric fish *Apteronotus leptorhynchus* as a proxy for the neural output of the pacemaker nucleus. The oscillatory neural activity of this brainstem nucleus drives the fish's EOD in a one-to-one fashion.

Each of these three anesthetics induced, at the concentrations tested (urethane: 2.5%; MS-222: 0.02%; eugenol: 30-60 μ L/L), within a few minutes a deep state of anesthesia, as assessed by the cessation of locomotor activity. This change in locomotor activity was paralleled by a dose-dependent, pronounced decrease in EOD frequency. Upon removal of the fish from the anesthetic solution, this effect lasted for up to 30 min (eugenol) respective 3 hours (urethane, MS-222). Each of the three anesthetics also led to a significant increase in the rate of 'chirps', specific amplitude/frequency modulations of the EOD. In addition, at 60 μ L/L eugenol induced a collapse of the EOD amplitude after about 3.5 min in half of the fish tested.

We hypothesize that the reduction in EOD frequency is mediated by specific modulatory effects of urethane, MS-222, and eugenol on neurotransmission, blocking of voltage-gated sodium channels, reduction of the excitability of neurons, and potentiation of GABAA-receptor responses within the circuitry controlling the neural oscillations of the pacemaker nucleus.

The findings of our study indicate strong effects of urethane, MS-222, and eugenol on CNS functions and call for caution when conducting neurophysiological experiments under general anesthesia and when interpreting their results.



Dose-dependent effect of eugenol on EOD frequency. The minimum value of median EOD frequencies determined at 30-s intervals was related to the median EOD frequency determined during the 30 min immediately preceding the 5 of the fish's exposure to the anesthetic solution.

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Circadian regulation of gene expression in the electric organ of the **T10** gymnotiform fish *Brachyhypopomus gauderio*

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Endogenous cellular circadian clocks have been shown to be major regulators of metabolic functions. The nocturnal gymnotiform fish Brachyhypopomus quiderio has emerged as a good candidate animal system to study the cellular and molecular mechanisms underlying the circadian regulation of energy metabolism. B. gauderio produce a weakly electric signal, referred to as an electric organ discharge (EOD), for habitat navigation and communication with conspecifics. The EOD displays sexual dimorphism, circadian regulation, and is highly energetically expensive. While both sexes of *B. gauderio* exhibit larger signals in the nighttime compared to the daytime, males exhibit a much larger signal increase in the nighttime. This enhanced nighttime EOD requires a large portion of a male's energy budget to be allocated to EOD production. In addition, the EOD circadian change is mediated at the cellular level by hormones, such as melanocortins and androgens, and their effects on electrogenic membrane channels and transporters, such as voltage-gated sodium channels and sodium-potassium ATPases. However, genes associated with the link between the circadian rhythm and energy regulation have yet to be fully characterized in the electric organ of *B. gauderio*. In this study, we used direct RNA sequencing to characterize the expression of genes in the electric organ from male (n=8) and female (n=4)B. gauderio, sampled during daytime and nighttime. When comparing nighttime to daytime, we found 231 upregulated genes and 108 down regulated genes in the EO of male B. qauderio, and 32 upregulated genes and 36 downregulated genes in the EO of female B. qauderio. Using a false discovery rate (FDR) statistical approach, we identified significant differentially expressed genes and categorized these genes into molecular mechanisms and regulatory pathways that may support the EOD enhancement at night. Of note, the results of this study showed upregulation of specific genes associated with the insulin-like growth factor 2 and ATPase pathway, providing more evidence to support the importance of this pathway on the circadian regulation of the electric signal and its energetic cost in *B. gauderio*.

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Electroreceptor development and evolution: insights from comparative molecular approaches

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The mechanosensory lateral line system of fishes and aquatic-stage amphibians responds to local water movement ("touch at a distance") via lines of neuromasts in the skin that contain mechanosensory "hair cells" (very similar to inner ear hair cells). Depending on their position, neuromasts are innervated by the anterior or posterior lateral line nerves projecting to the medial octavolateral nucleus in the hindbrain. Many species (lampreys; cartilaginous fishes; most nonteleost ray-finned fishes; lobe-finned coelacanths, lungfishes and aquatic-stage urodele and caecilian amphibians) also have electrosensory organs that respond to low-frequency cathodal (exterior-negative) electrical stimuli, such as the weak electric fields surrounding other animals (primarily used for hunting). They are innervated by the anterior lateral line nerve projecting to the dorsal octavolateral nucleus. Neuromasts, electrosensory organs and their afferent neurons all develop from the same embryonic primordia: lateral line placodes. Larval paddlefish and sturgeon electroreceptors are strikingly similar molecularly to hair cells, supporting the hypothesis that nonteleost electroreceptors evolved via the diversification of lateral line hair cells. Electroreception was lost in the ancestors of neopterygian fishes (gars, bowfin, teleosts). However, physiologically distinct (anodally sensitive) electroreception evolved at least twice independently within osteoglossomorph and ostariophysan teleosts. All electroreceptive teleosts have ampullary organs that respond to low-frequency environmental electric fields. The osteoglossomorph mormyroids and the ostariophysan gymnotiforms also independently evolved electric organs that generate high-frequency electric fields and tuberous organs that respond to electricorgan discharges. Ampullary and tuberous organs are distributed on both head and trunk, innervated by the anterior or posterior lateral line nerves projecting to novel "electrosensory lateral line lobes" in the hindbrain. Preliminary gene expression data from larval ostariophysans (a siluriform, Ictalurus punctatus, and a gymnotiform, Brachyhypopomus gauderio) provide strong support for the longstanding hypothesis that teleost electrosensory organs evolved via the modification of mechanosensory neuromasts.

Neurophysiological control of refuge tracking

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Feedback - impacts of an animal's behavior on sensory information used to control behavior links sensory and motor systems in the brain. For example, weakly electric fishes use feedback to maintain their position within a moving refuge. Through a suite of behavioral studies conducted in the laboratory of Noah Cowan, we have shown how fish rely on feedback, specifically the relative movement of the refuge and fish ('error velocity'), in the control of refuge tracking behavior. Here we show how feedback is encoded in midbrain circuits and may be used in sensorimotor control. Because sensory and motor activity are linked via feedback, we expect to find correlations in neurophysiological activity of single neurons to both past sensory signals and future motor acts. We recorded 75 neurons across 11 awake, freely swimming *Apteronotus leptorhynchus* using tetrodes implanted in the electrosensory midbrain while fish performed the refuge tracking task. A population of these neurons exhibited significant changes in firing rates to both past error velocity, and future acceleration of the fish. In other words, these neurons appear to simultaneously 'encode' both sensory feedback and motor output of the fish. Such neurons may manifest a critical computation that mediates the transformation of feedback to movement for refuge tracking.

A synthetized view of the electrosensory lobe of pulse Gymnotiformes

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In *Gymnotus omarorum* the electrosensory lobe (EL) incorporates two paths originated in distinct electroreceptor types. A) a fast path in which primary afferents contact a set of spherical neurons which directly project onto the mesencephalon, and B) the focus of this presentation, a slow path, where afferent project on cerebellum-like circuit of columnar organization. The output neurons are neurons differentiated anatomically by the presence (basilar BP) or absence (non-basilar NBP) of a basilar dendrite contacted directly by afferents. Functionally, BP show peculiar intrinsic properties and the ability to fire dendritic spikes while NBP are pacemaker neurons showing spike firing adaptation [1]. Here I revise evidence indicating that i) there is a EOD to EOD packet encoding of electric images; ii) this code has two constitutive aspects spike timing and spike number per EOD; and iii) packet encoding allows novelty detection at the EL. Seven extracellularly recorded unit types were considered representative of different neuron types according to a hierarchical cluster analysis of their post-EOD spike firing distributions. A phase locked unit type firing at about 5 ms after the EOD; the other types, referred to as phase preference units, fired once every 2 or 3 EODs at in the absence of objects, showed a silence between 7 and 9 ms suggesting the presence of a strong post-EOD inhibition, and were differentiated by their characteristic locations in the EL and responses to the changes in electric image[2]. They more probably fired with preferential modes after the EOD where low pass filtering of the signals or averaging showed field potential peaks at 5–7 ms, 12–13 ms, and 23 ms. Post-EOD spike count and timing and field potentials were modulated characteristically modulated when a metal rod was moved along the skin and a large "novelty potential" [3] preceding the next EOD and graded in amplitude with that of the pacemaker acceleration was recorded when provoking novelty responses by a stepwise reduction in the impedance of an object. Consistently, with a novelty detector this potential and the spike timing and count of the different unit types characteristically varied as a function of the change in impedance and the inter-step interval of the stimulus [3]. We synthesized our present view in two computer models: one of the whole electrosensorymotor loop [4] and another of an EL column.

Acknowledgements

This talk is a fresh view of old and recent memories. I have very much to thank to too many colleagues impossible to enumerate. We enjoyed doing it and as a Uruguayan musician wrote in a song "es lindo haberlo vivido pa' poderlo contar" (It's nice to have lived it to be able to tell it).

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Reproductive signals and signal evolution in wild electric fishes: perspectives from Brachyhypopomus

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The behavioral and neurohormonal basis of reproductive signaling is well described in the model species Brachyhypopomus qauderio, but reproductive signals from wild Brachyhypopomus assemblages, as with other gymnotiforms, are poorly known. Based on analyses of signal recordings from high-diversity assemblages across the Neotropics, with accompanying phylogenetic analyses, some generalities are emerging. Closely related species are more likely to exhibit signal divergences in regions of sympatric co-occurrence than in areas of allopatry, indicating a history of reproductive character displacement (RCD). Signal divergences primarily involve temporal (shape) components of the head-to-tail EOD recorded in the far field, with EOD peak power frequency and repetition rate playing a lesser role in species recognition. In some cases, closely related species in sympatry also exhibit signs of classical ecological character displacement, such as differences in microhabitat or body size. Understanding signal divergences in hypopomids is complicated by waveform shape plasticity. Laboratory studies report prominent P2 phase elongations in mature males of several species, which may mediate intrasexual contests and/or facilitate mate attraction. Nonetheless, our nocturnal field recordings suggest that males of species capable of exhibiting P2 elongation do so only very rarely. P2 elongations express low-frequency energy content detectable to electroreceptive predators. We suspect selective pressures from electroreceptive predators greatly limit how long P2 elongations last in the wild. We recently described a novel aspect of signal evolution in Brachyhypopomus — terminal reproductive investment. Annual Brachyhypopomus species show a gradual increase in size-adjusted EOD amplitude and size-adjusted gonad mass has the end of their one and only breeding season approaches, while size-adjusted non-gonadal body mass steadily declines. In contrast, a species that lives for two years (breeding in both years) shows similar changes in the proportioning of energetic resources to those observed in annual species, but only in its second and final year, not in its first year. Terminal investment involving the EOD occurs in both sexes and may be a major driver of reproductive signaling strategies in electric fish. Long-term field-based observations of signals in the context of life history and ecology provide a powerful means of understanding signal variation and evolution.

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Evolution of Electric Organ Discharge (EOD) in African weakly electric fishes: genomics and behavioral ecology of a magic trait

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The mormyrids comprise a species-rich group of African weakly-electric fish which has undergone an enormous radiation. This is particularily true for the genus Campylomormyrus which consists of about 15 closely related species mostly restricted to the Congo basin. In combined molecular, electrophysiological, and behavioural studies, we demonstrate that (1) cryptic species are hidden behind morphometrically similar (but electrophysiologically divergent) morphotypes, (2) divergence in Electric Organ Discharge (EOD) is associated with small, but significant morphometric changes regarding the feeding apparatus, and (3) EOD is the trigger of mate recognition. In my talk, I will summarize our work on proximate (genomic, transcriptomic, and histological underpinning) and ultimate (adaptive function for orientation, feeding, and mate choice) determinants of EOD evolution and divergence. I will discuss the idea that the EOD constitutes a "magic trait", i.e., a variable heritable trait which allows both for ecological diversification and species/mate recognition.

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A molecular phylogenetic tree and DNA barcode database for Gymnotiformes

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We present a species-level phylogeny for Gymnotiformes using the largest number of species of any study to date. We constructed both maximum likelihood and Bayesian inference molecular phylogenies using nine protein-coding genes for 189 gymnotiform species, representing approximately 70% of the order. We found strong support for two large clades that correlate with electric signal: 1) wave-type fishes (Apteronotidae + Sternopygidae) and 2) pulse-type fishes (Gymnotidae + Hypopomidae + Rhamphichthyidae). While most genera were monophyletic, several paraphyletic and polyphyletic genera indicate a need for taxonomic revisions, including Apteronotus, Eigenmannia, Porotergus, and Rhabdolichops. In conjunction with our phylogenetic study, we developed a DNA-barcode database for Gymnotiformes and used it to test the taxonomic accuracy of existing gymnotiform barcodes in online databases. We found many instances of misidentified sequences in Genbank and BOLD. Overall, our tree and barcode database provide a robust framework for future evolutionary, biogeographic, and taxonomic analyses of the order Gymnotiformes.

an electric fish potassium channel

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Dual mechanisms contribute to enhanced voltage dependence of

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Many mormyrids express a voltage-gated potassium channel (Kv1.7a) in their electric organs that has two unique properties: 1) it activates at hyperpolarized membrane potentials opening quickly and, 2) once open, remains open for tens of milliseconds even as membrane potential falls back to resting potential. The first property helps to shorten the action potential and make brief EODs, the second lowers internal resistance of the EO during a discharge. We studied Kv1.7a with Xenopus oocyte expression and molecular dynamics simulation to assess the contributions of two derived molecular features of this channel to these biophysical properties: a patch of four contiguous negatively charged glutamates in an extracellular loop (the "negative patch") and a glutamate in a helix (S3b) that faces the voltage sensor. We find that these negative charges act by separate, complementary mechanisms. In the closed state, the negative patch reduces the membrane surface charge biasing the channel to enter the open state while, upon opening, the negative amino acid in the S3b helix faces the second gating charge of the voltage sensor electrostatically biasing the channel to remain in the open state. This work highlights how two evolutionarily molecular novelties influence channel biophysics and influence the mormyrid EOD.

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Spatial coding of conspecific signals in the ELL of *A. leptorhynchus*

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The ability to localize the source of a signal often relies on complex neural dynamics to extract the spatial information. In weakly electric fish, behavioral data suggests that detection and localization of conspecifics is extremely sensitive. Little is known about the neural coding principles involved in encoding the spatial information of conspecific signals beyond the fact that the lower electrosensory areas are organized in topographic maps. Here we use a combination of neural recording and large-scale modelling to examine the encoding pattern and accuracy of the population of primary sensory neurons. Our data shows that a specific subpopulation of neurons encode the spatial information significantly better suggesting that the spatial processing pathway starts diverging early in the sensory system. Furthermore, we show how network inputs shape the information content and enhances spatial contrast.

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Action potentials in the pacemaker nucleus of *Apteronotus leptorhynchus* may lack resting potential

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The high-frequency electric organ discharges (600 to 1100 Hz) are driven by the medullary pacemaker nucleus in which all neurons fire action potentials that are synchronized with each other and with electric organ discharges in a 1:1 fashion. The train of intracellular action potentials recorded from pacemaker neurons typically have a peak potential at near zero millivolt and a trough potential, which is equivalent to the resting potentials in ordinary neurons, at hyperpolarized potential levels (-75 to -40 mV). When the recording electrode (a glass capillary sharp electrode) was slightly retracted or advanced while intracellular recording, however, the action potentials shifted positively (a depolarizing voltage shift) by the magnitude of the trough potential without changing the amplitude and shape of the original intracellular action potential. As a result, the trough potential is now near zero millivolt and the peak potential is at a voltage equivalent to the magnitude of the action potential. Full size action potentials without resting potentials are known in the medial giant fiber of the Kuruma shrimp (Hsu and Terakawa, 1996) in which a large axial space between the axon and the myelin surrounding it exists. The low resistance of the axial space contributes to a very high conduction velocity (210 meter/sec). In Apteronotus pacemaker nucleus there is a high demand for high conduction velocity in its axons. If the neurons use axons with an ordinary conduction velocity of central neurons (10 meter/sec) for mutual synchronization, action potentials take 1 millisecond to cross the diameter of the nucleus. An unusually high density of astroglia has been demonstrated in the pacemaker nucleus (Sîrbulescu et al. 2014). Anatomical substrate of the positively shifted action potentials is the subject of future studies in which involvement of astroglia will be examined.

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Neuromodulator receptor gene expression in electrosensory brain **T20** regions varies across species of electric knifefishes

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Neuromodulators, including monoamines and neuropeptides, modulate signal production and perception. Motor and sensory circuits regulating communication often express neuromodulator receptors, which allow neuromodulators to differentially modulate the production and perception of signals. Whether expression of neuromodulator receptor genes in these circuits varies across species that vary in sociality, however, is less explored. Weakly electric apteronotid knifefishes are an excellent system in which to investigate neuromodulation of sensory systems. Knifefishes vary in social behavior, electric signal perception is often sensitive to neuromodulators, and the neural pathways controlling electrocommunication are well-characterized. The electrosensory lateral line lobe (ELL) and the dorsal torus semicircularis (TSd) process electric signals. We used qPCR to quantify expression of several neuromodulator receptor genes (serotonin, dopamine, vasotocin, isotocin, substance P) in the ELL and TSd across three species that vary in sociality: territorial Apteronotus albifrons, semi-social Apteronotus leptorhynchus, and gregarious Adontosternarchus balaenops. We housed fish in isolation, same-sex pairs, or opposite-sex pairs overnight and collected brains the next morning. Gene expression did not vary consistently across sex or social context. Expression of every neuromodulator receptor gene of interest, however, varied significantly across species in both the ELL and TSd. Territorial A. albifrons and semi-social A. *leptorhynchus* had greater expression of many receptor genes than social A. balaenops. These results suggest a greater potential for neuromodulation of sensory processing in species that have less frequent, but more intense or unstable social interactions. Correlated gene expression was also limited both within species across context and across species within context. Gene expression in electrosensory circuits is, therefore, highly variable, and evolutionary labile.

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Shared neural substrates for seasonal and evolutionary shifts in **T21** sensorimotor integration

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Mormyrid fishes communicate using electric organ discharges (EODs). Electroreceptors specialized for communication respond to both self-generated (reafferent) and externally generated (exafferent) EODs. To distinguish between reafferent and exafferent EODs, an internal copy of the EOD command, termed corollary discharge (CD), signals the timing of EOD production. CD briefly inhibits central electrosensory neurons to block responses to reafferent EODs. EOD duration is diverse across species, and CD timing has evolved to maintain a precise match between the timing of inhibition and reafferent sensory input. Similarly, seasonal increases in testosterone reversibly elongate male EODs, and testosterone shifts CD timing in the brain to match the resulting shift in reafferent feedback. The mechanisms that keep CD precisely time-locked to reafferent input remain unknown. Here, we identify sites of both seasonal and evolutionary shifts in the timing of CD activity within this circuit. We treated Brienomyrus brachyistius with testosterone to elongate EODs and induce CD plasticity. We sequentially recorded field potentials from six nuclei linking electromotor, CD, and electrosensory pathways. We discovered that testosterone delayed synaptic input and elongated field potentials in the mesencephalic command-associated nucleus (MCA) of the CD pathway, which shifted downstream activity relative to controls. We recorded from these same areas in two Campylomormyrus species: one with short-duration EODs (0.2 ms), and one with long EODs (2-25 ms). We found long-EOD fish also had elongated MCA activity relative to short-EOD fish. Additionally, long-EOD fish showed delayed MCA activity relative to short-EOD fish, but unlike testosterone-treated fish, delays appeared to originate in MCA neurons rather than from delayed presynaptic input. These results reveal common substrates underlying CD shifts but suggest distinct mechanisms operating on seasonal and evolutionary timescales.

From hypoxia to hyperoxia: distribution and activity patterns of mormyrids across extreme spatiotemporal environmental gradients

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How fish cope with adverse and fluctuating environmental conditions in their natural habitats remains largely unknown. In the Lake Nabugabo system in Uganda, mormyrid fish inhabit a diverse range of habitats, from severely hypoxic swamps to partially hyperoxic lake and river environments. In this study, we employed a multi-channel electric organ discharge (EOD) logger to monitor the activity and distribution of mormyrid electric fish across these varying habitats. We recorded EOD activity, temperature, air saturation (AS), light intensity, turbidity, pH, and conductivity in the main lake, an inflowing river, and the surrounding wetlands. To capture temporal dynamics, EOD activity, AS, temperature, and light intensity were continuously recorded for 24 hours per site, with sampling repeated over two months during the short rainy season of 2023. Our results extend earlier findings of distinct species distribution patterns, with Gnathonemus longibarbis and Pollimyrus nigricans predominantly found in the well oxygenated lake (88 \pm 19% AS, 26.3 \pm 1.4 \star C) and river (72 \pm 14% AS, 22.4 \pm 1.5 \star C), and Marcusenius victoriae and Petrocephalus degeni mainly inhabiting hypoxic swamp habitats (19 ś 23% AS, 24.2 ś 1.7řC). This distribution suggests hypoxia tolerance as a critical factor in habitat and resource competition among these species. All species preferred structurally complex habitats during the day but ventured into less protected areas at night, occasionally moving hundreds of meters into the open lake. This study highlights the value of autonomous EOD recording technology for ecological research. It enables the observation of unperturbed fish behavior in natural settings over extended periods, addressing a significant gap in fish ecology knowledge.



A: Map of the Lake Nabugabo system. B: Sample data from a multi-channel EOD logger that was placed at Juma River. On the right, individuals and electric organ discharge waveforms of the four mormyrid species recorded in this study are shown.

Relationship between length and electrical organ discharges (EOD) in the electric eel *Electrophorus voltai* (Gymnotiformes: Gymnotidae)

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Amazonian electric eels have fascinated scientists for more than three centuries due to their large size, anguilliform shape, strong electric discharges, and physiology of their electrical organs. One hypotheses, dating from the late 1930s, proposes a probable positive relationship between the electrical voltage (v) of electrical organ discharges (EOD) and the length of the animals, which would imply an increase in voltage as the size of Electrophorus increases. However, it is important to note that most previous records of the species' strong EOD have been obtained from specimens of the species *Electrophorus varii*, with only a single record from *E. voltai*. In the present study, we report preliminary results of an analysis of the relationship between size and electrical discharges in *E. voltai* from the Araguari river basin in the Amapá State, Brazil. Data were obtained from 10 adult specimens of *Electrophorus voltai*, size ranging from 790 to 1450 mm. This analysis shows no positive correlation between size and voltage in this population of *E. voltai* (Pearson's r = 0.3607). More samples of *E. voltai* from other locations, including the type locality in the Xingu River, as well as additional comparisons with *E. varii* and *E. electricus* are needed to better understand the variation in EOD of electric eels, as well as their respective relationships with other variables such as sex.



Relationship between Total Length (cm) and Electric Voltage (v) of *Electrophorus voltai* from the Araguari River, Amapá, Brazil

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Habitat, natural activity and ethno-cultural use of electric eels in **T24** Suriname

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Electric eels, known for their powerful electric organ discharges (EODs), inhabit the upper streams of most of the Rivers in Suriname, South-America. However, there is a gap in understanding the species' activity, ecology, and behavior in these Surinamese river systems. This research explores the habitat, natural activity, and ethno-cultural use of electric eels in Suriname, with a focus on their ecological roles and interactions with local communities. Objectives include determining species differentiation, size and EOD capacity in the Corantijn and Coppename river basins in Suriname, assessing the danger of eel shocks to humans, analyzing habitat variations in electric events and soundscapes, and exploring the ethno-cultural significance of eels among local tribes. Utilizing specialized recording equipment, the study seeks to elucidate variations in Electric Organ Discharge (EOD) patterns and behavior across different habitats and time periods. Initial findings from February 2024 fieldwork show diel electric eel activity patterns. Interviews with approximately 50 members of local tribal communities in each of the two river basins, highlighted the use of eels for food, bait, and cultural practices, with natural repellents used to mitigate dangers. Next steps include detailed morphometric and acoustic analyses, EOD frequency and pattern studies, and additional fieldwork in August 2024 to enhance data collection. This study aims to enrich scientific understanding and support local community welfare through a comprehensive examination of the ecological and cultural aspects of electric eels in Suriname.

Specialized signals for conspecific electrolocation in weakly electric fish

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Brown ghosts have been widely used as a model to study social communication in fish. During social encounters, the sinusoidal electric fields of these south American gymnotiforms are heavily modulated in frequency (through "chirps"). This phenomenon has been typically considered to represent some kind of electric code to transmit information related to status, territorial ownership, and eagerness to fight or mate. Our recent work challenged this view and showed that chirp production relates more consistently with environmental clutter and locomotion rather than other social-related variables (Oboti et al., 2023). These findings suggested that electric field modulations could be used for electrolocation purposes. In this study, we aimed at further exploring this hypothesis by assessing the reciprocal positioning of brown ghosts during chirping. The idea is that If chirps are used as probes, they must be used when fish are well positioned to probe the spatial parameters of conspecific electric fields. Recordings chirps from interacting fish pairs were used to extract body coordinates during chirps and estimate both distances and orientations. Moving playback sources were then used to measure more precisely the chirp allocentric and egocentric referencing. We found that chirps are consistently produced when playback mimics are within the brown ghost foveal RF and at ranges within which rostrocaudal asymmetries in sinusoidal electric fields can be discriminated. Eventually, by simulating electroreceptor responses and electric images induced by chirps during social encounters, we provide further evidence to corroborate the idea that chirps can be used as probes for conspecific electrolocation.

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Detection and assignment of electrocommunication signals with **T26** deep learning

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Effective communication plays a crucial role in resource competition among animals, shaping dominance hierarchies and reducing the necessity for physical confrontations. The function of chirps, a common form of electrocommunication signal in electric fish, is still a topic of debate. Past studies have mainly focused on isolated fish due to the technical limitations in detecting chirps during interactions. Here, we introduce a deep learning pipeline for automated chirp detection in freely moving Brown Ghost Knifefish to investigate the function of chirps in staged competition scenarios and beyond. The pipeline applies deep convolutional neural networks for chirp detection, which are fine-tuned using labeled spectrograms of chirping fish. Following detection, it uses custom algorithms to link detected chirps with the specific fish producing them. This methodology facilitated the identification and allocation of chirps to individual fish, resulting in a substantial dataset of over 73,000 chirps detected during unconstrained interactions. Initial analyses focused on the patterns of chirping and their behavioral associations. A notable finding is a significant correlation between chirps emitted by subordinate individuals and the cessation of aggressive interactions when competing for shelter. Still, the chirps emitted by the subordinate fish did not seem to influence the behavior of the dominant fish in a measurable manner, which highlights the need for further investigation. Beyond laboratory settings, this automated methodology can be applied to field recordings as well. Prior research has illustrated the feasibility of markerless tracking of multiple individuals in their natural environments by capturing their electric fields on electrode grids. Building on this foundation, our approach provides the capability to extract the complete electrocommunication repertoire from such recordings. This advancement now represents the opportunity to extract information concerning identity, sex, movements, and communication signals for quantitative ethological studies in undisturbed populations using data from a single modality for the first time.

Poster

P1	Arteaga Avendano : Performance and motor behaviour of <i>Gnathonemus</i> <i>petersii</i> during object detection and size discrimination tasks	37
P2	Benda : Submergeable multi-channel loggers for recording electric fish all over the globe	38
P3	Cheng : Duration divergence of electric organ discharges (EODs) in African weakly electric fish	40
P4	Comas : Intrinsic connectivity of a multipurpose central pacemaker nucleus in $Gymnotus\ omarorum$	41
P5	Crampton : A field-portable 'EOD Machine' for recording EOD waveforms and pulse rate variation in free-swimming electric fish: an integrated open- source hardware and software solution	42
P6	Iulian : Computational modeling predicts regulation of central pattern gener- ator oscillations by size and density of the underlying heterogenous network of the pacemaker nucleus in the weakly electric fish <i>Apteronotus leptorhynchus</i>	43
P7	Kul: Strategies for context-specific learning in weakly electric fish	44
P8	Lehotzky : Supervised learning algorithm for analysis of communication signals in the weakly electric fish <i>Apteronotus leptorhynchus</i>	45
P9	Macpherson : Molecular evolution of vision in South American electric fishes (Gymnotiformes)	46
P10	Mayer: Modeling the electrosensory periphery of <i>Eigenmannia virescens</i>	47
P11	Perrone : Characterization of intersexual agonistic behavior in the weakly electric fish <i>Gymnotus omarorum</i>	48
P12	Root : Context dependent responses to communication signals in the ELL of <i>Eigenmannia virescens</i>	49
P13	Sinnott : A complete ontogeny of electric organ discharges (EOD) of three intragenus crosses from the mormyrid genus Campylomormyrus	50
P14	Valiño : Hormonal modulation of agonistic behavior during the breeding season in $Gymnotus \ omarorum$	51
P15	Vázquez : Hot fish: synchronization of behavioral rhythms by temperature in weakly electric fish	52

Vollbrecht: Swamp minds: weakly electric fish <i>Petrocephalus degeni</i> balance	
brain cell proliferation and apoptosis in hypoxia	53
Weygoldt : Musical Eigenmannias: signatures of the jamming avoidance response in field recordings?	54
	Vollbrecht: Swamp minds: weakly electric fish <i>Petrocephalus degeni</i> balance brain cell proliferation and apoptosis in hypoxiaWeygoldt: Musical Eigenmannias: signatures of the jamming avoidance response in field recordings?

Performance and motor behaviour of *Gnathonemus petersii* during **P1** object detection and size discrimination tasks

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While using active sensory systems animals invest energy to perceive. Amongst these animals, weakly electric Mormyrid fish were found to gradually adapt their sensing strategy to aid this process. We are interested in discovering how *Gnathonemus petersii* adjust their electromotor strategy to gain sensory information when a task requires the detection and comparison of stimuli.

Twelve fish were trained to swim through a corridor and turn to the correspondent side of the target object. Six of them took part in a size discrimination training and the other 6 in a detection task. In both circumstances, visual input of the objects was blocked by an opaque cloth, therefore the fish very likely solved the task with their electric sense. The discrimination task involved size and relative distance as two congruent cues. In individuals reaching the learning criteria, the distance cue was inverted during test trials. In the animals successfully performing the detection task, the object was shifted farther from the decision area during testing.

Detection performance during the test phase dropped to chance level when objects were farther than 8 cm, which is consistent with the literature. Object size discrimination was achieved either when the relative distance of the two objects was the same or when they differed by no more than one centimeter, suggesting that distance may be a cue in size discrimination.

We observed a noticeable trajectory side bias in some subjects. And such tendency seemed to be more pronounced when the target object was on the correspondent side of the bias. Respectively, during the detection task such bias was reduced in those conditions surrounding the detection threshold. We hypothesized about the role of a side biased trajectory while solving object discrimination and detection tasks. We also quantified the electromotor behaviour throughout the trials to address whether different motor strategies are recruited between learning strategies.

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Submergeable multi-channel loggers for recording electric fish all **P2** over the globe

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Field research on electric fish has been limited by their secretive and nocturnal lifestyles. In the last decade, this changed with the advent of electrode grids that recorded electric organ discharges (EODs) from all gymnotiform electric fish on an multi-electrode array, from which movements and electrocommunication were extracted (Henninger et al., 2018 & 2020, Madhav et al. 2018). However, this technology was costly and not easily applicable in the field. In contrast, microcontrollers and simple analog-digital converters have been used as single-channel loggers for assessing diurnal and spatial activity patterns of mormyrid fish (Mucha et al. 2022).

We have now combined both technologies to design the Teensy_Amp R4 family of 8 or 16 channel EOD loggers, which are cheap (less than \$100) and can be easily deployed in the field (https://github.com/janscience/Teensy_Amp). The signals are pre-amplified by a factor of 10 and digitized at 16-bit with up to 96 kHz sampling rate by an Texas Instruments PCM1865 audio chip, which provides additional gain of 1 to 100x, set by software. Each channel is connected to ground via a 100 kOhm resistance, eliminating the need for an additional reference electrode. Data is recorded by a Teensy 4.1 microcontroller as WAV files onto a micro SD card (https://github.com/janscience/TeeGrid). Optionally, water temperature can be logged as well. The logger can be powered by conventional power banks. A waterproof housing can be assembled from standard PVC tube components. The electrodes can be as simple as a bundle of isolated wires with solid copper ends. The advanced version uses a single 16-core cable with a stainless-steel wire electrode every 25cm. The advantage of multi-channel loggers over single-channel ones is the improved detection of individual fishes and, of course, the larger spatial coverage.

Since autumn 2023, these EOD loggers have been successfully used in Brazil, Uruguay, Uganda, Germany, and Suriname to record activities of electric fish, including electric eels, resulting in approximately 30 TB of data. Within less than a year, our EOD loggers have proven to be effective and reliable in both field and laboratory settings, making observations on electric fish in their natural habitats accessible to everyone.



The EOD logger in its water-proof housing with status LED, temperature sensor and the initial part of the 16 core electrode cable visible on its lower end (left).. Since autumn 2023 these loggers have been used in 5 different locations in Sout-America, Africa, and Europe.

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Duration divergence of electric organ discharges (EODs) in African **P3** weakly electric fish

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In the African weakly electric fish genus Campylomormyrus, electric organ discharge (EOD) signals are strikingly different in shape and duration among closely related species, contribute to pre-zygotic isolation and may have triggered an adaptive radiation. We performed mRNA sequencing on electric organs (EOs) and skeletal muscles (SMs; from which the EOs derive) from three species with short (0.4 ms), medium (5 ms), and long (40 ms) EODs and two different cross-species hybrids. We identified several candidate genes, including KCNJ2 and KLF5, their up-regulation may contribute to increased EOD duration. Hybrids between a short (C. compressirostris) and a long (C. rhynchophorus) discharging species exhibit EODs of intermediate duration and showed imbalanced expression of KCNJ2 alleles, pointing towards a cis-regulatory difference at this locus, relative to EOD duration. KLF5 is a transcription factor potentially balancing potassium channel gene expression, a crucial process for the formation of an EOD. In addition, we compared the KCNJ2 sequences of several Mormyridae species including 9 Campylomormyrus as well as other non-electric fish samples. It showed that this gene was under disruptive selection. We'd further planned gene knock out experiment on gene KLF5 and KCNJ2 in C. tshokwe using CRISPR CAS9 to test the function of both candidate genes. Unraveling the genetic basis of the species-specific modulation of the EOD in Campylomormyrus is crucial for understanding the adaptive radiation of this emerging model taxon of ecological (perhaps even sympatric) speciation.

Intrinsic connectivity of a multipurpose central pacemaker nucleus **P4** in *Gymnotus omarorum*

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In Gymnotiformes, the medullary pacemaker nucleus (PN) commands the electric organ discharge (EOD), whose rhythmic discharge can be modulated to cope diverse environmental and behavioral demands. The PN is composed of segregated subpopulations of pacemaker cells (PMc) and relay cells (Rc) connected in a feedforward manner. Rc send the descending command to lower levels of the electromotor system to emit stereotyped EODs. Modulatory inputs to PN neurons cause electromotor behaviors with different adaptive significance depending on its specific cellular target (PMc or Rc). Electrotonic coupling (EC) between PN neurons has been proposed as a critical functional characteristic of the intrinsic connectivity of the PN. To test whether EC is capable to guarantee the many functional requirements of the PN as a robust pacemaker and a flexible effector of a wide electromotor repertoire, intrinsic connectivity of the PN of *Gymnotus omarorum* was studied in brainstem slices of juveniles using electrophysiology, pharmacology and immunohistochemistry and evaluation of dye coupling between PN neurons.

PMc-PMc and Rc-Rc pairs showed low magnitude EC through non-rectifying contacts with a low-pass filter behavior. At the PMc, this could contribute to a robust yet modifiable pacemaker behavior of the PN; at the Rc would be suitable for the generation of a synchronous descending command. Weak PMc-Rc EC was also detected. This connection was non-rectifying and bidirectional but with direction-dependent filter behavior: high-pass in the PMc-Rc direction and low-pass in the Rc-PMc direction. Directionality and adequate timing of PMc and Rc activation would result from the existence of EC through axo-dendritic or axo-somatic connections and special anatomical characteristics of the PMc axon. The effect of specific blockers, the presence of dye coupling between PN neurons and immunohistochemical results suggest that neurons of the PN are connected via gap junctions probably formed by Cx35.



Schematic drawing of the putative intrinsic connections of the PN showing the signal transfer capability of different synaptic contacts and their possible functional roles.

A field-portable 'EOD Machine' for recording EOD waveforms and **P5** pulse rate variation in free-swimming electric fish: an integrated open-source hardware and software solution

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Several studies have used an 'EOD machine' to characterize the EOD waveforms of free-swimming electric fish. EOD machines detect the presence of a fish in a trigger zone at the center of the tank as it swims through an electrically transparent tube between tank-end compartments. We developed a hardware-software EOD machine based on a field-portable tank, custom amplifier and microcontroller, and custom software. Our arena comprises a modified 140 L cooler with a plastic mesh tube connected via fine-mesh funnels to end compartments. A cloth cover at the mesh tube center attracts the fish to the tank center by day. We designed a circuit board comprising a microcontroller and two variable-gain bio-amplifiers. Two such boards permit signals from four electrode pairs to be synchronously recorded on one microcontroller as consecutive 30 s long, 4-channel .WAV files. These recordings are stored and analyzed subsequently with a custom R program, parallelized for high-speed looping through 24+ hours of 30 s files. Four Ni-Cr electrode pairs are positioned in the arena, one at a 10 cm spacing in the mesh tube center (trigger electrode), one at the tank ends, and two positioned orthogonally across the tank's sides, each around 1/3 from the tank-end. The trigger electrode, tank-end electrode, and tank-side electrodes correspond to channels 1 through 4, respectively in each .way file. Our program separates the four channels of each .wav file, converts from PCM to floating source, and applies denoising digital filters. A trigger threshold is then used to index a specified number of EOD as the fish passes through the center of the trigger electrodes. Pulses with equivalent indices are then extracted from the tank-end electrode channel and used to computer an averaged EOD for analyses of amplitude (calibrated to a mV/cm at 10 cm value), duration, and integral. The recordings from all four electrodes are rectified and summed together, and the sequence of inter-pulse intervals logged with a peak-detection routine. Pulse rate is characterized for every recording, even when the fish does not swim through the trigger zone. Our program plots multiple aspects of the amplitude-calibrated EOD waveform and EOD pulse rate over a 24-hr period. We also calculate a proxy for fish movement based on the first derivative of amplitude fluctuation across the four electrodes. Our system works for electric fish under 30 cm body length, including species with all known EOD waveform types.

P6

Computational modeling predicts regulation of central pattern generator oscillations by size and density of the underlying heterogenous network of the pacemaker nucleus in the weakly electric fish *Apteronotus leptorhynchus*

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Central pattern generators are characterized by a heterogeneous cellular composition, with different cell types playing distinct roles in the production and transmission of rhythmic signals. However, little is known about the functional implications of individual variation in the relative distributions of cells and their connectivity patterns. Here, we addressed this question through a combination of morphological data analysis and computational modeling, using the pacemaker nucleus of the weakly electric fish Apteronotus leptorhynchus as case study. A neural network comprised of 60-110 interconnected pacemaker cells and 15-30 relay cells conveying its output to electromotoneurons in the spinal cord, this nucleus continuously generates neural signals at frequencies of up to 1 kHz with high temporal precision. We systematically explored the impact of network size and density on oscillation frequencies and their variation within and across cells. To accurately determine effect sizes, we minimized the likelihood of complex dynamics using a simplified setup precluding differential delays. To identify natural constraints, parameter ranges were extended beyond experimentally recorded numbers of cells and connections. Simulations revealed that pacemaker cells have higher frequencies and lower within-population variability than relay cells. Within-cell precision and between-cells frequency synchronization increased with the number of pacemaker cells and of connections of either type, and decreased with relay cell count in both populations. Network-level frequency-synchronized oscillations occurred in roughly half of simulations, with maximized likelihood and firing precision within biologically observed parameter ranges. These findings suggest the structure of the biological pacemaker nucleus is optimized for generating synchronized sustained oscillations.

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Ρ7

Strategies for context-specific learning in weakly electric fish

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Learning enables animals to adapt to changing environments. Simple forms of learning are known to be widely distributed throughout the animal kingdom, while complex forms of learning, such as context dependent learning, have mainly been studied in higher order vertebrates. Using a two alternative forced-choice paradigm, we investigate whether G. petersii (mormyrids) is capable of context specific object/place association. These fish produce electric signals (EOD) to orient in their environment. They can change their EOD rate depending on demand. Fish were trained to distinguish between sparse and dense environments. Fish were trained to indicate the perceived context by swimming to either of two spatially fixed goal compartments that contained a plastic or a metal object. Hence, fish could solve the task by either associating a context with these objects or with a fixed location. Preliminary data shows that fish are capable of contextdependent learning for which fish appear to associate both target object identity and object position with the context. Furthermore, task-acquisition is associated with altered swimming patterns. Fish took stereotypic paths through the set-up that changed when fish learned the task; e.g., one fish initially swam along the border of the setup, entering only one of the two goal compartments. Task-acquisition in this individual was associated with a switch of the trajectory towards sampling the arena from the middle of the setup. In general, inbound trajectories are more stereotyped than outbound trajectories. Fish sample their environment with a higher sampling rate (EOD/sec) on the inbound path. Sampling along the stereotyped trajectories appears to be object-specific, indicating that individual objects are of more importance in the evaluation of the context. Indeed, removing single objects increased the likelihood of incorrect choices during probe trials.

P8

Supervised learning algorithm for analysis of communication signals in the weakly electric fish *Apteronotus leptorhynchus*

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Signal analysis plays a preeminent role in neuroethological research. Traditionally, signal identification has been based on pre-defined signal (sub-)types, thus being subject to the investigator's bias. To address this deficiency, we have developed a supervised-learning algorithm for the detection of subtypes of chirps — frequency/amplitude modulations of the electric organ discharge that are generated predominantly during electric interactions of individuals of the weakly electric fish *Apteronotus leptorhynchus*. This machine learning paradigm can learn, from a 'ground truth' data set, a function that assigns proper outputs (here: time instances of chirps and associated chirp types) to inputs (here: time-series frequency and amplitude data). By employing this artificial-intelligence approach, we have validated previous classifications of chirps into different types and shown that further differentiation into subtypes is possible. This demonstration of its superiority compared to traditional methods might serve as proof-of-principle of the suitability of the supervised-machine-learning paradigm for a broad range of signals to be analyzed in neuroethology.

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P9

Molecular evolution of vision in South American electric fishes (Gymnotiformes)

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Vision in vertebrates is initiated by a photon of light absorbed by a visual pigment-chromophore complex within the photoreceptor cells of the eye. This triggers activation of a visual transduction cascade that eventually leads to a neural signal that light has been perceived. Neotropical electric fishes (Gymnotiformes) are a widespread and diverse freshwater group that is known to use electroreception for activities that in other fishes would be mainly visually guided. As these fishes tend to be nocturnal, and generally have small eyes, we sought to investigate whether there is evidence for a sensory tradeoff with vision. Using comparative analyses of selection, we found evidence for adaptive shifts in spectral tuning, but no evidence for relaxed selection that would be consistent with a sensory tradeoff hypothesis.

Modeling the electrosensory periphery of *Eigenmannia virescens* P10

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The electrosensory system of the electric fish is a well established model system for electrophysiological studies and computational analyses. Electric fish such as *Eigenmannia virescens* surround themselves with an electric field by regularly discharging their electric organ (Electric organ discharge, EOD). At the same time, cutaneous electroreceptors, namely the tuberous (active) and ampullary (passive) electroreceptors, sense distortions of the own field as well as exogeneous fields. These signals are evaluated and used in the context of navigation, prey detection and electrocommunication. First, peripheral information undergoes a non-linear transformation into on- and off pathways when relayed to pyramidal neurons in the hindbrain. There, active and passive information is kept separated. Then, on the level of the midbrain (torus semicircularis, the homologue to the inferior colliculus), these parallel streams are integrated. So far, none of these transitions are fully understood.

As a first step to address these fundamental transitions of a neuronal code we here aim at modeling the electrosensory periphery by fitting leaky integrate-and-fire (LIF) models to individual electroreceptor afferents, based on spontaneous and stimulus driven response properties. For this we characterize the response properties of the recorded neurons and then individually fit models using the simulation-based-inference approach¹.

Creating such a library of realistic model neurons will enable us to analyze computational principles. In particular to test the hypothesis of the beneficial role of integrating information carried by the active and the passive electrosensory system in the context of electrocommunication in $Eigenmannia^2$.

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Characterization of intersexual agonistic behavior in the weakly electric fish *Gymnotus omarorum*

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Agonistic behavior, present in all animal species, regulates conflict situations over limited resources. The genus Gymnotus is noted for its aggressive behavior, which becomes violent when agonistic encounters take place in confined spaces. Gymnotus omarorum is a weakly electric fish, and although it is a seasonal breeder, it shows aggression throughout the year, between males, between females, and most strikingly, between males and females, even during the reproductive period. It is a monomorphic species, there are no external sexual differences, neither anatomical nor electrical, not even during the reproductive period. Our group has extensive knowledge of the factors that modulate the agonistic behavior of this species in intrasexual dyads. Intersexual aggression is rare in nature, but its occurrence in the human species, gender-based violence, is the cause of many social and health problems. To find experimental models in which to explore the neural bases of this behavior is of great interest, and this species is the ideal model to study them. Here we describe the agonistic behavior of intersexual dyads of *Gymnotus omarorum* in their locomotor and electrical displays during the reproductive and non-reproductive seasons in semi-natural conditions. In most cases the heavier animal wins, regardless of sex. The conflict was longer during the non-breeding season than in the breeding season. No differences were found in the attack rate between dominant males and dominant females, nor between subordinate males and subordinate females. In the non-breeding season, only few transient electrical signals were observed, in contrast to the breeding season, when these signals are abundant. No differences were found in the duration of the conflict when females won from when males won. Surprisingly, there are no differences between the way females and males fight even in the reproductive period. We want to investigate the differences between this type of aggression and courtship.



An intersexual dyad of *Gymnotus omarorum* registered in the field laboratory during breeding season

P12

Context dependent responses to communication signals in the ELL of *Eigenmannia virescens*

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The weakly electric fish *Eigenmannia virescens* uses its electric field for navigation, prey detection, and communication, making it an ideal model for studying neuronal communication processes. In communication contexts, it employs interruptions of its self-generated field to convey information. These interruptions lead to signals that stimulate all three electrosensory pathways: the P- and T-units of the tuberous as well as the ampullary receptors. Accordingly, the receiving fish perceives these with all three types of electroreceptors. Previous work (Stöckl, 2014) described the neuronal responses of all three types of electroreceptors to interruption, revealing what is transmitted to pyramidal cells in the electrosensory lateral line lobe (ELL).

In a solitary context, i.e. when the fish chirps without another animal being present, the responses to interruptions in pyramidal cells were found to be stereotyped for each cell type (Metzner and Heiligenberg, 1991). Our current research aims to explore how pyramidal cells in the active maps of the ELL respond to these signals when they occur in the context of communication, i.e. when another animal is present. This study, which involved in vivo intracellular single-unit recordings of pyramidal cells in the ELL of *Eigenmannia virescens* during communication with conspecifics, revealed a more complex picture. Our electrophysiological data show that pyramidal cells can exhibit various response types depending on the properties of the stimuli. This indicates that a single neuron in the ELL can transmit different kinds of information based on the context.

These findings align with the complex environmental signal that *Eigenmannia virescens* must encode and interpret in its natural habitat, highlighting the complexity of its neuronal communication system.

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P13

A complete ontogeny of electric organ discharges (EOD) of three intragenus crosses from the mormyrid genus Campylomormyrus

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Weakly electric fish (WEF) communicate, navigate, and forage using discharges from their electric organ (EO) — EODs. Species of the pulse-type WEF genus Campylomormyrus exhibit a dramatic diversification in the shape and duration of their EODs, and questions remain regarding the ultimate mechanisms for this diversification. In a previous study utilising artificial breeding methods, intragenus crosses between five different Campylomormyrus species produced viable F1-hybrids (Kirschbaum et al. 2016). This study aimed to combine species differing in both EOD characteristics and EO structure, and successfully described the ontogeny of the EO and EOD of seven intragenus hybrids up to an intermediate stage of development. However, in pure Campylomormyrus species, it has been demonstrated that the shape and duration of the EOD can continue to change up to adulthood and sexual maturity (Kirschbaum et al. 2016, Nguyen et al. 2017). Here we complete ontogenetic description of the EOD for three F1-hybrids from the previous study - C. tshokwe (Cts) x C. tamandua (Ct); C. tshokwe x C. compressirostris (Cc); and C. rhynchophorus (Cr) x C. tshokwe — by including the EODs of adult, sexually mature fish. These hybrids were produced from species emitting both long (ca. 5-20ms, C. tshokwe and C. rhynchophorus) and short (ca. 200-250ts, C. compressirostris and C. tamandua) EODs; and species with both caudal (C. compressirostris, C. tshokwe and C. rhynchophorus) and rostral (C. tamandua) positions of the main EO stalk. In two out of the three crosses (Cts x Cc and Cts x Ct), EOD ontogeny is very similar, presenting a simple biphasic EOD of ca. 200ts duration when juvenile, then closely following the ontogeny of C. tshokwe. However, the development of these EODs appear to arrest at an intermediate C. tshokwe stage, presenting an EOD typical of a ca. 50mm C. tshokwe. In the third cross, Cts x Cr, again a typical juvenile EOD is presented, though with a slightly longer duration (ca. 250 ts). The EOD again progressed during ontogeny in a manner closely resembling the development of the pure Cts EOD, though in this cross the development continued to adulthood. The EOD shape of the C. rhynchophorus parent did not present at any stage in the ontogeny of this hybrid. Thus, in the EOD ontogeny of these three F1-hybrids, C. tshokwe EOD characteristics appear to dominate over those of the other three parental species.

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Hormonal modulation of agonistic behavior during the breeding **P14** season in *Gymnotus omarorum*

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Agonistic behavior, an adaptive social behavior arising from competition among conspecifics for limited resources, is regulated by sex steroids, primarily androgens and estrogens. While weakly electric fish are a pioneering animal model in neuroethology, the study of agonistic behavior and its hormonal modulation has focused on select species. One such species, Gymnotus omarorum, a gymnotiform native to Uruguay, has been a well-established model for studying the modulation of non-breeding agonistic encounters in both males and females. However, the agonistic behavior of G. omarorum during the breeding season and its hormonal modulation have not been previously explored. This study presents, for the first time, a characterization of G. omarorum breeding agonistic encounters in both sexes. Intrasexual dyads (n=24 male dyads and n=22 male dyads and n=22female dyads) were observed under semi-natural conditions, utilizing wild-caught fish isolated in their natural habitat prior to behavioral experiments. Both sexes engaged in highly aggressive encounters, leading to the establishment of a clear dominant-subordinate hierarchy following the conflict phase. Subordinate individuals of both sexes emitted electric signals (chirps and offs) to indicate their status. Furthermore, the role of androgens and estrogens in modulating this behavior was investigated. Estrogen synthesis was inhibited with an acute injection of fadrozole administered 30 minutes prior to interaction (n=7 control and n=11 treated male dyads, and n=10 control and n=12 treated female dyads). The results indicated that estrogens modulate the motivation to engage in fights in females and influence aggressive behavior once the hierarchy is established in males. Additionally, and rogen receptors were inhibited with an acute injection of cyproterone acetate administered 30 minutes before interaction (n=10 control and n=8 treated male dyads, and n=6 control and n=7 treated female dyads). The findings revealed that androgens only modulate the emission of offs in male subordinates and had no effect on female encounters. These results demonstrate that androgens and estrogens modulate breeding agonistic behavior via rapid mechanisms, most likely non-genomic. Furthermore, this hormonal modulation is sexually dimorphic during this season, in contrast to the monomorphism occurring during non-breeding agonistic encounters. Ultimately, this study highlights seasonal variation in the underlying mechanisms.

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Hot fish: synchronization of behavioral rhythms by temperature in P15 weakly electric fish

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Photoperiod acts as the main zeitgeber of physiological and behavioral daily rhythms (1), yet its unreliability in certain habitats has prompted animals to utilize other environmental cycles as alternatives. In poikilothermic animals, like the weakly electric fish *Gymnotus omarorum*, temperature emerges as a likely candidate (2). In murky, vegetation-laden waters where photoperiod cues are unreliable, G. omarorum exhibits robust nocturnal activity patterns, including heightened exploratory behavior and an increase in the rate of their electric organ discharge (EODr) (3,4). These changes in EODr are a behavioral display that is modulated in response to the perceptual and social needs of the animal (4,5). To explore the role of temperature as a synchronizer, we devised a semi-natural experimental setup mimicking their habitat while enabling high-throughput positional data and EODr value acquisition. By using this set-up we were also able to eliminate photoperiodic information while maintaining environmental temperature cycles, balancing the complexity of natural environments with controlled laboratory conditions. G. omarorum displayed a daily rhythm of locomotor activity and EODr increase even when the light/dark cycle is suppressed. This showcases that periodic light cues are not necessary for these animals to maintain their behavioral rhythms. Lastly, rhythms maintained constant phase relationships with natural temperature cycles, indicating temperature as the primary circadian synchronizer in both lighting conditions, which suggests that, in this species, temperature acts as the main synchronizer of circadian rhythms. Our study illuminates the adaptive strategies of species within habitats characterized by extreme lighting conditions, and underscores the significance of understanding behavior expression under natural conditions.

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Swamp minds: weakly electric fish *Petrocephalus degeni* balance **P16** brain cell proliferation and apoptosis in hypoxia

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Brain dysfunction during warming is triggered by a cascade of pathological changes stemming from oxygen limitations in aquatic habitats, significantly impacting the ecology, behavior, and physiology of fishes. We investigated apoptosis and compared it to brain cell proliferation in the brain of the weakly electric fish *Petrocephalus degeni*, shedding light on their adaptation mechanisms to the extreme hypoxia they experience in their natural habitat in swamps in Uganda. These fish inhabit an environment with minimal oxygen availability yet exhibit typical mormyrid brain characteristics, i.e. a very large brain with a gigantocerebellum. Thirty-three individuals were collected, with 11 sacrificed in the field and 22 exposed to artificial hypoxia or normoxia in a laboratory setting. PCNA analysis revealed that swamp-dwelling field fish displayed the highest rates of brain cell proliferation, with normoxic fish significantly outperforming hypoxic lab counterparts. Additionally, a TUNEL analysis staining apoptotic cells showed a higher apoptotic rate in fish in the hypoxic lab conditions than in fish from the swamp or kept under normoxic conditions in the lab. Our results suggest strong effects of both, oxygen regime and captivity, on the balance between brain cell proliferation and apoptosis, and remarkable resilience of the brains of these fish under naturally hypoxic conditions.

P17

Musical Eigenmannias: signatures of the jamming avoidance response in field recordings?

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When walking through a neotropical stream and listening to groups of gymnotiform wave-type electric fish with a fishfinder, the superimposed EODs usually sound pleasant, as if in harmony to each other. This suggests that the relative EOD frequencies of near-by electric fish are close to musical intervals like the minor and major third, the fourth, and the fifth. To check this hypothesis we analyzed various 16- to 64-channel logger and grid recordings obtained in Darien (Panama), Meta (Colombia), Leticia (Colombia), and Amapa (Brasil). From each data snippet (every minute a 10s snippet on every channel) we extracted 16 million fundamental frequencies of potential EODs (Henninger et al. 2020), calculated the ratio of the EOD frequencies in each data snippet, and checked whether ratios corresponding to musical intervals are preferred by the fish. We found that Eigenmannia did indeed prefer the major third, the fourth, and the major sixth, both in Darien and Leticia, and the fifth in Meta. In Apteronotus this was sometimes, but weakly expressed. In all other species groups, Sternopygus, Archolameus and Sternachella, we did not observe such a preference. This seems to coincide with the strength of the jamming avoidance response (Stamper et al. 2012) and also needs to be discussed in the context of the encoding of beats by electroreceptor afferents (Barayeu et al. 2023).



Probability densities of EOD frequency ratios (left) and histograms of EOD frequencies (right) computed from grid recordings in Panama, May 2014 (Eigenmannia) and in Colombia, October 2019 (Apteronotus).. Note the aligment of EODf ratios to musical intervals.

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Author index

Ahmed, Mariam, 15 Akhshi, Amin, 11 Arteaga Avendano, Maria Paula, 37 Ayoubi, Sirine, 33 Baker, Clare, 18 Barayeu, Alexandra, 9 Bartsch, Peter, 7 Bashivan, Pouya, 11 Baumann, Otto, 22 Benda, Jan, 3, 7, 9, 34, 38, 47, 54 Borde, Michel, 41 Caputi, Angel, 20 Carlson, Bruce, 28 Carnevale, Vincenze, 24 Chacron, Maurice, 11 Chang, Belinda, 46 Cheng, Feng, 22, 40 Comas, Virginia, 41 Cox, Shann, 17 Crampton, William, 6, 21, 23, 42, 46 Datovo, Aléssio, 30 de Santana, Carlos David, 30 de Santana, David, 7 Dunlap, Kent, 53 Elarbani, Khouloud, 50 Engelmann, Jacob, 37 Eske, Annika, 15 Fleig, Wilhelm, 33 Fortune, Eric, 19 Freiler, Megan, 27 Grewe, Jan, 9, 33, 47, 49 Haag, Michael, 6, 42 Hauser, Frances, 46

Henninger, Jörg, 54 Huang, Chen, 23

Immani, Swapna, 24 Iulian, Iulian, 43

Jalabert, Cecilia, 5 Janzen, Francesco, 21, 23 Jarzyna, Martin, 28 Juby, Nicole, 23 Jung, Sarah, 44

Kawasaki, Masashi, 26 Khadra, Anmar, 11 Kirschbaum, Frank, 7, 22, 50Klovdah, Kaitlin, 53 Korniienko, Yevheniia, 50 Krahe, Rüdiger, 29, 33, 53, 54Kul, Nur-Gizem, 44

Lehotzky, Dávid, 15, 45 Lindner, Benjamin, 9 Lovejoy, Nathan, 21, 23, 46

Macpherson, Esme, 46 Marsat, Gary, 25 Mayer, Sarah, 47 Mendes-Junior, Raimundo Nonato, 30 Metzen, Michael, 11 Migliaro, Adriana, 3, 52 Milam, Oak, 25 Molina, Jorge, 4, 6, 54 Mucha, Stefan, 3, 29, 38

Narain, Devika, 32 Nguyen, Linh, 50 Nzimora, Kingsley, 50

Oboti, Livio, 33 Osorio-Ospina, Juan, 4 Pedraja, Federico, 3, 33 Perrone, Rossana, 48, 51 Poon, Lok, 6, 21, 42 Pouso, Paula, 41

Quintana, Laura, 5, 51, 52

Raab, Till, 34, 54 Ramachandra, Keshav, 25 Root, Kathrin, 49

Sabaj, Mark, 23 Salazar, Vielka, 17 Schleimer, Jan-Hendrik, 13 Schlungbaum, Maria, 9 Schneck, Christina, 33 Schreiber, Susanne, 13 Sehuanes, Juan, 54 Silva, Ana, 3, 5 Sinnott, Andrew, 50 Smith, G. Troy, 27 Soma, Kiran, 5 Suma, Antonio, 24 Tiedemann, Ralph, 22, 40, 50

Todorovic, Jelena, 24

Valiño, Guillermo, 51 Vollbrecht, Marie-Luise, 53 von Drenkmann, Helen, 33 Vázquez, Juan, 52

Waddell, Joseph, 21 Weerdmeester, Liz, 13 Weygoldt, Patrick, 34, 38, 54

Zakon, Harold, 24 Zubizarreta, Lucia, 5 Zupanc, Günther, 15, 43, 45

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