# Modelling the Spinal Cord of a Tadpole

Exploring different ways to model the spinal cord in the *Xenopus* frog

by

**Alexia Wilson**

December, 2025

A thesis submitted to the
Graduate School
of the
Institute of Science and Technology Austria
in partial fulfillment of the requirements
for the degree of
Master of Science

Committee in charge:
Tim P. Vogels
Lora B. Sweeney
Maximilian Jösch

The Master's thesis of Alexia Wilson, titled "Modelling the Spinal Cord of a Tadpole: Exploring different ways to model the spinal cord in the *Xenopus* frog", is approved by

| | | |
|---|---|---|
| ..... | ..... | ..... |
| **Supervisor:** | **Co-Supervisor:** | **Committee Member:** |
| Tim P. Vogels,<br>ISTA, Klosterneuburg,<br>Austria | Lora B. Sweeney,<br>ISTA, Klosterneuburg,<br>Austria | Maximilian Jösch,<br>ISTA, Klosterneuburg,<br>Austria |

Signed page is on file.

ISTA Master's Thesis, ISSN: 2791-458

I hereby declare that this thesis is my own work and that it does not contain other people's work without this being so stated; this thesis does not contain my previous work without this being stated, and the bibliography contains all the literature that I used in writing the dissertation.

I declare that this is a true copy of my thesis, including any final revisions, as approved by my thesis committee, and that this thesis has not been submitted for a higher degree to any other university or institution.

I certify that any republication of materials presented in this thesis has been approved by the relevant publishers and co-authors.

.....

**Alexia Wilson**

December, 2025

Signed page is on file.

# ABSTRACT

Left–right alternation is a defining feature of spinal locomotor circuits, yet the level of neuronal detail required to generate and maintain this pattern remains unclear. This thesis investigates how models spanning multiple levels of abstraction—from biophysically detailed Hodgkin–Huxley (HH) neurons to adaptive integrate–and–fire (I&F) formulations and synfire-chain modules—can account for the generation of fictive swimming in the spinal cord of the *Xenopus laevis* tadpole. The guiding hypothesis is that a small set of neuronal mechanisms is sufficient to reproduce the essential features of rhythmic alternation, and that moving between modeling scales helps distinguish core principles from biological detail.

A minimal bilateral HH network comprising only four canonical neuron classes—excitatory descending interneurons (dINs), inhibitory commissural interneurons (cINs), ipsilateral inhibitory interneurons (aINs) and motoneurons—served as a biophysical proof of concept. Tuned to reproduce experimentally observed firing modes, the model demonstrated that rebound-prone dIN excitability, contralateral inhibition and modest electrical coupling are sufficient to generate stable alternating activity, even in very small networks. These results motivated the transition to simpler models capable of efficient analysis and scaling.

Adaptive exponential I&F (AdEx) neurons were calibrated to physiological recordings using simulation-based inference, yielding tonic and phasic/rebound templates that preserved the key dynamical signatures of the HH model. Phase-plane analysis clarified the mechanisms underlying single-spike responses and rebound firing in dINs. At network level, the I&F models robustly reproduced left–right alternation, while highlighting constraints on synaptic kinetics and adaptation needed to avoid multi-spike responses.

Finally, a synfire-chain framework provided a complementary, timing-centric perspective, demonstrating how precise spike synchrony, synaptic delays and minimal inhibitory coupling can generate alternating left–right sequences in a feedforward setting. Together, these approaches converge on a common conclusion: rebound-prone ipsilateral excitation combined with precisely timed contralateral inhibition constitutes a sufficient substrate for alternating spinal rhythms.

By integrating bottom-up and top-down modeling strategies, this thesis provides a unified, extensible framework for studying spinal pattern generation. The results show that essential locomotor dynamics can be captured across multiple abstraction levels, offering both mechanistic insight and practical tools for future data-driven investigations of spinal circuit development, robustness and modulation.

# ACKNOWLEDGEMENT

This work was made possible through the generous support of my grant funding (IPC 61), for which I am deeply grateful.

I would like to express my sincere gratitude to my supervisors, Tim P. Vogels and Lora B. Sweeney, for their guidance, support and insight throughout this thesis. Their expertise, critical feedback and encouragement shaped both the scientific direction of the work and my development as a researcher.

I am also grateful to the members of both labs for creating a stimulating, supportive and collaborative research environment. Discussions, feedback and shared problem-solving greatly enriched this work. I thank past and present lab members for their generosity with time, ideas and code, and for making the day-to-day process of research both productive and enjoyable.

I would like to thank Jonas Ingmanns for his invaluable support, advice and encouragement throughout this journey.

I am lastly grateful to the Institute of Science and Technology Austria (ISTA) for providing the necessary resources and facilities to complete this project to the best of my ability.

# About the Author

Alexia Wilson completed a BA in biological Natural Sciences at the University of Cambridge in 2020. She then joined the Vogels lab at ISTA for a scientific internship. During the intership she applied to and got accepted for the PhD Program at ISTA, starting in 2021. Following a year of rotations, she joined the Vogels and Sweeney labs to be co-supervised by Tim P. Vogels and Lora B. Sweeney. In 2023 she wrote a review on spinal interneurons together with Lora B. Sweeney, published in *Frontiers*. Due to health problems she decided to terminate the PhD early with a Masters Degree. The PhD project was therefore turned into a Masters Thesis.

# List of Collaborators and Publications

Alexia C. Wilson and Lora B. Sweeney, 2023. Spinal cords: Symphonies of interneurons across species. **Frontiers in Neural Circuits**, Volume 17. Re-used in whole in Chapter 1.

# Contents

# List of Figures

# 1 Review of Spinal Interneurons in Mouse and Zebrafish

**Statement of prior publication:** *This chapter is an umodified reproduction of my previously published review article [355].*

## 1.1 Introduction

Vertebrates exhibit a wide range of movement patterns. Across species and evolutionary time, they have transitioned from axial-based swimming to limb-based locomotion. Between species, they have uniquely adapted their movement repertoires to their environment, physiological needs, and mode of locomotion (Fig.1.1; [172, 319, 16]). Fish, for example, rely on precise and alternating contraction of segments along the rostrocaudal axis to generate slow, undulatory swimming. Mice coordinate the flexor and extensor muscles of the limb to grasp food pellets, run on a wheel, swim, and perform stereotyped repetitive grooming behaviors. Frogs adopt fish-like undulatory movement as tadpoles, transition to limb-based locomotion during metamorphosis, and as adults, predominantly rely on synchronous limb movement [78, 269, 274, 19, 82]. This contrasts with other amphibians, such as salamanders, which maintain both undulatory tail and alternating limb movement throughout life. Like salamander, limbed reptiles, and most mammals, including mice and humans, similarly alternate their limb muscles at all speeds as a default behavior [146, 281, 188]. Notable exceptions to this are snakes, which have lost their limbs and exhibit only axial body movement. These many differences in movement between species raise the question of how their underlying motor circuits differ.

### 1.1.1 Spinal Circuit and Function

Over a century ago, Sherrington and Brown demonstrated that motor circuits of the spinal cord were the core of movement generation. Sherrington highlighted the integrative nature of spinal, sensory and central circuits in reciprocal motor action in the cat [313]. Brown then proposed the half-center model, in which rhythm is generated by two half-centers in the spinal cord that reciprocally inhibit each other [51]. Together, these two half centers, and their constituent spinal circuits, were dubbed central pattern generators (CPGs). Experimental evidence for such a CPG organization and

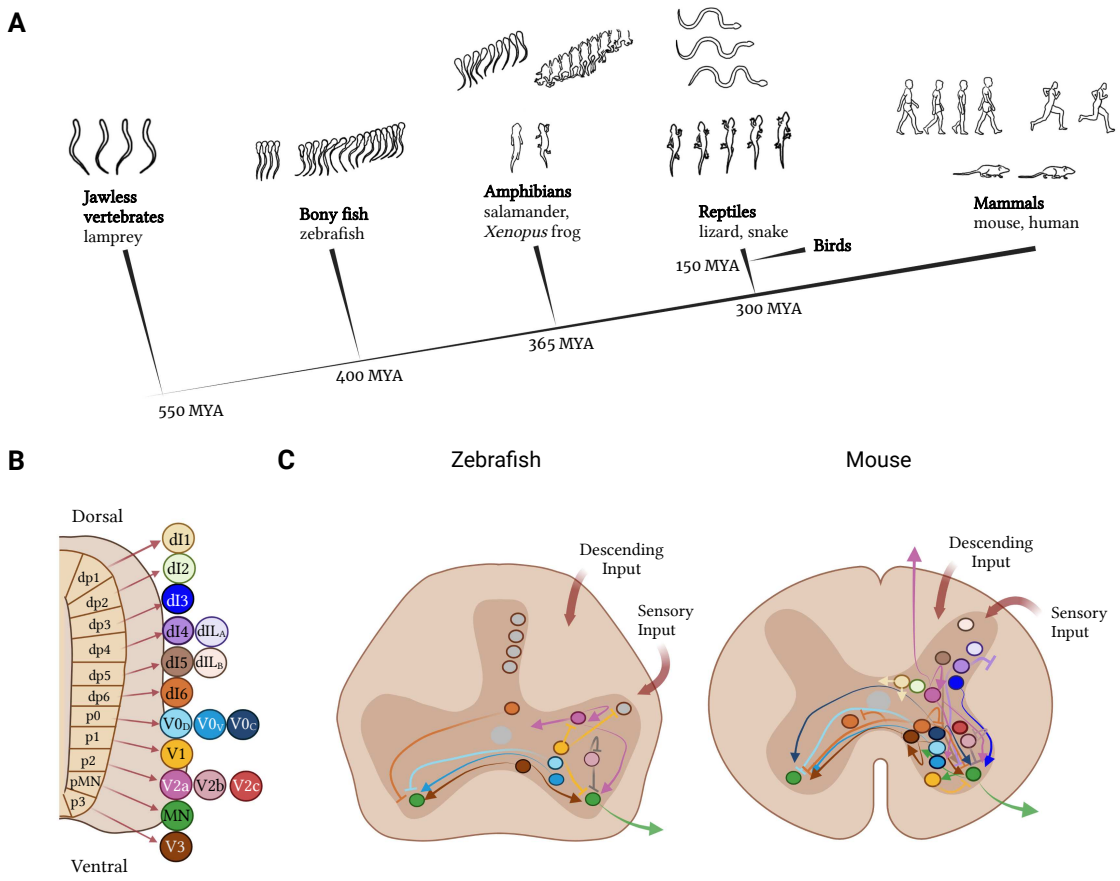


**Figure 1**

Figure 1.1: A cross-species comparison of the neural basis of vertebrate movement. (A) Cladogram of vertebrate evolution with illustrations of movement patterns for each of the species listed as examples. The lamprey is the most primitive vertebrate and exhibits simple, undulatory swimming; zebrafish display more complex swimming patterns; the frog and salamander use both tail and limbs for movement; reptiles exhibit diagonal limb coordination; and mammals display complex fore-/hindlimb gaits. (B) Cardinal neuron classes that make up the spinal cord circuitry are derived from 11 progenitor domains. Some domains give rise to more than one neuron class, e.g., the p2 domain gives rise to the V2a, V2b, and V2c interneurons. (C) Comparison of interneuron subtypes and projection patterns in the spinal cord of zebrafish versus mice. Colors represent different neuron classes; gray represents neurons without a clear cardinal class identity.

initial characterization of spinal reflex behaviors was first described in invertebrates, and then, in the spinalized cat [356, 121, 124, 122, 225, 282, 333]. Although evolutionarily distant, in both, "normal" locomotor patterns with appropriate excitation were present even in the absence of descending input, supporting that rhythm-generating modules were intrinsic to the spinal cord. Later studies in the lamprey and Xenopus tadpole revealed that ipsilateral excitatory drive combined with reciprocal inhibition

made up the core architecture of the vertebrate CPG [122, 61, 274].

Since Sherrington and Brown, our understanding of spinal circuits has rapidly advanced due to the development of genetic approaches for identifying and manipulating neurons and physiological tools for recording, activating, or suppressing them [228]. It is still believed that CPGs in the spinal cord, consisting of motor neurons and interneurons, are the modules responsible for transforming constant input into rhythmic output. However, we now understand that each unit of the CPG is composed of multiple neuronal subtypes [147].

Motor neurons, the best-characterized example of this subdivision, form molecularly- and anatomically-distinct columns, divisions and pools based on the body region and muscle they target [89]. This specificity is dictated by a single family of transcription factors, the Hox genes [344, 14, 90, 263]. Motor columns divide into motor pools which are further partitioned into alpha, beta, and gamma subtypes based on their fiber versus spindle innervation pattern [89]. The alpha subclass is further subdivided into fast-fatigue, fast-fatigue resistant or slow types based on the specific fiber type they innervate [127, 8]. The sequential and coordinated activation of these motor neuron types by a network of excitatory and inhibitory interneurons underlies coordinated movement.

Like motor neurons, excitatory and inhibitory interneurons in the spinal cord, can be similarly compartmentalized by their molecular, anatomical and functional properties [49, 182, 308]. They subdivide into at least 11 classes based on their developmental origin, gene expression and anatomical projection pattern: six dorsal classes (dI1-6), and five ventral classes (V0, V1, V2a, V2b, and V3; [142]. From a molecular perspective, recent single-cell sequencing work in the developing and adult mouse spinal cord has suggested that these eleven classes can be split into further numerous molecularly distinct cell types [159, 94, 41, 290]. Birth date, projection range, and motor/sensory function divides them even more [254]. Even a single class, such as V1, can contain up to 50 distinct subpopulations [126, 36, 129, 336]. From a physiological perspective, the response properties of interneurons also segregate them, exemplified by the recruitment of distinct excitatory V2a subtypes at slow or fast locomotor speeds [374, 10]. This demonstrates a broad organization of interneurons into cardinal classes and yet, a precise subdivision of the neurons within these classes based on their molecular and physiological characteristics.

### 1.1.2 Cross-species approach to study spinal circuit scaling

This large amount of spinal neuron subtype heterogeneity could provide the link between specialized vertebrate movements and their underlying spinal circuits. Many

recent studies have sought to test this possibility using species with diverse motor outputs and high levels of genetic access, such as the zebrafish and mouse [139, 148].

Here, we aim to lay the foundation for a complementary cross-species approach. Such an approach could differentiate cell types required for swimming (lamprey, fish, tadpoles) versus limb movement (frog, mice, humans, horses), or distinct movement capabilities between closely related species, such as rodents that hop versus run (kangaroo rat versus mouse) or mammals with varying gaits (mouse, horse, human). These approaches have the potential to pinpoint shared versus species-specific neural components of movement, taking us one step closer to determining how they correspond.

Such shared components include the precise coordination of muscle groups along the rostrocaudal, dorsoventral, and left–right body, and body-part axes; the variation of movement in a speed-dependent manner; and the ability of increasing drive to recruit additional motor units sequentially [166, 152, 151, 79, 232, 130]. This graded recruitment enables smooth transitions from slower or weaker to faster or stronger movements. In addition, reflex coordination has a modular organization, exemplified by studies in the frog, in which locomotion can be fractionated into motor primitives for each reflex [249, 161]. This principle is likely to extend across limbed species [211]. Finally, for an organism to survive in its environment, it must also integrate sensory information and vary the type, amplitude and speed of its movement accordingly [366].

Many components of movement however are species-specific, with one of the best examples being the speed-dependent expression of gaits. In many tetrapods, faster locomotion is achieved by gait transitions: walking at slow speeds, trotting at intermediate speeds and galloping at high speeds [35, 270, 128]. Horses exemplify this: the phase relationship of their limbs relative to each other varies between each of their speed-dependent gaits. A species-specific molecular mechanism has even been identified for this phase relationship with a mutation in the Dmrt3 gene resulting in the misspecification of a dorsal interneuron population and the appearance of either unnatural or additional gaits [11]. Additionally, in limbed vertebrates, spinal cord composition varies across the rostro-caudal axis [91, 126, 336, 89]. In this review, we largely focus on limb levels when discussing spinal cord architecture in limbed species.

Mechanistically, these shared and specific features between vertebrate species raise several fundamental questions that this review aims to explore. Is cell type heterogeneity in the spinal cord a correlate of movement diversity? At what level – molecular, anatomical and/or physiological – do cell types converge or diverge across species, and to what extent do these properties correspond? How do conserved features of movement, such as left–right coordination, map onto spinal cord cell types? And how do these maps vary for divergent features, such as gaits? Moreover, given the

variation in sensory and cognitive inputs between species, do spinal circuits similarly vary and if yes, for which cell types and on what level?

There has never been a better time to make such cross-species comparisons. Single-cell sequencing has enabled detailed molecular comparisons of neuronal classes in the spinal cord within and across species [159, 297, 94, 312, 290]. It is now possible to record hundreds of neurons in an actively-moving animal, empowering us to validate and extend findings which previously could only be made in an isolated spinal cord [224, 207]. We can also now take advantage of the vast anatomical and physiological knowledge of spinal neuron function in simpler organisms, in which they have been more comprehensively studied [148], to make novel predictions about their role in more complex ones.

### 1.1.3 Cardinal classes across vertebrates

In this review, we provide arguments to support the hypothesis that, as you move from simple swimming to limb-based movement across vertebrate evolution, spinal interneurons are compartmentalized into distinct molecular, anatomical and functional subclasses. Although these changes in spinal circuitry are accompanied by parallel changes in higher brain centers [252, 138, 185], these topics will not be discussed in this review. Here, we focus on interneurons in the spinal cord, structuring our discussion using the cardinal class organization of mammals, which captures both molecular and functional properties of each neuron class [94]. We start with the ventral excitatory classes: ipsilaterally-projecting V2a neurons and bilaterally-projecting V3 neurons. We then describe the mixed excitatory and inhibitory commissural class of V0 neurons and the inhibitory ipsilateral V1 and V2b neurons. Finally, we end with the dorsal inhibitory dI6 and mixed dI1-5 neurons, of which the least amount is known. In each section, we summarize our current knowledge of the conservation and divergence of cell type architecture across vertebrate species, focusing on zebrafish and mouse and, when possible, providing examples from lesser-studied species such as turtle and chicken.

## 1.2 V2a excitatory neurons

Ipsilaterally-projecting V2a excitatory neurons arise from the p2 progenitor domain and are defined by the transcription factor VSX2 in zebrafish and mice [21, 340]. During development, p2 progenitors differentiate into at least two subpopulations: V2a excitatory neurons, discussed here, and V2b inhibitory neurons, marked by GATA2/3 expression and discussed below [186, 245, 227, 258]. In zebrafish, where interneurons

are often named by their projection patterns, these neurons correspond to the circumferential descending (CiD) cells [156, 191]. Glutamate is a key neurotransmitter employed by V2a excitatory neurons in all species, with expression of the vesicular glutamate transporter 2 and blockage of V2a-derived motor neuron EPSPs by glutamate receptor antagonists, shared properties of zebrafish and mice [191, 80, 10].

In the lamprey, ipsilateral excitatory neurons provide the drive for the locomotor network [85, 61, 58, 115]. Although it is unknown whether they express Vsx2, their connectivity pattern and functional role as the drivers of movement suggest they may represent a primitive V2a population. In the lamprey, tadpole, zebrafish and mouse, this group of neurons receives descending and peripheral sensory input, and excites other V2a interneurons, commissural interneurons and motor neurons [85, 61, 60, 253, 257, 191, 192, 214, 220, 80, 326, 99, 10, 162, 217, 235]. Recurrent connections between V2a neurons generate consistency in motor output [61, 60, 69, 162, 235] and connections between V2a and commissural neurons implicate the V2a population in the coordination of the left and right side [61, 80, 99, 235]. Recent studies detailing how V2a neurons drive tail and limb movement patterns in zebrafish and mouse, respectively, provide a framework to understand how molecular, anatomical and functional subtypes correspond and scale with movement complexity.

### 1.2.1 Zebrafish V2a neurons

In zebrafish, V2a excitatory neurons are both necessary and sufficient to induce a normal swim pattern. Supporting this, action potentials in V2a neurons usually occur before those in motor neurons [10], and optogenetic activation of this class generates swimming [106], implicating them as drivers of the swim circuit. V2a neurons are also present in the hindbrain, where optogenetic activation drives, while inactivation impairs or stops, swimming [192]. Acute and selective ablation of V2a neurons has three potent effects on swimming activity: an increase in the threshold for its initiation, a decrease in locomotor-related burst frequency, and a change in the rostrocaudal propagation of activity [107]. Similar changes were seen when swimming was induced by electrical stimulation or NMDA application, suggesting they are due to perturbations in the excitability of the swim circuit [107]. This experimental evidence provides strong support that V2a neurons are crucial drivers and determinants of locomotion in zebrafish.

It was also observed in zebrafish that specific V2a subpopulations are recruited in a speed-dependent manner. As the fish's swim speed increased, ventrally located V2a neurons were recruited before dorsally located ones [191, 233, 231, 17]. Selective ablation of dorsal V2a neurons decreased peak, without altering sustained, swim

frequency; whereas optogenetic activation of the entire V2a population recruited ventral, but not dorsal, subpopulations [107]. This suggested a speed-dependence of V2a excitatory subpopulations in tail locomotion, dictated by their dorsoventral location and recruitment threshold.

The physiological role of V2a neurons in the activation and speed-dependent modulation of motor output was shown to map onto three spatially distinct microcircuits: one for driving slow, one for intermediate and one for fast motor neurons [191, 10]. These circuit modules are spatially segregated along the dorsoventral axis of the spinal cord and arrayed such that the slow is recruited before the fast one [329]. Each module preferentially targets either slow or fast motor neurons and has different anatomical and functional properties [329]. The slow V2a neurons preferentially target slow motor neurons, have unidirectional caudally projecting axons, and exhibit bursting activity; they display significant short-term potentiation, which decreases the number necessary to activate motor neurons [329, 10]. The fast V2a neurons preferentially target fast motor neurons, show no bursting activity, and project in both the caudal and rostral directions; they lack short-term potentiation and produce a weaker excitatory drive, requiring a larger number of them to be activated to generate a motor neuron response [329, 10]. The excitatory drive to V2a neurons and motor neurons of the same speed class is further organized in a continuum, such that at faster locomotor speeds, the drive to the intermediate and fast class is increased [10]. This circuit organization allows for a smooth transition from slow to fast swimming with increased drive.

The functional role of V2a speed-dependent microcircuit segregation is also evident during zebrafish development. Paralleling the development of swim behavior—where strong contractions are needed for early escape swimming while slow, sustained swimming emerges later—the dorsal, fast V2a class forms earlier than the ventral, slow class [191]. In early larvae, V2a neurons fall into two morphological classes: those with ascending and descending axons, and those with only descending axons [236]. Within the descending population, more dorsal V2a neurons project longer axons and make denser synapses onto proximal motor neurons than ventral V2a neurons [236], an organization suited to recruiting the entire motor pool during fast escape. By contrast, the ventral V2a population innervates smaller motor-neuron territories and fires in-phase with motor neurons, supporting finer control required for late-stage slow swimming [237].

An alternative V2a subdivision has been proposed in which molecular, morphological, and electrophysiological features—rather than anatomy and speed—segregate these neurons into two groups that control either timing or amplitude [235]. "Timing" V2a neurons preferentially connect to other V2a excitatory neurons and V0d inhibitory neurons, receive primary-afferent input, and set locomotor frequency, while

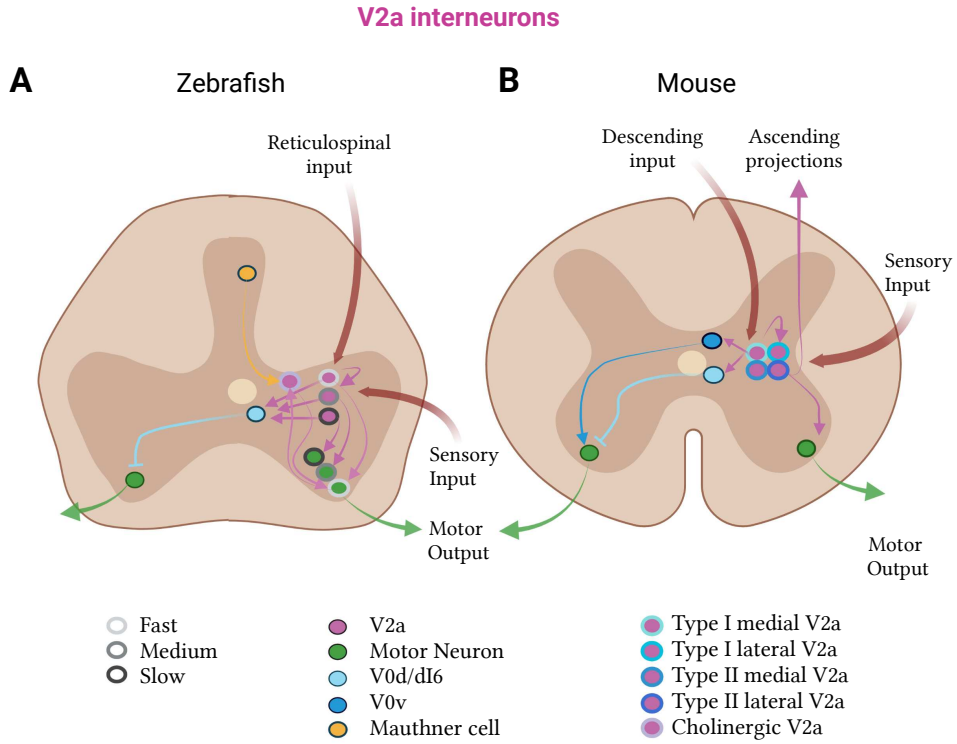


**Figure 2**

Figure 1.2: Excitatory V2a subtypes in zebrafish and mice. (A) In zebrafish, the V2a class (pink) is divided into fast (light gray, outline), medium (medium gray, outline) and slow (dark gray, outline) subtypes. The fast subtypes are more dorsal than the slow subtypes. Each subtype receives reticulospinal and sensory input, and projects to the corresponding fast, medium or slow class of motor (green) and V0d (light blue) neurons. Zebrafish also have a cholinergic subclass of V2a neurons, which receive input from Mauthner cells and have bidirectional connections to fast motor neurons. (B) In mice, the V2a population receives descending and sensory input, projects to inhibitory V0d and motor neurons, and has recurrent connections. Mouse V2a neurons subdivide into type I and type II, which further divide into medial and lateral subtypes (shades of blue, outlines). Type I V2a neurons connect to V0 neurons (V0d in light blue and V0v in dark blue); type II V2a neurons have ascending connections.

"amplitude" V2a neurons predominantly target motor neurons and shape movement strength [235].

Recent work also identifies a cholinergic V2a subpopulation integral to the escape-swim circuit. These neurons form bidirectional electrical connections with ipsilateral motor neurons, and their ablation disrupts escape responses by preventing amplification and distribution of the command [150]. How this cholinergic group relates to previously defined V2a subtypes remains to be determined.

Taken together, zebrafish possess anatomically and physiologically distinct sub-

classes of ipsilateral V2a excitatory neurons. This contrasts with the lamprey and larval *Xenopus* tadpole, where evidence for sharply defined subclasses is limited [57, 327, 264]. These subdivisions likely enable smooth muscle activation and set both the strength and frequency of the diverse swimming patterns seen during zebrafish development, with future work needed to clarify the molecular bases of these distinctions.

### 1.2.2 Mouse V2a neurons

As in zebrafish, V2a excitatory neurons in mouse provide premotor excitation and help regulate speed-dependent microcircuits. This conserved role in driving locomotion and stabilizing its tempo is supported by in vitro and in vivo experiments showing that perturbing V2a neurons alters locomotor rhythm and coordination [80, 376, 375, 370]. In particular, genetic and physiological manipulations of the V2a pool affect burst amplitude, cycle period, and left–right coupling, consistent with anatomical evidence that V2a neurons project to commissural populations involved in alternation [80].

Multiple V2a subtypes have been identified in mice. Electrophysiological recordings from identified V2a neurons revealed diverse response profiles—including rhythmically firing, subthreshold-rhythmic, tonic-firing, and quiescent cells—with only a subset increasing recruitment as locomotor frequency rises [376]. Rough anatomical position was a poor predictor of firing phenotype, underscoring functional heterogeneity within the class [376].

Converging lines of work point to organizational principles beyond simple dorsoventral position. Imaging and systems studies highlight local vs. longer-range V2a contributions to pattern generation and speed control, as well as heterogeneity in how V2a neurons interface with downstream targets [305, 254, 308]. Complementary single-cell transcriptomic atlases of the mouse spinal cord further resolve V2a neurons into molecularly distinct subsets (including medial/lateral tendencies and rostrocaudal specializations), suggesting circuit roles that vary across axial vs. limb levels [367, 94, 41, 290]. These data align with functional results showing that V2a neurons contribute to limb patterning and flexor–extensor coordination during locomotion [370].

Altogether, V2a neurons in mouse exhibit subtype diversity consistent with graded muscle recruitment as speed increases and with tetrapod-specific demands for limb control. Compared with fish, mouse V2a diversity appears more tightly linked to body level and task demands, fitting the broader motor repertoire available along the neuraxis [375, 254, 308].

### 1.2.3 Cross-species perspective on V2a

Common and divergent properties of V2a neurons have emerged across species. In all vertebrates, the V2a population provides key premotor excitation for locomotion [57, 191, 80, 10, 370]. Notably, in mice they do not appear to be strictly necessary for rhythm generation itself [375]. In higher vertebrates such as zebrafish and mice, functional, anatomical, and molecular V2a subdivisions confer added roles in pattern regulation. A prominent feature is differential recruitment with speed, which suggests that locomotor drive can be selectively routed to particular motor pools by specific V2a subpopulations [10].

Functional distinctions also track with connectivity. In lamprey and Xenopus tadpoles, V2a-like cells excite motoneurons, other V2a-like neurons, and commissural inhibitory interneurons [57, 257, 215]. These motifs persist in zebrafish and mouse, alongside supraspinal projections and long-range interactions [191, 192, 80, 100, 376, 10, 236, 162, 235]. In mice, V2a neurons are functionally heterogeneous [305]. The split between cells that do or do not receive locomotor-related synaptic drive [375] may mirror zebrafish groups that were proposed to separately control timing versus amplitude of locomotion [235].

In summary, V2a excitatory neurons are vital drivers of movement. In zebrafish, this role is specialized into strong and weak subgroups differentially active during escape versus slow swimming [107, 10, 237, 236, 329]. In mice, only about half of V2a neurons receive locomotor-related drive, with the function of the remainder still unclear [375]. Cervical specializations likely support precise forelimb control via ascending/non-local V2a pathways [162]. V2a neurons also contribute in a speed-specific manner to maintaining left–right alternation at high (but not low) frequencies, likely through excitation of commissural V0 populations [80, 375]. Given these roles, the mouse V2a population is probably more molecularly diverse than in zebrafish, a hypothesis increasingly testable with single-cell atlases [367, 94, 41, 290].

## 1.3 V3 excitatory neurons

V3 excitatory neurons, best studied in mice, are glutamatergic *Sim1*-expressing neurons that derive from the p3 progenitor domain during development [48, 334, 372, 360]. Their anatomical projection pattern is a key defining feature. Like V2a neurons, they project ipsilaterally but, unlike them, the majority of V3 neurons also have contralateral projections that extend caudally [372]. Such bilaterally projecting neurons do not appear to be part of the pattern-generating network in the lamprey spinal cord [54, 53, 59, 70, 204]. They are, however, present in zebrafish, where they occupy a single ventromedial domain across the length of the spinal cord and match

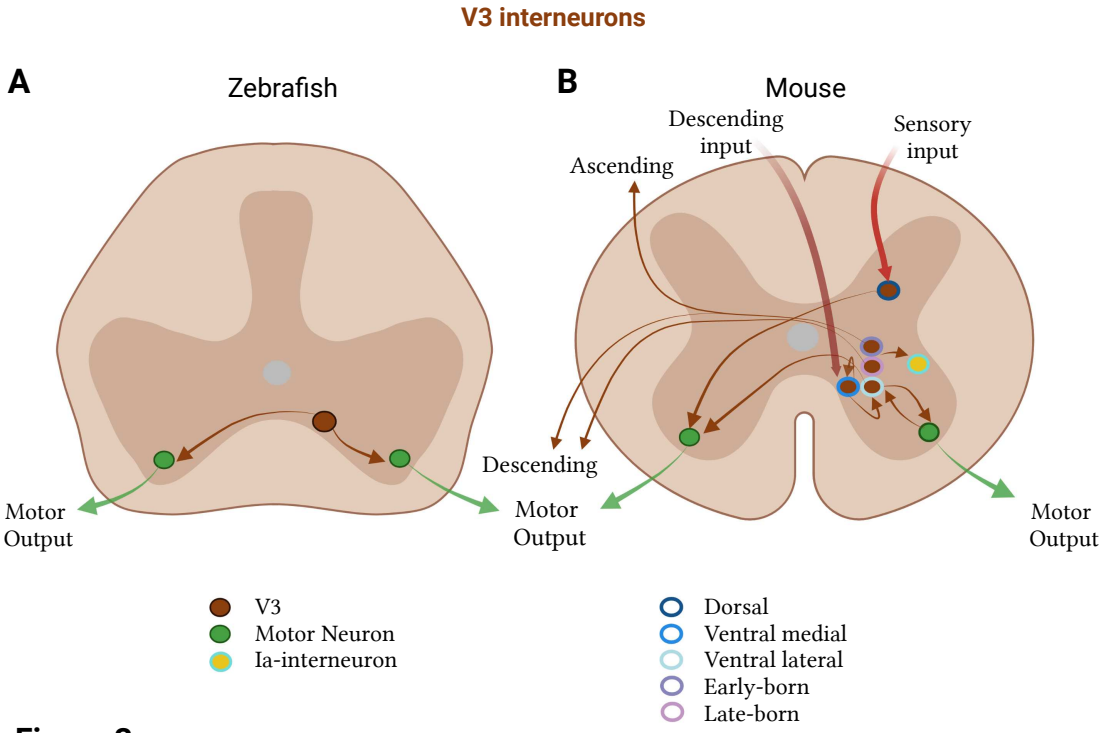


**Figure 3**

Figure 1.3: Excitatory V3 subtypes in zebrafish and mice. (A) In zebrafish, V3 neurons (brown) project to motor neurons (green) on both sides of the spinal cord and (B) in mice are divided into a dorsal (dark blue, outline) and ventral class, the latter of which is further subdivided into a medial (medium blue, outline) and lateral (light blue, outline) group. The ventral medial group receives descending commands, forms recurrent connections with itself, and projects to the ventral lateral group. The lateral group projects to motor neurons on the ipsi and contralateral side. Motor neurons project back to V3 neurons in mice. V3 neurons also subdivide by their birth time into an early-(dark purple, outline) and a late-born (light purple, outline) subpopulation, which form either both an descending and ascending, or only a descending, projection, respectively. V3 neurons in mice also project to Ia- interneurons.

the projection properties of the anatomically defined ventral medial (VeMe) neurons [156, 170, 169, 353]. In contrast, in the mouse, V3 neurons migrate after differentiation to form spatially and physiologically distinct subpopulations [44]. This raises the question of whether V3 subdivision is important for more complex locomotion in limbed, as opposed to finned, vertebrates.

### 1.3.1 Zebrafish V3 neurons

Zebrafish V3 excitatory neurons comprise a spatially and functionally homogenous population of neurons active during fictive swimming (Fig.1.3A; [42, 353]). They are critical for the recruitment of motor neurons, as their activation increases swim

strength and ablation reduces motor neuron activity [42, 353]. They regulate the burst amplitude independent of the burst frequency, thus providing drive but not influencing the speed of swimming or underlying locomotor rhythm [42, 353]. One proposed function of the zebrafish V3 population is the relay of excitatory drive to coordinate motor units, according to the desired amplitude of locomotor bursts. The bilateral and descending projection pattern of the V3 class in zebrafish is consistent with this hypothesis, as they contact multiple motor units. Moreover, the timing of V3 spikes relative to those of motor neurons suggests a role in providing excitatory drive during locomotion [42, 353].

### 1.3.2 Mouse V3 neurons

In mice, V3 neurons play a more specific role in the symmetry of motor control by ensuring the balance of motor output between the left and right sides of the spinal cord [373]. Suppression of V3 synaptic transmission produces loss of coordination and stability during locomotion—raising the coefficient of variation, but not the mean, of burst duration and period—while optogenetic/chemogenetic activation lengthens contralateral motor bursts and slows stepping [373, 88]. Together, these data suggest that the function of V3 excitatory neurons diverged across evolution to meet the demands of tetrapod locomotion, where precise left–right balance is critical.

V3 neurons in mice are also highly spatially and functionally heterogeneous, unlike the more uniform population described in zebrafish. They are found across multiple laminae and along both dorsoventral and rostrocaudal axes in the postnatal spinal cord [373, 44, 40]. Electrophysiology and morphology further divide them into dorsal and ventral subgroups with distinct properties: dorsal V3s, active primarily during running, display complex branching and low gain, whereas ventral V3s, active during both swimming and running, have simpler morphology, higher gain, and tonic firing better suited to reliably relay motor commands [44].

Within the ventral population, a lateral and a medial V3 subgroup can be distinguished [74]. Lateral V3 neurons excite both ipsi- and contralateral motor pools and make bidirectional (electrical/glutamatergic) connections with ipsilateral motoneurons; their contralateral targets are likely to include motoneurons, consistent with the prevalence of V3-derived synapses on contralateral motor neurons [74, 373]. Medial V3 neurons occupy a distinct ventromedial layer that receives descending motor commands [230] and form synapses with both medial and lateral ventral V3s, suggesting a role in integrating reticulospinal drive that is then distributed to appropriate motor pools via the lateral subgroup [74].

Recent work also subdivides V3 neurons by birth date and projection pattern:

early-born V3s possess both ascending and descending commissural projections, whereas late-born V3s exhibit descending and local commissural projections; early-born cells span multiple laminae while late-born cells are confined ventrally [95]. Notably, some ascending lumbar V3 axons terminate contralaterally in cervical segments and appear crucial for diagonal limb synchronization; modeling shows that selectively removing ascending V3s abolishes trot while preserving gallop and bound, highlighting gait-specific roles for V3 subpopulations [368].

### 1.3.3 Cross-species perspective on V3

V3 excitatory neurons are similar in fish and mice but exhibit greater complexity in the latter, arising from spatially distinct subpopulations with specialized functions [373, 44, 74, 95, 368]. The split into dorsal and ventral subgroups is thought to support integration of sensory information in limbed vertebrates [44]. Further division of the ventral pool into medial/lateral and early/late-born subgroups may enable finer control of limb muscles by channeling drive to specific ipsi- and contralateral motor pools [74, 95, 368].

In mice, stimulating motor neurons can initiate locomotor-like activity and shape the rhythm via a glutamatergic pathway [240, 112, 74]. The bidirectional connections between ventrolateral V3 neurons and motoneurons may therefore allow motoneurons to contribute to rhythm generation and help maintain left–right balance [74, 373]. In addition, V3 neurons synapse onto inhibitory Ia interneurons, potentially facilitating burst termination and adding another layer of control over the locomotor cycle [373]. These features align with proposed roles of V3 subpopulations in enabling gait transitions in mammals [292, 87, 368].

Across species, V3 neurons—like V2a neurons—provide excitatory drive. In zebrafish, they appear crucial for recruiting motor units and regulating burst amplitude [42, 353]. In mice, they help balance activity across the two sides of the spinal cord and integrate, relay, and direct sensory and descending motor commands [373, 74]. Molecular studies are beginning to illuminate how gene-expression programs map onto these divergent functions; notably, V3 neurons in both mice and zebrafish are molecularly heterogeneous [247], raising the possibility that molecular subclasses underlie the anatomical and functional diversity described above.

## 1.4 V0 excitatory and inhibitory commissural neurons

Commissural interneurons are a common feature of all vertebrate spinal cords. In the lamprey, both excitatory glutamatergic and inhibitory glycinergic commissural

interneurons have been identified [293]. They can be divided into three groups based on their reticulospinal inputs [54, 53, 316, 59, 238, 239, 38]. Excitatory commissural neurons in lamprey seem to be selectively involved in the movement of the dorsal fin [316, 238, 239]. They are active in phase with both ipsi- and contralateral fin motor neurons, and drive their simultaneous activation during straight swimming. Inhibitory commissural neurons have been shown by modeling and experimental studies to be necessary for the generation of bilateral undulatory swimming, but not unilateral rhythm generation [77, 7, 154, 13, 68, 70, 178, 204]. They receive input from excitatory ipsilateral neurons, and, on the contralateral side, drive motor neurons, lateral inhibitory neurons, and other inhibitory commissural neurons [54, 53]. They decrease burst frequency and coordinate and stabilize the activity on both sides of the spinal cord [53, 70, 204].

In other vertebrates such as zebrafish and mice, ventral commissural interneurons, termed V0 neurons, derive from the p0 progenitor domain and are characterized by expression of the transcription factor DBX1, which is required for their development and commissural connectivity [244, 265, 189]. In both zebrafish and mice, they extend axons rostrally for two to four spinal cord segments, and either mono- or poly-synaptically synapse onto motor neurons, suggesting that they are important for diagonal coordination [244, 265, 65, 209, 267, 335, 279]. In zebrafish, there is evidence that the V0d population forms reciprocal connections with itself, and projects to ipsilateral inhibitory neurons [325, 299]. Additionally, both V0d and V0v populations likely connect to V2a neurons and motor neurons [233, 145, 335, 299, 284]. In mice and rats, the V0 class is also thought to form reciprocal connections with other commissural neurons, and project to ipsilateral inhibitory neurons on the opposite side of the spinal cord [196, 37, 65, 267]. Compared to the monosynaptic connections in zebrafish, the projections to motor neurons in mice tend to be disynaptic [196, 65, 267, 233, 335].

In zebrafish and mice, this V0 class is composed of both excitatory V0v and inhibitory V0d subtypes (Fig.1.4). Excitatory V0v interneurons derive from the ventral DBX1 progenitor domain and transiently express the homeodomain protein EVX1. Inhibitory V0d neurons derive from the dorsal DBX1 domain and lack EVX1 expression, but unlike V0v, express PAX7 [244, 265]. The V0d neurons are largely GABAergic or glycinergic, whereas the V0v neurons are glutamatergic [244, 338]. As in the mouse and in contrast to the lamprey, extensive studies in zebrafish support that the V0 population is highly heterogeneous in its connectivity and function, with both the anatomically defined bifurcating multipolar commissural descending (MCoD) and unipolar commissural descending (UCoD) neurons best corresponding to the V0v class and the glycinergic commissural bifurcating longitudinal (CoBL) neurons, to the V0d class [298].

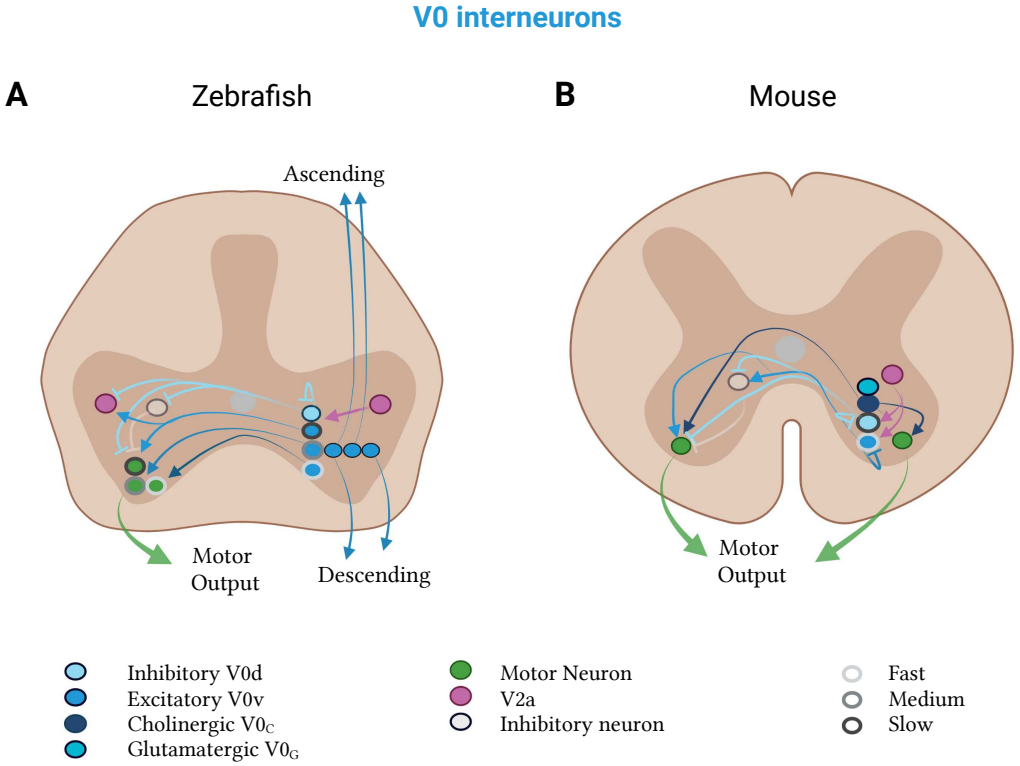


**Figure 4**

Figure 1.4: Mixed V0 subtypes in zebrafish and mice. V0d neurons (light blue) inhibit, and V0v neurons (medium blue) excite, contralateral motor neurons (green). (A) Zebrafish V0d neurons receive input from V2a neurons (pink) and project to other V0d, contralateral inhibitory (gray) and ipsilateral excitatory (pink) neurons. The V0v neurons project to V2a neurons and divide into a rhythmic and a non-rhythmic group. The rhythmic group is further split into fast (light gray, outline), medium (medium gray, outline) and slow (dark gray, outline) subtypes, which project to motor neurons of the respective speed class. Excitatory V0v neurons also segregate by projection pattern into ascending, descending and bifurcating subpopulations. (B) Mouse V0d and V0v neurons control slow and fast speeds, respectively. Both classes receive input from V2a neurons. The V0v class additionally projects to contralateral neurons (gray), which inhibit motor neurons. Mouse-specific V0c neurons (dark blue) are cholinergic and project to motor neurons on both sides of the spinal cord. Mouse-specific V0g neurons (turquoise) are glutamatergic and their projection pattern is as of yet unknown.

### 1.4.1 Zebrafish V0 neurons

Zebrafish V0v and V0d neurons coordinate, regulate, and drive a variety of locomotor features during development, highlighting their diverse contributions to higher-order vertebrate movement [232, 233, 231, 298, 119, 187, 311, 271]. As with other interneu-

ron classes in zebrafish, they have historically been defined by projection pattern, with the best-studied being the multipolar commissural descending (MCoD) V0v subpopulation that develops at a later stage of neurogenesis [231, 298]. In late-stage larvae, V0v neurons are important for slow swimming [271, 232, 233, 231, 119]. They keep the head stable during this form of locomotion by coordinating the activities of diagonal trunk muscles, enabling the characteristic S-shaped bends [187]. During fast swimming, V0v neurons are not active and the head is no longer stabilized at higher frequencies [233, 231, 311]. Additionally, ablation of the V0v population decreases spontaneous swimming, suggesting that these neurons also contribute to the general excitability of the motor circuit [233, 187]. As zebrafish mature into adults, the physiological properties of this class change; adult V0v neurons display speed-dependent recruitment during swimming, with many recruited at fast speeds [39].

The anatomical and electrophysiological properties of V0v neurons are consistent with a role in coordinating diagonal activity during swimming (Fig. 4A). Their axons cross the midline and descend, making direct monosynaptic connections with motor neurons contralateral and caudal to the presynaptic V0v neurons [156, 233]. They fire in a highly phasic manner, with spike timing slightly preceding nearby motor neurons [187]. Modeling suggests that they may also connect to excitatory V2a neurons at later developmental stages [284].

Reflecting these diverse physiological roles, zebrafish V0v neurons are highly heterogeneous. They can be divided into three subclasses based on temporal order of development and axonal projection patterns [298]. The first to develop is the V0v subclass with ascending projections, followed by those with bifurcating projections, and finally those with descending projections—where ascending/descending correspond to unipolar UCoD and bifurcating to multipolar MCoD anatomical subclasses [298]. Each subclass includes rhythmic and non-rhythmic types; the rhythmic group further splits into subsets recruited at slow, intermediate, or fast speeds [298], explaining the speed-dependent recruitment seen in adults.

Less is known about inhibitory V0d neurons. Anatomically, the morphology of the glycinergic commissural bifurcating longitudinal (CoBL) population matches that of V0d [298]. Like inhibitory commissural interneurons in lamprey, they likely provide mid-cycle inhibition onto motor neurons and other neurons, important for left–right motor alternation [299]. Both V0d and Dmrt3a-expressing dI6 neurons have monosynaptic inhibitory connections to neuronal populations active during fictive swimming, including contralateral motor neurons (Fig. 4A) [299]. The V0d population tends to fire during faster, stronger movements, whereas the dI6 subpopulation fires during normal fictive swimming [299]. Both populations are active in phase with nearby motor neurons, suggesting that they inhibit contralateral motor neurons when the ipsilateral side is active—a conserved feature of undulatory swimming.

### 1.4.2 Mouse V0 neurons

Like zebrafish, mice also have an excitatory V0v and an inhibitory V0d population (Fig.1.4B). Excitatory V0v neurons in mice coordinate diagonal limb muscles during walking, analogous to their role in diagonal muscle coordination during zebrafish swimming. Ablation of the V0v, or V2a neurons that innervate them, at the cervical level mainly impacts left–right hindlimb, but not forelimb or interlimb, coordination [288]. This suggests evolutionary conservation of this long-range, cross-body diagonal function of V0 neurons across species. In addition, mice which lack V0 neurons exhibit increased co-bursting between the left and right sides of both flexor and extensor muscles, which is expressed as quadrupedal hopping at all frequencies of locomotion [209, 338]. This further demonstrates that V0 neurons also contribute to left–right alternation in mice.

The conservation of V0 subtype function between species is also demonstrated by the similar role of zebrafish and mouse V0 neurons in speed-dependent motor control. Selective ablation of just the inhibitory V0d neurons leads to a lack of left–right alternation at slow locomotor speeds, mixed coordination at medium, and normal alternation at high speeds [338]. Conversely, ablation of only V0v neurons has the opposite effect: normal alternation at slow speeds and hopping at intermediate and high speeds [338]. This high-speed hopping is also observed in V2a mutants [80]. V2a excitatory neurons in mice project to V0v neurons, providing a circuit-level mechanism for this phenotype [80]. Computational modeling suggests that V0v neurons may also project to contralateral inhibitory interneurons that contact motor neurons [314, 87]. Thus, as in zebrafish in which V0 subpopulations are segregated by the speed of locomotion they influence, in mice V0d control slow, and V0v high, speed locomotion.

Unlike zebrafish, there is emerging evidence in mice of two other excitatory classes of V0 neurons: the cholinergic V0C and glutamatergic V0G neurons [365]. Both populations are marked by the transcription factor PITX2. V0C neurons, the best studied of these two populations, provide cholinergic C-bouton input to motor neurons and V1-derived Ia interneurons on either one or both sides of the spinal cord (Fig.1.4B) [365, 317, 332]. Genetic inactivation of V0C output results in behavioral deficits in task-dependent motor performance [365]. Their firing activity is tightly phase-locked to that of motor neurons, and when they are inactivated, motor neuron firing and muscle activation are impaired [365]. Conversely, their activation increases the excitability of motor neurons by reducing the afterhyperpolarization following each action potential [241]. This suggests that V0C excitatory neurons activate motor neurons to ensure firing at rates appropriate for the desired locomotor task, a property of the V0 population thus far described only in mice.

### 1.4.3 Cross-species perspective on V0

Across vertebrates, the V0 class is important for long-range coordination of rostrocaudal and left–right body parts [209, 338]. One might therefore predict that these are the neurons that vary the most between tetrapods that differ in their default mode of locomotion along these axes, such as frogs and mice. Within the V0 class, however, there is remarkable conservation of subtypes between species, with excitatory/inhibitory, diagonal-coordinating, and speed-dependent classes highly conserved [209, 338, 298]. A notable exception is the presence in mice—but not in zebrafish—of the excitatory V0C and V0G subclasses [365]. Additionally, the speed-dependent subdivision described for zebrafish V0v neurons has not yet been systematically investigated in mice [298]. Thus, V0 specialization may not increase quantitatively over vertebrate evolution but may change qualitatively according to each species' specific left–right coordination requirements. Future work should test whether these functional differences are mirrored at molecular and anatomical levels, and to what extent they are conserved in simpler vertebrate systems.

## 1.5 V1 and V2b inhibitory ipsilateral neurons

In addition to commissural excitation and inhibition, ipsilateral inhibition is a key component of more complex swim and limb spinal circuits. Two types of ipsilaterally projecting inhibitory neurons exist in the lamprey—ipsilateral inhibitory neurons (IINs) and lateral inhibitory neurons (LINs) [287, 57, 56]. IINs inhibit both motor and commissural neurons, whereas LINs generally inhibit only commissural neurons [54, 56, 285]. LINs additionally receive inputs from ipsilateral excitatory, contralateral, and dorsal neurons [287, 54, 57]. Early computational models of the lamprey locomotor network incorporated ipsilateral inhibition [351]; however, because ipsilateral inhibition is not required to generate a hemicord burst pattern [68], these neurons are often excluded and may instead chiefly inhibit dorsal fin motor neurons [203, 239, 204]. This supports the hypothesis that the dorsal fin circuit was a precursor to lateral fin and, ultimately, flexor–extensor limb circuitry. In larval *Xenopus*, by contrast, ipsilateral inhibitory neurons (aINs) appear to be a core component of the pattern-generating network for escape swimming [216, 213].

In zebrafish and mice, ipsilateral inhibitory interneurons comprise two major molecular classes, V1 and V2b (Fig. 1.5). V1 neurons arise from the p1 progenitor domain, express Engrailed-1 (EN1), use GABA and/or glycine, and target ipsilateral motor neurons as well as other ipsilateral inhibitory interneurons [302, 168, 295, 9, 227, 22, 30, 317, 32, 371, 315, 67, 190, 309]. In zebrafish, the homologs of mammalian V1 neurons are the circumferential ascending (CiA) cells, which likewise express *en1*,

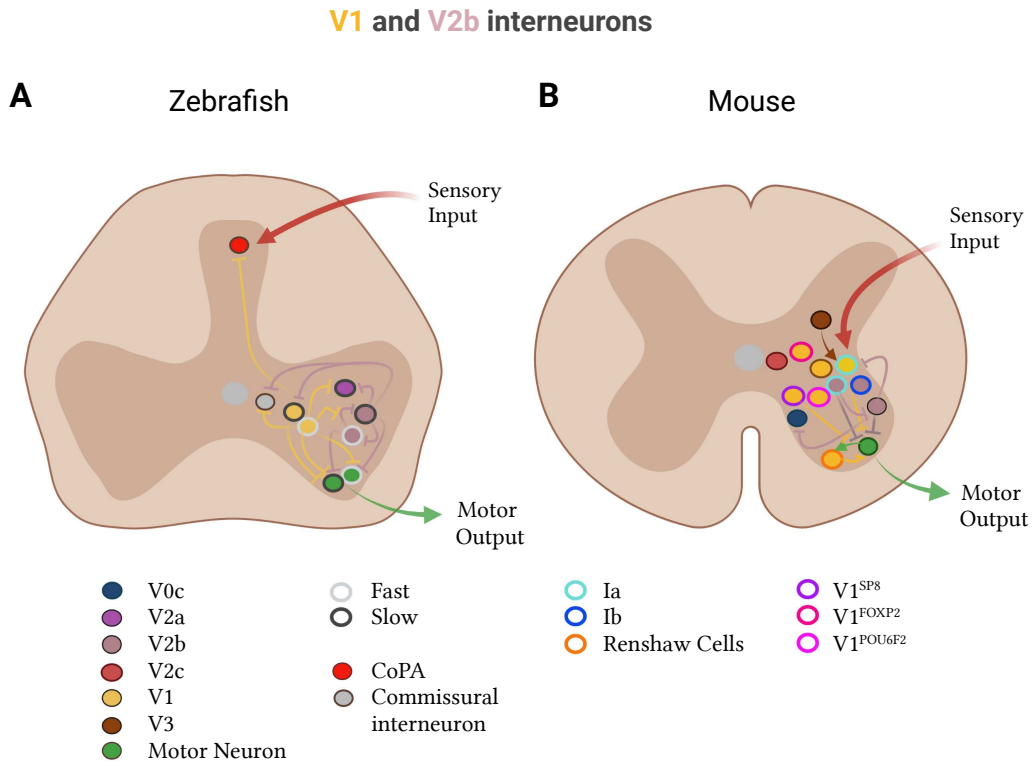


**Figure 5**

Figure 1.5: Inhibitory V1 and V2b subtypes in zebrafish and mice. (A) In zebrafish, V1 (yellow) and V2b (brown) divide into fast (light gray, outline) and slow (dark gray, outline) subtypes. V1 neurons: The fast V1 subgroup (yellow, light gray outline) inhibits both slow (green, dark gray outline) and fast (green, light gray outline) motor neurons in addition to slow-type V2a neurons (pink, dark gray outline). The slow V1 subgroup (yellow, dark gray outline) inhibits slow motor neurons (green, dark gray outline). V1 neurons also project to dorsal CoPA neurons (red) which receive sensory input, V2a neurons (pink), V2b neurons (brown), and commissural neurons (gray). V2b neurons: Slow V2b neurons (brown, dark gray outline) inhibit fast motor and other V2b neurons. Fast V2b neurons (brown, light gray outline) inhibit slow motor and other V2b neurons. V2b neurons in zebrafish also project to V2a, V1 and commissural neurons. (B) In mice, V1 neurons and subdivide into Ia-interneurons (light blue, outline), which receive sensory input and inhibit motor output; Renshaw cells (orange outline) which form recurrent connections with motor neurons; and four clades: Sp8 (purple outline), FoxP2 (pink-red outline) and Pou6f2 (pink outline). V1 neurons also receive input from V3 neurons (brown). V2b neurons include Ia- and Ib- (dark blue, outline) interneurons. V2b-derived Ia-interneurons inhibit motor and other V2b neurons. V2b neurons also inhibit V0c neurons. An additional V2c class is present in mice (red-pink) with an unknown projection pattern.

project ipsilaterally, and contact motor neurons, other ipsilateral inhibitory and excitatory neurons, and commissural neurons [168, 213, 190, 309].

The V2b ipsilateral inhibitory population derives from the LHX3-expressing p2

progenitor domain in mice and is defined by expression of the transcription factors GATA2/3 [227, 258, 67]. This GATA3-expressing population appears well conserved across vertebrates and beyond—present in the spinal cord of chicks and even the ventral nerve cord of worms [186, 349]. In zebrafish, V2b inhibitory neurons are thought to correspond to the anatomically defined ventral longitudinal (VeLD) neurons, which similarly express *gata3* and project ipsilaterally, but derive from the pMN domain [29, 156, 258, 22, 310]. In both mice and fish, V2b neurotransmitter profiles change during development: a large proportion initially uses GABA [22] and later transitions to glycine [227, 371, 67]. In mice, the p2 progenitor domain also gives rise to a third class, the V2c neurons, which express SOX1 and GATA3 transiently in early development [256]. Diversification of the V1 and V2b inhibitory populations nevertheless seems key for producing an expanded repertoire of movement patterns in mice compared to fish.

The V2b class largely projects axons caudally in both zebrafish and mice [227, 50, 67]. In zebrafish, V2b neurons contact motor neurons and multiple interneuron classes on the ipsilateral side—including V2a, V1, V2b, and commissural neurons [307]. In mice, there is evidence that they project to V0c neurons, V1 neurons, and motor neurons [371, 315]. Notably, the well-studied Ia- and Ib-inhibitory interneuron populations, which control basic flexion–extension and autoinhibitory reflex circuits respectively, derive from V1 and/or V2b lineages [23, 50]. Together, V1 and V2b neurons are necessary for flexor–extensor alternation in mice [371, 50].

### 1.5.1 Zebrafish and frog V1 and V2b neurons

The existence of the V1-homologous CiA and V2b-homologous VeLD neurons in zebrafish, which lack the same extent of flexor–extensor divisions as four-limbed vertebrates, suggests that flexor–extensor coordination was only a role that this population took on later in evolution or alternatively with pectoral and pelvic fin control [341, 348]. Ablation of V1 inhibitory neurons in both zebrafish and mice leads to reduced fictive locomotor speeds [140, 190]. This occurs via reduced inhibition and thus a longer intersegmental delay and locomotor cycle period [339, 347, 131]. In contrast, in vivo optogenetic suppression of V2b activity in zebrafish leads to an increase, and activation to a decrease, in tail beat frequency [67]. In the *Xenopus* tadpole, there is only one ipsilateral inhibitory neuron class, the ascending interneuron (aIN) population, which is thought to modulate the swim cycle by providing in-phase inhibition to motor and other rhythm-generating neurons [213, 276]. Thus, in-cycle inhibition of locomotion represents a conserved feature of V1 neurons in motor pattern generation across vertebrates.

Along with their role in regulating the motor pattern, the V1 inhibitory class is also believed to play an essential part in sensory integration in both *Xenopus* and zebrafish through reflex inhibition during movement via dorsal interneuron connectivity [213, 168, 197, 309]. This is reminiscent of the presynaptic inhibition of spinal sensory feedback necessary for smooth movement in mice, although in mice this function is likely carried out by dorsal interneurons [120].

Like motor and V2a excitatory neurons, V1 inhibitory neurons can be divided into slow and fast subtypes in zebrafish [190]. The mechanism by which V1s regulate motor output in this speed-dependent manner is through direct connections with motor and V2a excitatory neurons [190]. During fast swimming, strong in-phase inhibitory inputs from fast-type V1 neurons suppress the activity of slow-type V2a and motor neurons [190]. During slow swimming, slow-type V1 inhibitory neurons act on slow circuits by providing inhibition to regulate the cycle frequency. When swimming changes from slow to fast, fast-type V1 neurons are thought to shut down the slow circuit. In parallel, the fast subpopulation regulates the period of fast circuits by tuning their inhibition to the strength of excitation they receive [195, 190]. This regulation is important for deactivating slow muscles and slow-type motor neurons during fast swimming [345, 181, 64, 237, 195].

V2b inhibitory neurons in zebrafish can be divided into two subclasses by neurotransmitter and morphological properties: V2b-mixed and V2b-gly subpopulations [67]. Both express glycine, with the V2b-mixed also expressing GABA. The two subtypes are indistinguishable physiologically and are found along the rostrocaudal axis, with V2b-mixed located more ventrally and V2b-gly more dorsally. Both classes synapse directly onto motor neurons but target speed-specific circuits: V2b-mixed targets slow and V2b-gly targets fast motor neurons. The V2b-gly subclass innervates more of the dorsal spinal cord than V2b-mixed [67]. Additionally, rostral V2b neurons inhibit more caudal V2b neurons, leading to long-range circuit disinhibition. Locally, V2b-mixed and V2b-gly neurons make reciprocal connections with each other, which may stabilize the circuit at a desired speed [67].

### 1.5.2 Mouse V1 and V2b neurons

Similar to zebrafish, ipsilateral inhibitory neurons in mice are also required for regulating locomotor speed. Pax6-knockout mice, which lack V1 inhibitory neurons, display prolonged motor neuron activation that leads to slowed stepping—a phenotype replicated when V1 inhibitory neurons are acutely silenced or hyperpolarized [140, 111]. More recent studies have revealed additional heterogeneity in V1 function, with the type of manipulation producing different effects on the frequency of motor output

[111, 110]. This supports further subdivision of V1 interneurons by connectivity into functionally distinct subpopulations, a concept previously proposed in computational models [315].

Gain- and loss-of-function experiments in mice also show that V1 and V2b neurons contribute directly to the coordination of limb movement. Mice lacking V1 inhibitory neurons have defects in flexor–extensor alternation: during the step cycle they exhibit defective extension and prolonged flexion, causing an overall hyperflexion of the limb [50]. Mice lacking V2b inhibitory neurons show an increase in extension and a lack of flexion, causing overall hyperextension [50]. Optogenetic activation of V2b neurons also suppresses extensor activity [50]. It is therefore believed that V1 neurons restrict flexor activity during stance and facilitate the swing-to-stance transition, while V2b neurons facilitate the stance-to-swing transition by suppressing extensor activity during swing [50]. The Ia-interneurons, derived from V1 and V2b neurons, and Ib-interneurons, derived only from V2b neurons, are the predominant neuron types for controlling flexor–extensor alternation [4, 50].

Blocking both V1- and V2b-derived neurotransmission in the isolated mouse spinal cord leads to synchronous flexor and extensor activity and marked deficits in limb-driven movements, but normal left–right alternation [371]. Conversely, the commissural interneurons that contribute to left–right alternation (see V0 section) do not affect flexor–extensor alternation [352, 189, 371]. Two conclusions follow. First, V1 and V2b neurons are primarily responsible for controlling flexor–extensor alternation by acting within the ipsilateral spinal cord. Second, the rhythm-generating circuits on each side of the cord are largely decoupled from those that control alternation across the cord.

One unique feature of V1 inhibitory neurons in mice is their well-characterized physiological and transcriptional subtype diversity [36]. The V1 class includes the well-studied Renshaw cells and reciprocal Ia-interneurons, both implicated in flexor–extensor [104, 113, 295, 9, 23, 331, 371]. Notably, recurrent and reciprocal V1 types make up $< 25\%$ of the V1 class [295, 9], leaving open the question of what constitutes the other $\sim 75\%$. More recent work shows that the V1 class can be grouped into $\sim$50 transcriptionally distinct subtypes—or four clades—based on combinatorial expression of FOXP2, SP8, POU6F2 and other factors [36, 129, 336]. Each clade has a distinctive settling position, physiology and synaptic connectivity [36, 129].

Settling position, in particular, constrains input specificity, forming inhibitory microcircuits that selectively act on motor pools innervating different proximodistal muscles—evident in differences in V1→MN connectivity for hip, knee and ankle [36]. Segmental differences in transcriptionally defined V1 subsets at limb- versus non-limb levels have also been observed [126, 336]. This extensive molecular heterogeneity sug-

gests parallel anatomical or functional diversity. One possibility is that it supports precise motor-pool innervation and coordination; if so, similar heterogeneity would be expected in V2b neurons, which is only beginning to be examined at a molecular level [126]. Altogether, the high degree of molecular diversification in ipsilateral inhibitory neurons appears key to the expanded movement repertoire in mice relative to zebrafish.

### 1.5.3 Cross-species perspective on V1/V2b

In aquatic vertebrates, V1 and V2b inhibitory neurons control the speed of swimming and ensure faithful rostral-caudal propagation of activity [216, 221, 193]. In tadpoles and zebrafish for example, they provide in-phase inhibition to the CPG, including motor neurons, to regulate the length of each swim bout [216, 221]. In zebrafish specifically, V1 and V2b inhibitory neurons can be split into speed-specific subtypes and act as a brake on the locomotor circuitry [193].

It is likely that the zebrafish circuit organization is also present in the tadpole. Li et al. [221] demonstrated the presence of direct connections between aINs (corresponding to V1 inhibitory neurons) and dINs (likely V2a excitatory neurons). The aINs are known to provide early-phase inhibition to motor neurons. There was a strong correlation between aIN-derived inhibitory inputs and the frequency of swimming [221].

In addition to their shared role in the regulation of motor output across vertebrates, V1 and V2b neurons are specialized for flexor-extensor coordination in limbed vertebrates such as mice [50]. Their innervation patterns are biased in their connectivity with flexor and extensor motor pools to ensure smooth transitions through the step cycle [50]. Moreover, in tetrapods, this idea of motor pool specialization of ipsilateral inhibitory circuits can be extended further, as the settling position of V1 neurons predicts their subtype and innervation patterns [36]. It is thus likely that the diversity of V1 neurons may also enable other aspects of motor pool coordination such as fine motor control, which remains to be tested and is a crucial difference between vertebrate species.

## 1.6 Other ventral neurons

There are two additional types of ventral interneurons identified in mice, which have not been assigned to one of the cardinal classes described above. However, these types, marked by HB9 and SHOX2, are of interest since they are thought to be candidates for the rhythm-generating neurons [175, 174, 357, 358, 173, 52, 377, 101, 66]. The

HB9 neurons have a mixed neurotransmitter phenotype and progenitor domain origin. Blocking only glutamatergic transmission had no impact on locomotion, while blocking all synaptic transmission caused defects in the frequency of locomotion but not its left–right or flexor-extensor phase [66, 201]. The SHOX2 population, also known as V2d neurons and partly overlapping with V2a neurons, are ipsilateral excitatory neurons, form recurrent connections, and project to motor neurons [101, 153]. Similar to HB9 neurons, silencing them or blocking their transmission affects rhythm, but not pattern, generation [5, 227, 101]. It will be of interest to determine whether these intrinsic rhythm-generating neuron types, which have not been described in fish or frogs, are conserved between vertebrates with less or more varied locomotor demands.

## 1.7 dI6s inhibitory neurons

Modulation of motor output by dorsal interneurons is a conserved feature across all vertebrates and has typically been associated with the gating of sensory input. In the lamprey, one class of dorsal neuron has been identified: glutamatergic primary sensory neurons, which can be subdivided into touch and pressure cells [75, 114]. In zebrafish and mouse, dorsal interneurons are implicated in sensory-to-motor transmission but are much less well-characterized than ventral neurons. These putative sensory-related populations will be discussed at the end of this review.

The exception to this sensory compartmentalization of dorsal interneurons is the inhibitory dI6 population (Fig.1.6). Like all dorsal populations, the dI6 neurons express LBX1 at early embryonic stages [149, 248]. Originating from the dp6 progenitor domain, they additionally express a combination of DBX2 and PAX transcription factors [11, 167]. The dI6 inhibitory neurons fall into three subtypes based on the expression of DMRT3 and WT1: those that express one, the other or both [142, 11, 304]. In zebrafish and mice, dI6 neurons connect to other dI6 neurons and project commissurally to contact motor neurons on the contralateral side to regulate left–right alternation, rhythm generation and locomotor pattern [323, 149, 248, 37, 209, 142, 268, 103, 144, 158, 261, 300, 348].

### 1.7.1 Zebrafish dI6 neurons

The dI6 population in zebrafish is inhibitory, expresses Dmrt3a, and has been termed the CoLo (commissural local) neurons based on its anatomical projection pattern [212, 301]. The function of dI6 neurons seems to vary between developmental stages in zebrafish [301].

At escape swimming stages and in the absence of the dI6 population, initiation of the body bend is impaired [301]. This impaired response is specific to Mauthner-

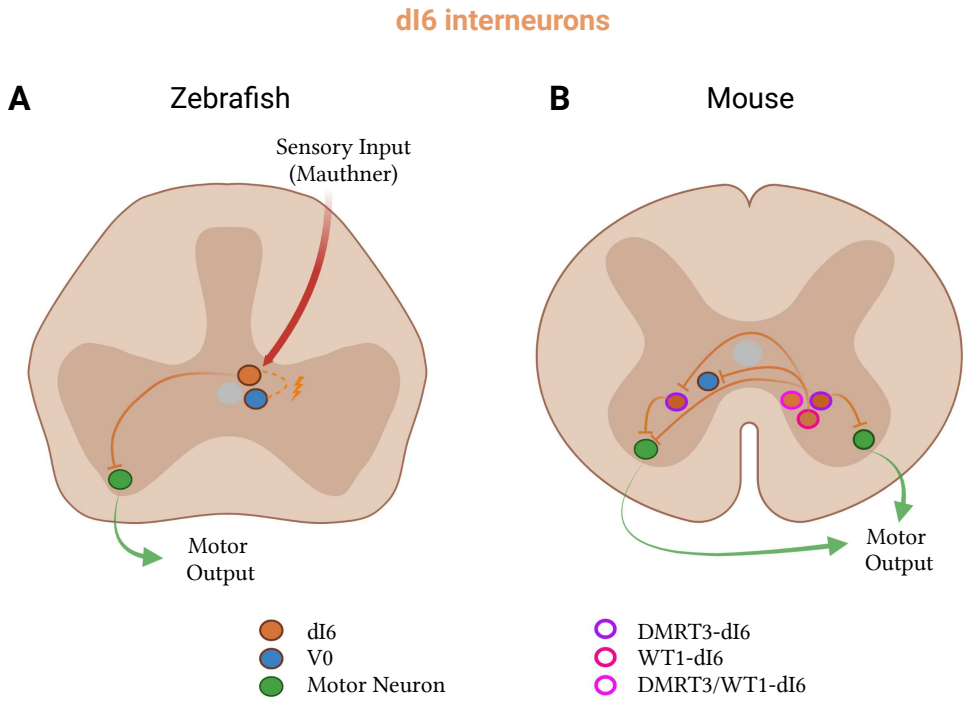


**Figure 6**

Figure 1.6: Inhibitory dI6 subtypes in zebrafish and mice. (A) In zebrafish, dI6 neurons (orange) receive input from Mauthner cells, form electrical connections with V0 neurons (blue), and project to contralateral motor neurons (green). (B) In mice, dI6 neurons split into three subclasses: DMRT3- (purple outline), WT1- (pink outline), and DMRT3- and WT1-co-expressing (pink-red outline). WT1-dI6 inhibit contralateral V0 and DMRT3-dI6 neurons, while DMRT3-dI6 inhibit motor neurons on both sides of the spinal cord.

mediated escape, in which stimulation of Mauthner cell on one side of the body activates motor neurons on the same side and simultaneously inhibits those on the opposite side [362]. Further studies showed that dI6 neurons are electrically coupled to commissural interneurons and monosynaptically connected to contralateral motor neurons (Fig.1.6A; [96, 301]). The presence of these connections, together with the altered response of dI6-ablated zebrafish, implies that the dI6 neurons usually function in escape to inhibit the firing of contralateral motor neurons [301]. Larval dI6 neurons were also found to be inhibited and thus inactive during swimming [301], suggesting that their contribution is limited to the Mauthner-mediated escape response at this stage.

At this early swim stage, recent evidence has also linked Dmrt3a-expressing neurons to the regulation of abductor motor neurons in the pectoral fin of zebrafish [348]. Abductor and adductor motor neurons alternate in their spiking, like flexor and ex-

tensor motor neurons in mammals [348]. Abductor, and not adductor, motor neurons receive strong inhibitory synapses from Dmrt3a neurons. In their absence, the timing of abductor neuron firing was impaired, while adductor unaffected. In larval zebrafish, dI6 neurons thus also regulate fin movement via abductor/adductor coordination.

During later-stage beat-and-glide larval swimming, genetic ablation of Dmrt3a led to fewer and shorter movements with decreased velocity and acceleration [93]. This contrasted with very early coiling stages in which the loss of protein had no effect [93], supporting that Dmrt3a-expressing neurons may only be recruited when the fish needs to perform stronger escape movements.

In adult fish, Satou et al. [300] demonstrated that these neurons were rhythmically active during locomotion, increased their firing probability at slow speeds, and provided mid-cycle inhibition onto contralateral motor neurons. When ablated, there was a decrease in maximum swim speed [300]. This suggests that dI6 function may change during development: first necessary for strong body bends in larvae and later, required in a speed-dependent manner in adult zebrafish.

### 1.7.2 Mouse dI6 neurons

In mice, like zebrafish, a subset of dI6 inhibitory neurons similarly expresses DMRT3. However, expression of WT1, together with GABAergic and glycinergic neurotransmitters, define a broader population of dI6 neurons in the mouse (Fig.1.6B; [142, 11, 158]). This population can be further divided into subtypes based on morphology, electrophysiology, neurotransmitters, birth order, transcription factors and axon guidance gene expression [11, 144, 304, 261, 194, 179].

Reinforcing a conserved role of the DMRT3-expressing dI6 subset in cross-body inhibition, *Dmrt3*-null mice exhibit impaired left–right as well as fore–hind limb coordination [11]. Other defects include a decrease in swim duration when mice are placed in water, and, when not in water, an increase in twitching movements, an inability to run at high speeds and a decrease in alternation of hindlimb steps during air-stepping [11]. Consistent with these behavioral observations, the DMRT3-expressing dI6 neurons in mice are known to contact V1 neurons and motor neurons on both sides of the spinal cord and are rhythmically active during fictive locomotion [11, 144, 261]. This indicates a conserved role between mice and zebrafish for the DMRT3-dI6 neurons in regulating rhythm and coordinating activity on either side of the spinal cord.

To examine the function of the rest of the dI6 population, Schnerwitzki et al. evaluated *Wt1*-knockout mice [304]. Neonatal mice with this deletion displayed uncoordinated and variable locomotor activity: a slower walk with a decreased stride

frequency and increased stride length, and loss of left–right and fore-/hindlimb coordination [304]. The anatomical projection pattern of WT1-dI6 neurons is consistent with these defects. Whereas the only known targets of the DMRT3-dI6 are the motor neurons on both sides of the spinal cord [11], the WT1-dI6 neurons have commissural projections and terminate close to, and likely onto, the DMRT3-dI6 and V0 neurons [158, 304]. V0 neurons are proposed to excite contralateral inhibitory interneurons, which in turn contact motor neurons [338, 314, 87], conferring an indirect function in contralateral inhibition onto this WT1 population. This indirect function was further supported by the acute silencing of WT1-dI6 neurons [158]. Acute silencing resulted in the elimination of left–right, but maintenance of flexor–extensor alternation, and WT1-cell bursting was tightly coupled to fictive locomotor activity of motor neurons [158]. Since WT1-dI6 do not contact motor neurons directly, but instead contact commissural interneuron subtypes, they were thus proposed to indirectly gate the activity of rhythm-generating neurons.

### 1.7.3 Cross-species perspective on dI6

Across vertebrates, dI6 neurons play a consistent role in regulating the firing of contralateral motor neurons to coordinate the left and right sides of the body. This role in left–right coordination has recently been shown to be essential for generating the characteristic gaits of horses, with mutations in *Dmrt3* associated with the emergence of new gaits in Icelandic horses [11]. Thus, in mice and likely other four-limbed mammals, dI6 neurons have diverged in their molecular, anatomical and functional properties to control same-side inhibition, rhythm and gait generation [11, 304, 158]. These diverse roles in limbed vertebrates seem to map differentially onto the molecularly distinct DMRT3 and WT1 subpopulations. DMRT3-dI6 neurons contact motor neurons directly [11] whereas WT1-dI6 neurons do not and instead receive multisynaptic input and target contralateral interneurons [158, 304].

These anatomical differences seem to confer each subpopulation with different roles in locomotion in the mouse. Work in horses, mice and zebrafish suggest that the DMRT3-dI6 neurons, with their monosynaptic connections onto motor neurons, are important for the coordination of body bend and gaits, likely via sensory integration [11, 304, 93] and potentially through flexor–extensor regulation as has been demonstrated in zebrafish [348]. WT1-dI6 neurons, in contrast, are proposed to gate the rhythm-generating circuitry in general by integrating supraspinal and proprioceptive input, as well as releasing motor neurons on the opposite side of the spinal cord from same-side inhibition, although it is yet to be shown experimentally whether this is indeed the case [304].

## 1.8 Other dorsal interneurons

The dorsal horn of the spinal cord, as the main target area of primary somatosensory afferent axons, is classically implicated in sensory processing of higher order vertebrates [234, 343, 342, 3, 47]. Our understanding of the interneuron circuitry in the dorsal horn however, is more limited than that of the ventral horn. On a developmental level, we know the molecular determinants of the identity of many classes. On an anatomical and physiological level however, the connectivity and functional properties of these molecularly defined dorsal populations remains unclear.

In mice, these interneurons project to a variety of targets including supraspinal structures, motor neurons, cutaneous afferents, proprioceptive terminals and other dorsal interneurons (Fig.1.7A; [164, 6, 210, 206]). In lamprey, dorsal interneurons similarly relay and process sensory information, consisting of a glutamatergic dorsomedial, lateral and giant interneuron population [286, 287, 306, 114]. In zebrafish, four dorsal populations have been identified anatomically, including glutamatergic commissural primary ascending (CoPA), glutamatergic and glycinergic commissural secondary ascending (CoSA), glycinergic commissural longitudinal ascending (CoLA), and glycinergic dorsal longitudinal ascending (DoLA) neurons [156, 169]. However, a function has only been well-defined for CoPA neurons, which drive early touch-mediated larval escape [266]. The lack of extensive characterization or diversity of dorsal neurons in the lamprey, tadpole and zebrafish (Fig.1.7A; [55, 76, 75, 218, 266]), likely indicates a simplification or absence of some of these populations in aquatic vertebrates – a hypothesis that will likely be tested by molecular cell type profiling in the future.

In all vertebrates, it is well known that sensory inputs can modulate locomotion in a phase-dependent manner [123, 121, 12, 303, 46]. In recent work, an inhibitory population of glycinergic deep dorsal horn parvalbumin-expressing interneurons (dPVs) were found to be active during locomotion, joining the ROR$\beta$- and SATB2-expressing populations in representing inhibitory interneurons involved in a cutaneous sensorymotor pathway [171, 200, 255]. The medial deep dorsal horn is an area of large convergence of cutaneous and proprioceptive inputs, and the dPVs integrate these multimodal sensory inputs to modulate cutaneous-evoked muscle inhibition in a state- and phase-dependent manner.

In the mouse, in which dorsal neurons are clearly present in large numbers and arguably best studied, six cardinal classes have been defined based on developmental origin, marker expression, settling position, projection pattern, and neurotransmitter type (Fig.1.7B; [164, 206, 159, 297, 367]). More recently however, an alternative organization has been proposed in which neuron types display a laminar organization that correlates with their function in encoding a specific somatosensory reflex program

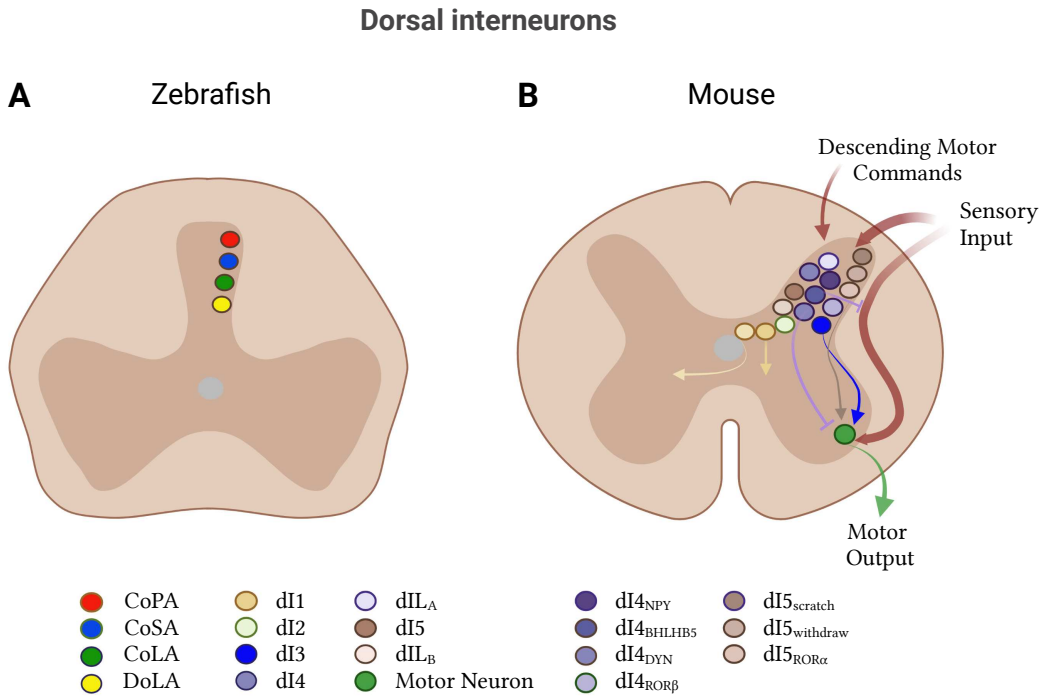


**Figure 7**

Figure 1.7: Other dorsal interneurons in zebrafish and mice. (A) In zebrafish, four classes of dorsal interneuron have been identified: glutamatergic commissural primary ascending (CoPA, red), glycinergic commissural secondary ascending (CoSA, dark blue), glycinergic commissural longitudinal ascending (CoLA, green), and glycinergic dorsal longitudinal ascending (DoLA, yellow). CoPA neurons drive touch-mediated larval escape. (B) In mice, dorsal neurons are divided into seven classes: dI1-5, dILA and dILB. The glutamatergic dI1 class (yellow) subdivides into ipsi- and contralateral populations. Little is known about the dI2 class (light green). The dI3 class (dark blue) receives cutaneous afferent input and excites motor neurons (green). The dI4 class subdivides according to sensory modality: the NPY subclass (dark purple) is associated with mechanical itch, BHLHB5 (medium-dark purple) with chemical itch, and DYN (medium-light purple) with nociception. ROR$\beta$ neurons (light purple) gate sensory afferent transmission. They also include dILA (light purple-pink) and dILB (light pink-brown) classes. dI5 neurons associated with scratch (dark brown) are located in laminae I/II, and with paw withdrawal reflex (medium brown) in laminae II/III. ROR$\alpha$ neurons (light brown) receive descending motor commands and project onto motor neurons, and function in corrective motor adjustments.

[133]. How this organization links to cardinal class identity is an active area of current study [210, 133, 290]. Here, we summarize the properties of dorsal interneurons in the mouse in relation to their developmental cardinal class identity: ipsilateral and contralateral dI1, dI2, dI3, dI4, dIL, and dI5.

### 1.8.1 dI1/dI2 excitatory neurons

dI1 and dI2 neurons originate from ATOH1- and NEUROG1-expressing progenitors, respectively [206]. Atoh1 mouse mutants lack dI1 neurons, extend NEUROG1 expression, and thus generate more dI2 neurons [143]. They display minor motor defects [364], that may result from either the loss of dI1 or gain of dI2 neurons. The dI1 population in mice is composed of an ipsilateral (dI1-ipsi) and a contralateral LHX2-expressing (dI1-contra) population, both of which express the transcription factors BARHL1/2 and are glutamatergic [359, 98, 226]. During development, Barhl2 specifies dI1 subtype diversity such that in Barhl2-null mice, the dI1-ipsi subpopulation expresses the dI1-contra transcription factor LHX2, and less of the dI1-ipsi-enriched transcription factor BARHL1 [98]. The dI1-ipsi population in these mice also exhibit a dI1-contra settling and projection pattern, suggesting that BARHL2 is important in specifying dI1 subtype diversity [98]. Unlike dI1, dI2 interneurons lack BARHL2 and LHX2/9, and instead are characterized by FOXD3, LHX1 and LHX5 expression during development [18, 94]. In mice, very little is known about dI2 anatomy or function. A recent study in chick however, showed that dI2 neurons at limb levels receive sensory and premotor interneuron input, and project to the cerebellum [155]. Silencing of dI2 neurons in chick results in abnormal hindlimb stepping [155], implicating them in the high-level coordination of limb movement.

### 1.8.2 dI3 excitatory neurons

The dI3 interneuron class is distinguished from other spinal interneurons by the expression of the LIM homeodomain transcription factor ISL1 [222, 165]. This class is known to receive direct low-threshold cutaneous afferent input and form excitatory glutamatergic connections with motor neurons and other rhythm-generating interneurons [63, 62]. It was demonstrated that the elimination of glutamatergic transmission from these neurons leads to a loss in grip strength in mice, implicating this population in grasping, likely by gating sensory transmission [63]. Bui et al. suggested that the dI3 neurons are important for functional recovery following spinal cord transection, since their removal had little effect on locomotor activity but a large negative impact on recovery [62]. They proposed that dI3 neurons compare sensory and locomotor input to compute a prediction error, which could be used to correct locomotor output. It is likely that this is also the mechanism that allows this group of interneurons to produce the appropriate grip force [62].

### 1.8.3 dI4 inhibitory neurons

dI4 interneurons, defined by the expression of PTF1A and GBX1/2 during development [136, 242, 354], can be segregated by their birth timing into two subpopulations: the early-born dI4 and late-born dILA population in mice, which are characterized by the homeodomain factors LHX1/5 and PAX2, respectively [136, 30]. For both populations, PTF1A is necessary for dI4 interneurons to adopt a GABAergic neurotransmitter profile [136, 242, 354]. The connectivity and synaptic differentiation of these interneurons is determined by their sensory targets [30]. In general, silencing dI4 leads to hypersensitivity to mechanical or thermal stimuli and increased pain and itch responses, whereas their activation has the opposite effect [15, 102, 125, 262, 81, 109, 243], directly implicating these populations in the processing of sensory stimuli. Through feed-forward inhibition of motor neurons and presynaptic inhibition of sensory and other interneurons, one such role of dI4 is to filter sensory signals according to the phase of the locomotor cycle [108, 282, 289, 120, 141]. Recent work has also shown that dI4 subtypes segregate according to sensory modality – with those expressing NPY preferentially associated with mechanical itch, BHLHB5 with chemical itch, and DYN with nociception [280, 102, 184, 45, 199]. Another subset of dI4 neurons expressing ROR$\beta$ modulate the motor output during walking by gating sensory afferent transmission [200].

### 1.8.4 dI5 excitatory neurons

LBX1-positive neurons are divided into two populations, one expressing PAX2 (dI4, dI6, and dILA) and inhibitory, and the other TLX3/LMX1B (dI5 and dILB) and excitatory [149, 248, 72, 242]. The dI5 dorsal progenitor domain also produces the excitatory dILB subset [149, 248]. The dI5 interneuron population expressing TLX1/3, LMX1B and ASCL1 (MATH1), conveys information about itch, temperature, static and dynamic touch [134]. Ablation of dI5 strongly affects different aspects of somatosensation [337]. Recent studies have revealed that sensory modalities map onto spatially, instead of molecularly, distinct dI5 subpopulations in the spinal cord [133]. The scratch reflex, for example, is produced by cells in lamina I/II, and the paw withdrawal reflex by those in lamina II/III. Additionally, distinct modules encode low-threshold mechanical stimulation [2], and static and dynamic tactile reflexes, with the latter falling largely into the molecularly distinct ROR$\alpha$ subpopulation [33, 34, 330, 71, 361, 133]. This population receives descending motor commands and projects onto motor neurons. It is a key part of the spinal touch circuitry that underlies corrective motor adjustments [45].

In summary, most of the work on dorsal populations has been carried out in mice.

Due to the multi-modal nature of sensory perception and integration in the mouse as compared to the fish, it is thus not surprising that current evidence supports that the dorsal interneuron populations of mice are more numerous and exhibit much greater heterogeneity than in simpler vertebrates. This hypothesis that the dorsal spinal cord expanded and diversified over vertebrate evolution can be addressed in the future with high-throughput molecular and physiological techniques that are increasingly becoming feasible in non-mammalian and less characterized vertebrates.

## 1.9 Discussion

Our comparison of interneurons across vertebrates finds several potent examples of neuron-to-function conservation, but just as many of new neural classes and subtypes that emerge with the more muscle groups and complex movement patterns of higher order species. Accordingly, the division of existing cardinal classes into multiple subclasses appears to be a prevalent theme going from lamprey to zebrafish to mice and more broadly, from swimming-to-limb-based movement.

### 1.9.1 Conservation of interneurons across vertebrates

One common theme to all vertebrate spinal circuits is the conservation of the three-part basic spinal rhythm-generating circuits beginning with the most primitive extant vertebrate, the lamprey. This architecture of motor neurons, ipsilateral V2a-type excitatory neurons, and commissural V0-type inhibitory neurons is present in the lamprey, tadpole, zebrafish and mouse spinal cord. In addition, zebrafish and mice have all ventral cardinal classes including not only the V2a (Fig.1.2) and V0 (Fig.1.4), but also the excitatory V3 (Fig.1.3) as well as the inhibitory V1 and V2b (Fig.1.5), and dorsal dI6 (Fig.1.6) populations.

We also observe functional conservation between finned and limbed vertebrates, with subclasses for left–right and rostral-caudal coordination, as well as those for graded muscle recruitment with increasing drive, common to both. In zebrafish and mice, for example, the excitatory V2a, V0, and inhibitory V1/V2b neurons are composed of multiple speed-specific subtypes, thus controlling the frequency of locomotion. The V0v and V0d subpopulations are important for coordinating diagonal activity and providing mid-cycle inhibition in both species. These subclasses are thus responsible for pan-vertebrate features such as speed-dependent recruitment of motor neurons, and coordination along and across the body axis.

### 1.9.2 Species-specific interneuron subpopulations

Between vertebrate species that swim versus walk however, there are notable differences. In some cases, existing neuron types for swimming seem to have taken on new roles in limb-based movement. For example, V1 and V2b inhibitory neurons, in addition to coordinating basic features of locomotion across vertebrates such as the frequency of movement, also regulate flexor-extensor alternation in limbed vertebrates. The dI6 class, necessary for the escape response and mid-cycle inhibition to motor neurons in zebrafish, also controls gaits in limbed organisms. The V0 classes have additionally taken on the role of speed-dependent left–right coordination in mice.

From this literature review, it is also clear that the more complex and variable the movement patterns and gaits of a vertebrate, the more their interneurons have been compartmentalized into distinct subtypes. This subdivision is based on factors such as birth date, projection range, physiology, recruitment threshold and function. Exemplifying this, the V2a population in mice is split into one subpopulation that receives locomotor drive and one that does not. These subpopulations differ in their marker expression and projection patterns along the rostrocaudal axis of the spinal cord. This diversity is directly linked to greater dexterity of the forelimbs. In the same manner, the subdivision of the V1 class in mice into around 50 molecularly distinct types is likely to have allowed the class to take on new functions in flexor-extensor control, mediated by Ia- and Ib-inhibition [23, 50, 36, 129, 336]. The V3 population, which seems to contribute to excitatory drive in fish and mice, can be split into subpopulations in mice, which have not been found in zebrafish, and are necessary for the generation of the trotting gait [44, 74, 95, 368]. These subdivisions allow them to control the balance of activity on both sides of the spinal cord more precisely, essential for locomotion on land.

Entirely new cardinal classes of interneurons also seem to have developed to facilitate limbed locomotion. The V0c and V2c subclasses were found in mice, homologues of which have not been discovered in zebrafish. Many new sensory dorsal interneuron classes are additionally present in mice but have not been identified in lamprey, zebrafish, or tadpoles, possibly enabling specific features of locomotion in terrestrial sensory environment. Dorsal neurons in mice are molecularly heterogeneous and have overlapping but sensory-specific functions [134]. From work in the cat, it has become clear that sensory inputs are important for initiating and maintaining an appropriate locomotor rhythm by regulating phase changes during stepping and modulating the amplitude of motor output [333]. Dorsal interneurons also project supraspinally and receive descending input to regulate descending pathways [45, 155]. Since dorsal spinal interneurons regulate these sensory pathways, one can be certain that they will exhibit large differences between water- and land-based animals, consistent with

current observations of dorsal subtype radiation from zebrafish to mouse (Fig.1.7). The specific differences at a molecular, anatomical and functional level will be an interesting area to examine in the future.

Location, connectivity, proportion and number of neurons of each class are also important parameters that can change the output of a neuronal circuit in a species-specific manner. The same class could be present in two species, but its function could differ. Exemplifying this, some classes can be subdivided differentially between species based on the location of their cell body. For the V1 population, this location can be important to enable precise connectivity between interneurons and motor neurons, for example Bikoff et al. [36]. The grouping of V2a interneurons, based on rostrocaudal location, also exemplifies this principle. As has been best shown in the turtle, there are also large differences in distribution between interneurons connected to functionally distinct motor pools [137]. Premotor interneurons that project to axial muscles are distributed symmetrically on either side of the spinal cord, while those that connect to limb motor neurons are mainly ipsilateral. These interneuron subpopulations can be distinguished by their genetic profile and neurotransmitter identity. On the other hand, an interspersed distribution of interneuron subtypes that are recruited at different speeds can allow smooth transitions between speeds, as exemplified by the well-defined slow, intermediate and fast circuits of zebrafish [24].

Changes in connectivity between neuron classes are likely to have also been necessary for producing more complex movement patterns. Supraspinal connectivity of the cervical V2a subtypes in mice enables precise motor control of the forelimbs [162]. Inter-connectivity of interneurons may also be important for relaying, gating and distributing motor commands. Recent studies have also highlighted that interneurons subdivide based on whether they project locally or long-range [254]. V3 neurons, for example, are divided into a local and an ascending population that is important for trot in mice [368]. The V2a class, in addition to their role in regulating flexor-extensor activity, has long-range V2a and V2b neurons that are important for ipsilateral body coordination [163]. In limbed vertebrates, that require coordination at and across highly variant regions of the body, it is likely that more examples of such local and long-range divisions with distinct functions will be found for other classes in the future. This may be especially important for limbed vertebrates with high dexterity, such as mice and humans, and movements that require intricate limb-torso or inter-limb coordination, such as trotting in horses.

However, it is important to consider that similar locomotor outputs can be generated with different circuit connectivity. This has been demonstrated in the crab stomatogastric system, and two related species of nudibranchs, and has been proposed to ensure robust circuit function given individual variability [229, 294]. This

suggests that there could be some flexibility in how spinal circuits connect to generate a movement pattern.

It is also still unclear to what extent the diversity of spinal neuron types in limbed vertebrates scaled with evolutionary time. Spinal neuron-type diversity increased with the complexification of the body plan and the control of limbs, as we have summarized here. Yet some of the core elements needed for limb control, such as flexor and extensor limb motor neurons and Ia-inhibitory interneurons, are likely to already present in skates and sharks [83, 183]. Therefore, these elements may have existed in the ancestor of all vertebrates with paired appendages, not just those with limbs. Future molecular and functional studies across even more finned and limbed vertebrates will be crucial in addressing this question.

### 1.9.3 Beyond interneurons

Although we have chosen not to focus on motor neurons in this review, it is now clear that they can also influence the pattern and frequency of locomotion, as well as connectivity with premotor networks [137, 176, 20]. In zebrafish, this is mediated by gap junctions, which allow motor neurons to exert influence over the strength of excitation retrogradely [328], whereas in mice, this is mediated by synaptic glutamate release [240, 251]. Additionally, motor neurons receive strong input from each other, with the fast-type receiving greater excitation than the slow-type [31].

The traditional concept of interneuron subtype-based locomotor pattern generation is now also being brought into question by work in the turtle. The spinal networks for swimming and scratching movements in turtles largely overlap [25, 157]. The identification of spinal neurons in the turtle is still much less advanced than in the mouse. However, it is clear that the interneurons active during swimming and multiple forms of scratching likely contribute directly to motor output, while the interneurons specialized for one behavior receive hyperpolarizing inhibition during the others [28, 26, 27]. Additionally, recordings from the lumbar spinal cord of the turtle demonstrate that the neural circuitry exhibits "rotational dynamics," rather than alternating activity as proposed in the half-center model [223]. These observations, although not inconsistent with the diversity of interneurons present in the spinal cord, propose a new way of thinking about pattern-generation as a neural network, as opposed to neural subtype, property.

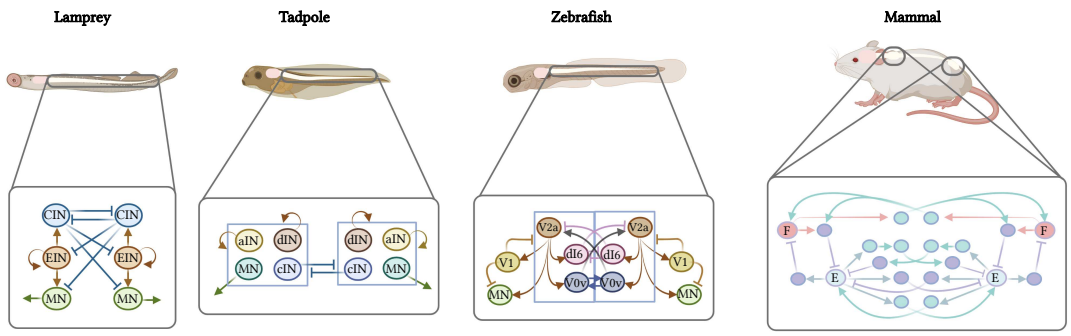


**Figure 8**

Figure 1.8: Computational models of the lamprey, tadpole, zebrafish and mammal spinal cord networks. The region of the spinal cord modeled is indicated. Far left: lamprey CPG with excitatory interneurons (EINs), inhibitory commissural interneurons (CINs), and motor neurons (MNs). Middle left: CPG model of the tadpole with the same neuron types as the lamprey plus additional ipsilateral inhibitory neurons (aINs). Middle right: CPG of the zebrafish larva at a stage when it can perform beat-and-glide swimming. Compared to the tadpole model, this model has additional contralateral excitatory neurons. Far right: model of the mammalian spinal cord showing connections between right and left rhythm generators (red: flexor unit, F; blue: extensor unit, E) at the limb level. Turquoise circles represent excitatory interneurons while purple circles represent inhibitory interneurons.

### 1.9.4 Computational dissection of neuron-to-behavior relationships across species

Computational models have emerged as a useful tool to guide our understanding of the function of interneurons in rhythm generation across species (Fig.1.8; [208, 105, 178, 180, 203, 204, 160, 198, 296, 277, 291, 314, 43, 87, 88, 202, 116, 368]), and can help us to address some of the questions raised in this review, namely whether movement complexity parallels neuronal complexity. They can explain experimental observations, by testing the minimal requirements to reproduce these observations in silico. They can also help generate hypotheses about how neuronal components can be expected to change their configuration to produce varying output across development, stimuli, or species.

In zebrafish, CPG models have provided a potent demonstration of how the addition of interneuron classes can lead to increasingly complex motor patterns during development [284]. Three models were built to test the neuronal basis of the zebrafish's transition from single-coiling to double-coiling to beat-and-glide swimming. The first model generated single coiling with three components: a pacemaker kernel, V0d-equivalent interneurons, and motor neurons. The addition of V2a and V0v-equivalent neurons, and new chemical synapses, in the second model resulted in the

emergence of double coiling. Finally, in the third model, the division of the circuitry into network oscillators, the addition of V1-equivalent interneurons, and a change to mainly chemical synapses gave rise to beat-and-glide swimming. The transition from simple to more complex architecture in these models supports the hypothesis that movement complexity requires increased neuronal and synapse heterogeneity, and generates testable predictions of which neuronal components are required at each stage.

Spinal cord models have also provided a foundation for dissecting how sensory feedback shapes movement. In lamprey and salamander, they have underlined the importance of such feedback in gait transitions and action selection, showing that changes in descending input upon sensory stimulation produced variant motor output [208, 105, 180, 160, 198, 296, 202]. In tadpole, they have shed light on the biophysical properties of the spinal circuits that control time-delays in swimming in response to sensory stimulation [277, 43, 116].

CPG models have also assigned a function, or lack thereof, to anatomically- or molecularly-defined neuron subtypes. It was previously believed, for example, that ipsilateral inhibitory interneurons formed a key part of the rhythm-generating circuitry in the lamprey. However, biophysical models demonstrated ipsilateral excitatory and commissural inhibitory, but not ipsilateral inhibitory, neurons were required [178, 203, 204]. The maintenance of rhythm in the absence of ipsilateral inhibition resulted in a major revision to our understanding of the cellular basis of lamprey locomotion.

Comparing across species, models of the mammalian CPG are much more complex in their neuronal composition and connectivity than those of the lamprey, tadpole or salamander. The most recent mammalian CPG model consists of 12 interneuron types, as opposed to the two in the lamprey [178, 204, 368]. Importantly, the CPG models can be, and are periodically, updated based on new experimental evidence [292, 314, 87, 88, 368], helping to integrate new data into the framework of our current understanding. The most recent update incorporates newly identify lumbar V3 neurons with ascending projections to cervical areas [368]. Experimental evidence shows silencing the entire V3 population led to instability of the trotting gait. However, only with the updated model can this defect be localized to the ascending V3 population.

CPG models also provide a means to translate between different forms of pattern generation, such as breathing and locomotion [132, 321, 92], thus yielding a generalizable understanding of variation in rhythmic circuits. Moving forward, they provide a powerful means to probe the cellular basis of locomotor variation within and across species.

## 1.10 Concluding remarks

Cross-species comparisons in vivo and in silico can drive observations and predictions about the diversity and function of spinal interneuron types in relation to movement. From this review and its comparison of vertebrate motor circuits, we observe that heterogeneity within a class correlates with finer-tuned control of muscles and a greater movement repertoire. This heterogeneity gives rise to a variety of circuit-level features that facilitate limbed, as opposed to swim, locomotion. Recent development of new tools to examine lesser-studied vertebrates along the swim-to-limb evolutionary trajectory will bring us closer to identify their spinal cords and the remarkable symphony of interneurons across species.

# 2 Review of Spinal Interneurons in Tadpole

## 2.1 V2a-like excitatory neurons in *Xenopus* tadpoles (dINs)

In hatchling *Xenopus laevis*, the principal ipsilaterally projecting excitatory interneurons that drive swimming are the descending interneurons (dINs) [327] (Fig.2.1). Functionally, these neurons occupy the V2a role in the tadpole locomotor CPG: they are glutamatergic, project descending axons on the ipsilateral side, and provide the core premotor excitation to the network. dINs form a longitudinal column spanning caudal hindbrain into spinal cord, fire reliably one spike per cycle during swimming, and excite other premotor elements and motoneurons. Stimulation of individual dINs in the caudal hindbrain can start and stop swimming, underscoring their driver role in rhythm initiation and maintenance [217].

Two intrinsic/network features help explain their reliability. First, dINs are electrically coupled to each other (via gap junctions), which promotes synchronous recruitment and stabilizes the rhythm [215]; pharmacological disruption of coupling weakens dIN activity and compromises swimming. Second, dINs express the hyperpolarization-activated current $I_h$ selectively, and the swim rhythm at stages $\tilde{3}7$–42 relies on post-inhibitory rebound (PIR) in dINs following phasic mid-cycle glycinergic inhibition from commissural inhibitory interneurons (cINs) [264]. Together, $I_h$ and PIR make dINs especially likely to fire on each cycle once the CPG is active.

In terms of inputs and outputs, dINs receive descending drive and cutaneous sensory-evoked inputs and in turn excite motoneurons and other CPG interneurons, forming the ipsilateral excitatory "half-center" that alternates with contralateral inhibition to generate left–right anti-phase swimming [246]. In contrast to zebrafish, where V2a neurons split into clear speed-tuned subclasses, evidence for sharply defined dIN subclasses in early *Xenopus* is limited; the embryonic network is notably compact (dINs, aINs, cINs, motoneurons) [275, 272]. Some functional diversity exists (e.g., repetitive-firing dIN variants under certain sensory conditions), but overall dINs appear comparatively homogeneous at these stages.

Altogether, the *Xenopus* dIN population exemplifies a minimalist V2a-like module: ipsilateral glutamatergic premotor neurons that are electrically coupled, endowed with rebound-friendly membrane properties, capable of triggering locomotion when singly activated, and essential for sustaining the swim rhythm once it begins. This simple

architecture—ipsilateral excitation plus reciprocal contralateral inhibition—provides a tractable vertebrate template for studying how V2a-type excitatory drive builds spinal locomotor patterns.

## 2.2 Commissural inhibitory interneurons in hatchling *Xenopus laevis*

In the hatchling tadpole swim CPG, contralateral inhibitory commissural interneurons (cINs) provide the glycinergic "reciprocal inhibition" that enforces left–right alternation (Fig.2.1). cINs fire one spike per cycle in anti-phase to ipsilateral motoneurons and generate mid-cycle IPSPs on the opposite side, a defining feature of the swim rhythm [323, 322, 274]. Anatomically, cIN somata lie dorsally; their axons cross ventrally at the midline and then T-branch to ascend and/or descend on the contralateral side, establishing intersegmental inhibition over measurable distances along the cord [323, 363, 324]. Glycinergic identity and the timing of the inhibitory waveform were established with physiology and pharmacology (e.g., strychnine block) and later supported by immunohistochemistry [322, 274].

Functionally, reciprocal inhibition from cINs is essential both for patterning (left–right alternation) and for rhythm maintenance. Classic slow pharmacology suggested rhythms could persist briefly without inhibition, but millisecond-scale, unilateral "fast silencing" showed that depressing reciprocal inhibition on one side rapidly stops activity on the other, revealing that cIN-mediated inhibition is critical for sustaining locomotor output in the intact network [246]. The longitudinal reach of cIN outputs—quantified by mapping mid-cycle IPSPs relative to identified cIN groups—supports a role in coordinating rostrocaudal progression of the pattern during swimming [324, 363].

cIN inhibition also shapes the cellular mechanism of cycle-to-cycle timing. Mid-cycle glycinergic IPSPs onto ipsilateral excitatory premotor neurons (dINs) help set the next spike through post-inhibitory rebound, integrating with the dINs' intrinsic properties to produce a reliable, alternating rhythm [274]. Other inhibitory classes (e.g., ipsilateral ascending aINs) gate sensory pathways during locomotion, but cINs are the principal source of the crossed inhibition that organizes bilateral alternation in this minimal vertebrate CPG [216, 274].

## 2.3 Ascending inhibitory interneurons in *Xenopus laevis* hatchlings

In the hatchling Xenopus laevis spinal cord, the principal ipsilateral inhibitory class is the ascending interneuron (aIN) (Fig.2.1). These neurons are glycinergic, extend a characteristic ipsilateral ascending axon (often with a short descending branch near

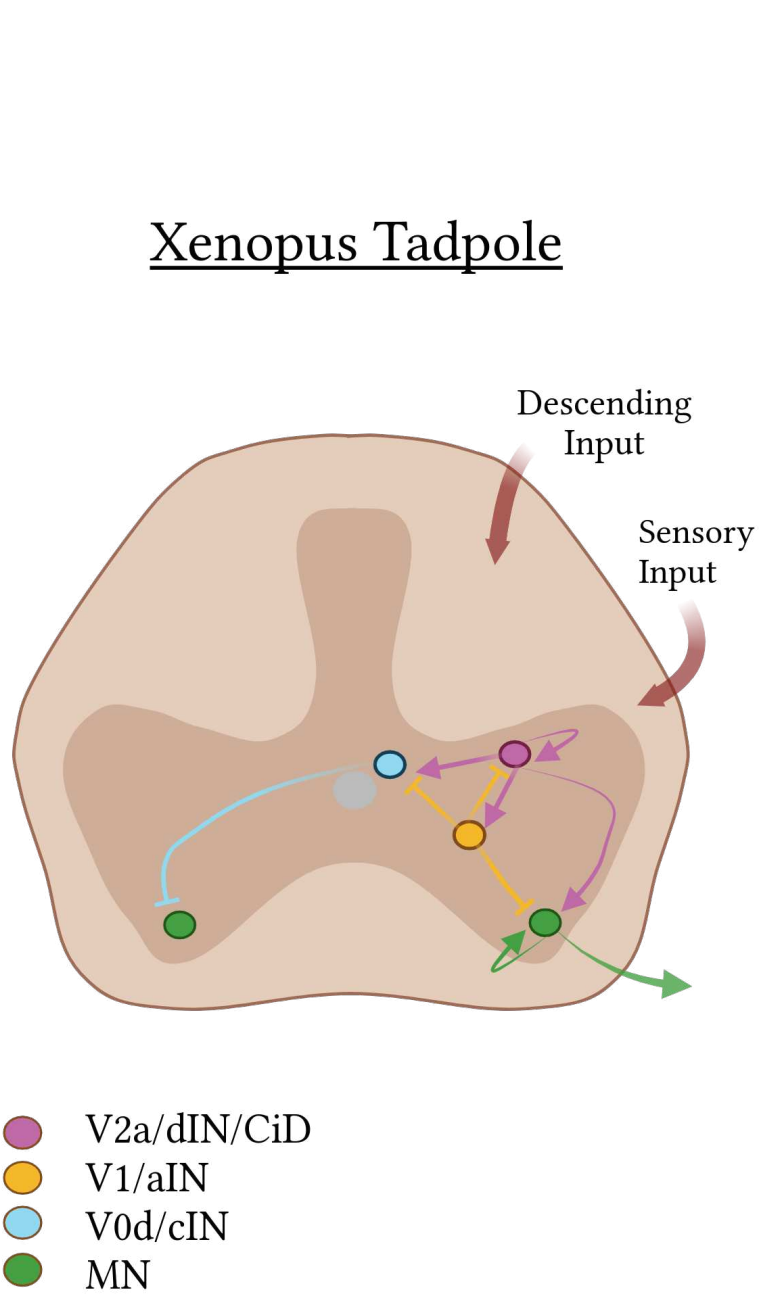


Figure 2.1: Overview of the types and connections of spinal interneurons in the *Xenopus* tadpole. In pink are the ipsilaterally-projecting excitatory descending interneurons, equivalent to V2a neurons. In yellow are the ipsilaterally-projecting inhibitory ascending interneurons, equivalent to the V1 neurons. In blue are the contralaterally-projecting inhibitory neurons, equivalent to the V0d neurons. In green are the motor neurons which project to the muscles. The spinal cord also receives descending and sensory input. Neurons and connections of just one side shown for simplicity.

the soma), and participate directly in the swim central pattern generator (CPG). Functionally, they deliver in-cycle (on-phase) inhibition within the ipsilateral half-center and thus help shape each swim cycle's timing and gain [216, 318, 274, 273, 73]. Morphology and role place aINs alongside the V1-like inhibitory population in other vertebrates: in early Xenopus, this single ipsilateral inhibitory class appears to subsume tasks that are later divided among multiple subtypes in species with more elaborate locomotor repertoires [73, 273].

A hallmark contribution of aINs is sensory gating during movement. During fictive swimming, aINs provide rhythmic, glycinergic postsynaptic inhibition to cutaneous sensory pathway interneurons, reducing reflex gain so that reafferent touch does not disrupt the ongoing motor pattern (classical "gating") [318, 216]. In addition, paired recordings show that aINs supply negative-feedback inhibition to motoneurons and other CPG interneurons, curtailing firing and stabilizing the rhythm—i.e., acting as an intrinsic brake on ipsilateral excitation [221]. Together, these actions allow strong but orderly motor bursts while keeping reflex pathways appropriately tempered during locomotion [216, 221, 274].

Recent work has begun to link aIN intrinsic properties to their recruitment in the developing network. In hatchlings, the reliability with which inhibitory interneurons (including aINs) fire during swimming correlates inversely with input resistance, and synaptic drive scales with those intrinsic properties—an organization that differs from the classic size principle and likely supports robust rhythm generation in an immature circuit [117]. Overall, aINs exemplify a compact solution for ipsilateral inhibition in the tadpole CPG: they gate sensory inflow, apply on-phase feedback to motor and premotor targets, and help set bout structure and frequency within a small, well-defined neuron set [216, 221, 274, 73].

## 2.4 Motor neurons in *Xenopus* tadpoles

In hatchling Xenopus laevis, spinal motoneurons form the final common pathway of the swim CPG, projecting axons out the ventral roots to segmental myotomes and firing in a left–right alternating pattern that propagates rostrocaudally during fictive swimming [274, 278] (Fig.2.1). At these early stages, individual MNs typically produce one spike per cycle in tight synchrony with their segmental pool, providing a faithful readout of premotor drive [274]. Anatomically, axial MN somata lie ventrally; their central axon collaterals run longitudinally and make en passant synapses within the spinal cord [278].

Excitatory premotor drive to MNs is glutamatergic and has the classic dual-component profile: a fast non-NMDA component and a slow NMDA component first defined in the embryo spinal cord by Dale & Roberts [86, 84]. These components

summate across cycles and help sustain rhythmic output once swimming is initiated [274]. On the inhibitory side, MNs receive mid-cycle, glycinergic IPSPs from commissural interneurons that enforce left–right alternation; blocking glycine receptors with strychnine disrupts the pattern, demonstrating the necessity of reciprocal inhibition for proper motor output [322]. In addition, the ascending inhibitory interneurons (aINs) provide negative-feedback inhibition that can limit MN firing and gate sensory inputs during locomotion [221].

MNs also contribute centrally to the rhythm via MN→MN synapses. Paired recordings reveal both cholinergic chemical synapses and electrical coupling between synergistic MNs on the same side; together these connections add fast on-cycle excitation and promote reliable, synchronous firing [259, 260]. Developmentally, electrical coupling is strong at hatch and helps synchronize the MN pool; pharmacological reduction of coupling desynchronizes output, underscoring its role in pattern robustness [369].

Overall, tadpole MNs are driven by dual-component glutamatergic excitation, sculpted by precisely timed glycinergic inhibition, and locally reinforced by cholinergic and electrical MN→MN coupling. This compact arrangement supports the simple, robust axial swim pattern that has made the Xenopus preparation a canonical vertebrate model for linking identified premotor circuits to motor output [274, 278].

## 2.5 Modelling the spinal cord of *Xenopus*

*Xenopus* offers a uniquely tractable system for studying spinal cord function and development across a natural transition in locomotion. In early larvae, a compact and well-characterised spinal central pattern generator (CPG), consisting of the four spinal neuron types described above, produces axial, undulatory swimming. During metamorphosis, hindlimb circuits emerge and are progressively reweighted until limb-driven hopping predominates, with clearly defined stages marking intermediate network configurations [274, 273, 250] (Fig.2.2). This staged progression provides an experimentally aligned timeline for testing hypotheses about circuit addition, reorganisation, and coordination (e.g., axial–limb interactions and left–right coupling), while standard ex-vivo preparations allow stable recordings of fictive locomotion that link identified neuronal properties to network output [274, 273]. Practically, *Xenopus* combines high fecundity, rapid development, and excellent experimental access (intracellular/whole-cell recordings, calcium imaging, opto/chemo-genetic manipulations). Importantly, unilateral ("half") manipulations can be achieved by injecting a single blastomere at the two-cell stage to generate left–right mosaic animals, enabling within-animal controls for causal circuit analyses [320]. Together, these features make Xenopus a powerful model for linking cellular and synaptic mechanisms to the evolv-

ing computations of the spinal locomotor system [274, 273].

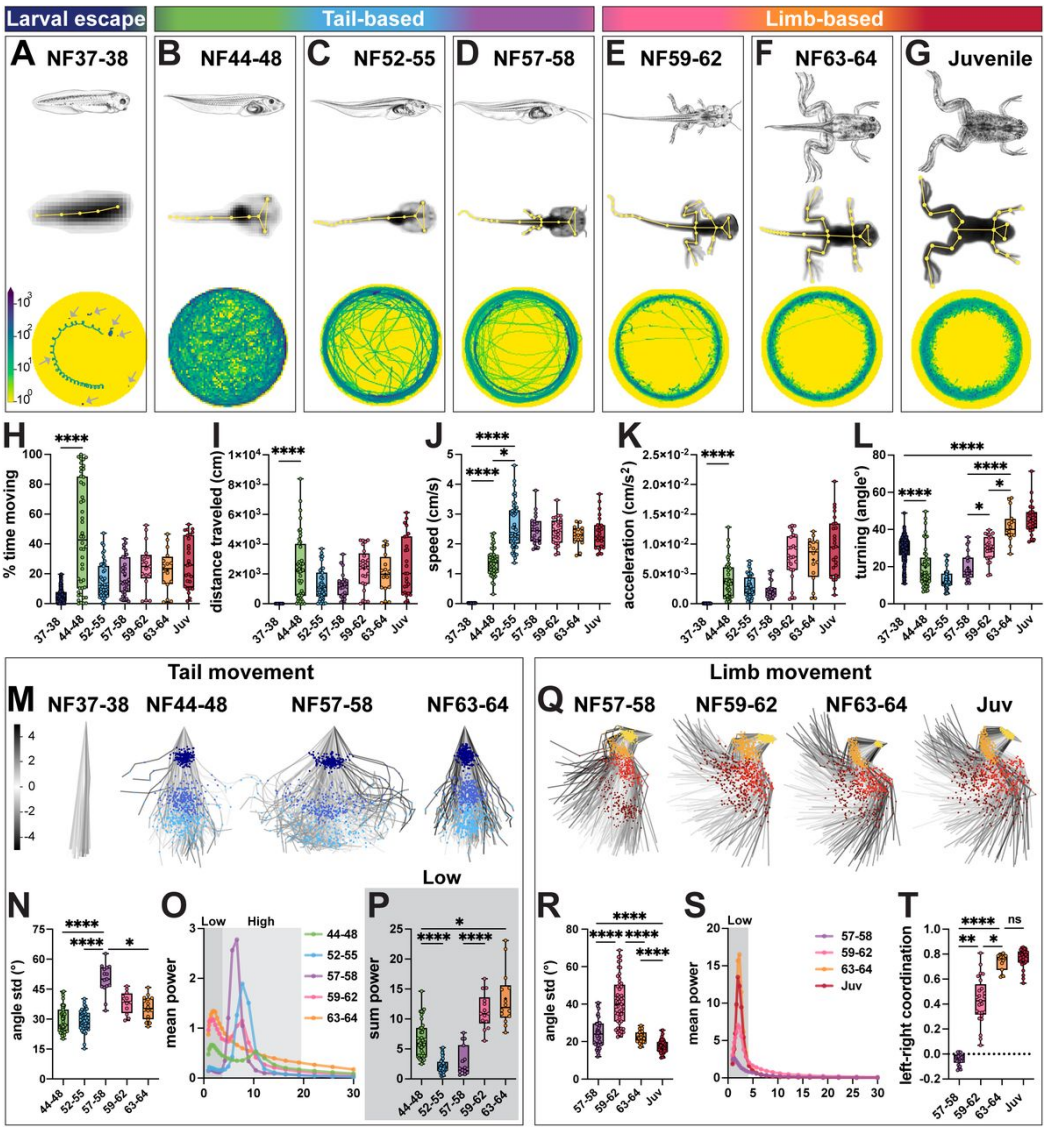


Figure 2.2: Loss of tail and emergence of limb movement during frog metamorphosis. A-G. Larval escape, tail-based and limb-based locomotion during Xenopus laevis metamorphosis. Trajectories of the distance traveled show distinct patterns at each stage: coiling escape swimming (A), free-feeding exploration (B) and a transition to edge tracking at juvenile (G) stage. H-L. Quantification of tadpole and frog movement: percentage of time spent moving (H), length of the distance traveled (I), speed (J), acceleration (K), and turning (L). M-P. Range and frequency of tail movement across tadpole metamorphosis. Range of movement at the tail tip (N). Mean power spectrum of frequency of tail tip oscillations for each stage of metamorphosis, with low (0.9-4.5 Hz, dark gray) and high (4.5-20 Hz, light gray) frequency bins highlighted (O). Amount of tail tip movement in the low frequency bin, represented by the sum power (P). Q-T. Gain of hindlimb movement during frog metamorphosis. PCA plots represent position of the hindlimb and its range of movement (Q). Quantification of range of knee movement (R). Mean power spectrum of knee oscillations for each stage of metamorphosis (S). Coordination of the left and right knees (T). (Taken and caption modified from [350].

# 3 Hodgkin–Huxley Neurons for Modeling the *Xenopus* Tadpole Spinal Cord

## 3.1 Overview

As outlined in the previous chapter, the early *Xenopus laevis* spinal circuit can be approximated by a compact set of identified neuron classes—descending excitatory interneurons (dINs), commissural inhibitory interneurons (cINs), ascending inhibitory interneurons (aINs), and motoneurons (MNs)—that together generate left–right alternating swimming [272]. In this chapter I build a proof-of-concept network using Hodgkin–Huxley (HH) neurons to capture class-specific excitability and key interactions observed experimentally. I first recall the HH formalism, then describe how I obtained (i) tonically firing HH neurons to represent aINs, cINs, and MNs, and (ii) a transiently firing HH neuron to represent dINs, following the biophysical logic reported for tadpole dINs (rebound excitability, electrical coupling) [215, 264, 217, 117]. I then examine the impact of adding calcium conductances and strengthening gap junction coupling between dINs, and finally assemble an eight-cell bilateral network (four cells per side) that already exhibits alternating activity and silence, consistent with mutual inhibition across sides [323, 322, 219]. Since the aim was methodological exploration, I conclude by motivating a subsequent transition to a simpler integrate-and-fire (I&F) model with far fewer parameters.

## 3.2 Methods

An HH neuron represents the membrane as a capacitor in parallel with voltage-gated ionic conductances:

$$C_m \frac{dV}{dt} = I_{\text{app}} - g_{\text{Na}} m^3 h\,(V - E_{\text{Na}}) - g_{\text{K}} n^4\,(V - E_{\text{K}}) - g_{\text{L}}\,(V - E_{\text{L}}) - \sum_c g_c\, a_c(V,t)\,(V - E_c) + I_{\text{syn}} + I_{\text{gap}}. \tag{3.1}$$

Here $m, h, n$ (and any additional $a_c$) are gating variables obeying first-order kinetics,

$$\frac{dx}{dt} = \alpha_x(V)\,\big(1 - x\big) - \beta_x(V)\,x,$$

with $\alpha_x, \beta_x$ chosen to set class-specific excitability. Synaptic and electrical coupling are

$$I_{\text{syn}} = \sum_j g_{ij}^{\text{syn}}\, s_{ij}(t)\,(V - E_{\text{syn}}), \qquad I_{\text{gap}} = \sum_j g_{ij}^{\text{gap}}\,\big(V_j - V_i\big),$$

with $s_{ij}$ obeying standard rise/decay dynamics. This formalism reproduces spike initiation, refractoriness, and subthreshold integration with biophysical interpretability [177].

To model the tadpole CPG, I create four neuron classes representing the dINs, cINs, aINs and MNs. The cINs, aINs and MNs are given the same parameters, as in [116], as they all respond to current injection in a similar way, firing tonic spikes. The parameters of the dINs are hand-tuned so that they show transient spiking in response to current injection.

## 3.3 Results

### 3.3.1 Modeling aINs, cINs, and MNs

To represent aINs, cINs, and MNs I used a standard HH core ($g_{\mathrm{Na}}/g_{\mathrm{K}}/g_{\mathrm{L}}$) and tuned the leak reversal $E_{\mathrm{L}}$ and applied current $I_{\mathrm{app}}$ to set rheobase and baseline firing. The sodium and potassium peak conductances could also be modified to shape upstroke speed and inter-spike interval. With modest depolarizing current these cells fire repetitively (type-I/II depending on exact kinetics (Fig.3.1). Figure 3.1 shows an example voltage trace of a repetitively-firing HH neuron (a), and the traces of the gating variables (b-e) during the period of this trace. Inhibitory vs. excitatory is implemented at the synapses (glycinergic GABA/glycine for aINs/cINs with $E_{\mathrm{syn}} \approx -70\,\mathrm{mV}$; glutamatergic for premotor excitation), not in the intrinsic HH equations [322, 219].

### 3.3.2 Modeling dINs

Hatchling dINs are ipsilateral glutamatergic premotor neurons that fire a reliable single spike per swim cycle and can both initiate and sustain the swim rhythm; they are electrically coupled and exhibit post-inhibitory rebound (PIR) [215, 217]. To capture their hallmark transient response to constant depolarization, I adapted the HH parameters following the logic used experimentally and in modeling for tadpole dINs [117, 264]. I reduced the delayed rectifier potassium conductance ($g_{\mathrm{K}}$) and enhanced the persistent sodium conductance ($g_{\mathrm{NaP}}$). The leak conductance ($g_{\mathrm{L}}$) and membrane time constant ($\tau_{\mathrm{m}}$) were also adjusted as they contribute to the dINs being more easily depolarised but less able to sustain repetitive spiking. In this configuration, a dIN does not fire repetitively to a maintained $I_{\mathrm{input}}$—it gives a single spike. However, it spikes again after brief release from depolarization, following a transient hyperpolarization, or when it receives a well-timed inhibitory input (from a cIN), due to PIR—exactly as reported in vivo (Fig.3.2). Figure 3.2 shows that the dIN will fire on rebound after a hyperpolarising current when a depolarising +200nA current is interrupted by a

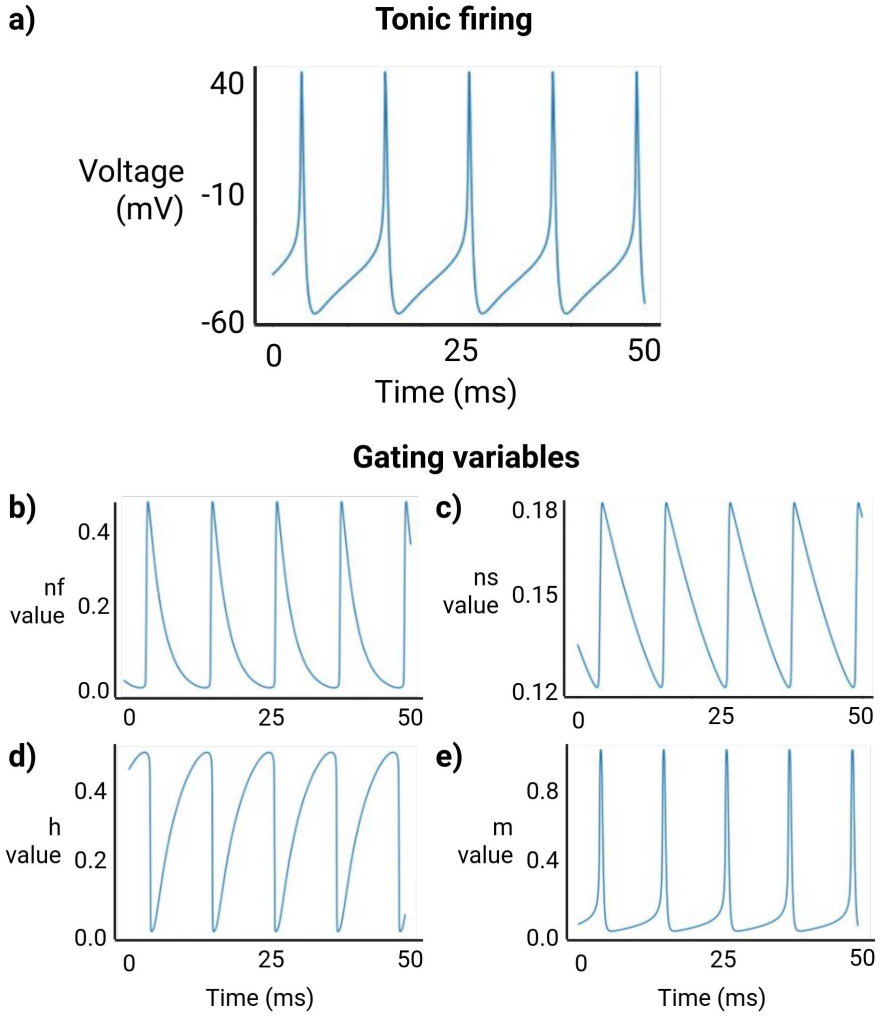


Figure 3.1: Voltage Trace and Gating Variable Evolution during Tonic Firing of a Hodgkin-Huxley Neuron. a) Voltage trace for a tonically-firing Hodgkin-Huxley neuron over a simulation time of 50ms. b) Trace over this period of the: b) nf value, corresponding to the fast potassium channel; c) ns value, corresponding to the slow potassium channel; d) h value and e) m value, both corresponding to the sodium channel.

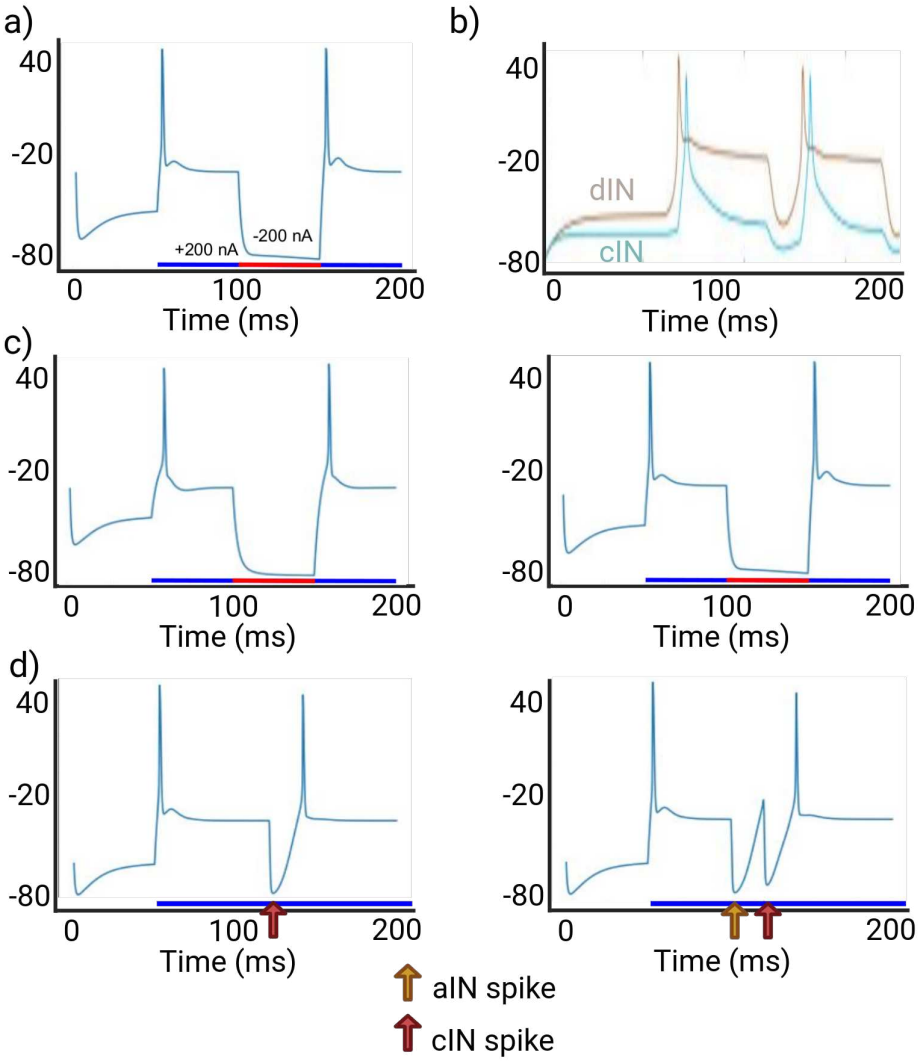


Figure 3.2: a. Voltage Trace from my Hodgkin-Huxley dIN, receiving -200nA between 100 and 150ms and +200nA between 50 and 100ms and 150 and 200ms. b. An example cIN and dIN trace from Ferrario et al., 2018. c. Greater depolarisation leads to a faster rebound spike. On the left the depolarisation in 100nA and the spike times are 57.64ms and 158.87ms. On the right wth 200nA depolarisation, the spike times are 52.85ms and 153.75ms. d. One extra spike from a connected aIN leads to the dIN firing a faster rebound spike. Left: with just one spike from a connected cIN, the dIN spikes at 52.84ms and 141.8ms. Right: with an additional spike from a connected aIN, the dIN spikes at 52.84ms and 140.81ms.

hyperpolarising -200nA current (a), as in [118] (b). A greater depolarisation current and one input spike from an aIN both lead to the rebound spike occurring faster (c-d). [322, 219, 215].

### 3.3.3 Calcium channels and gap junction coupling in dINs

I next tested two dIN specializations, in order to determine their importance in the model dINs: caclium channels and gap junctions.

**Adding calcium conductances.** Introducing a voltage-gated calcium current ($I_{\mathrm{Ca}}$) effectively lowers spike threshold and increases depolarizing drive during the after-spike trajectory. In the model this raised the achievable firing rate under transient inputs and made rebound more robust—consistent with the idea that additional inward currents facilitate reliable cycle-by-cycle recruitment at swim frequencies (Fig.3.3; [264]). Figure 3.3 shows how adding calcium channels to two different neurons increases firing frequency for both.

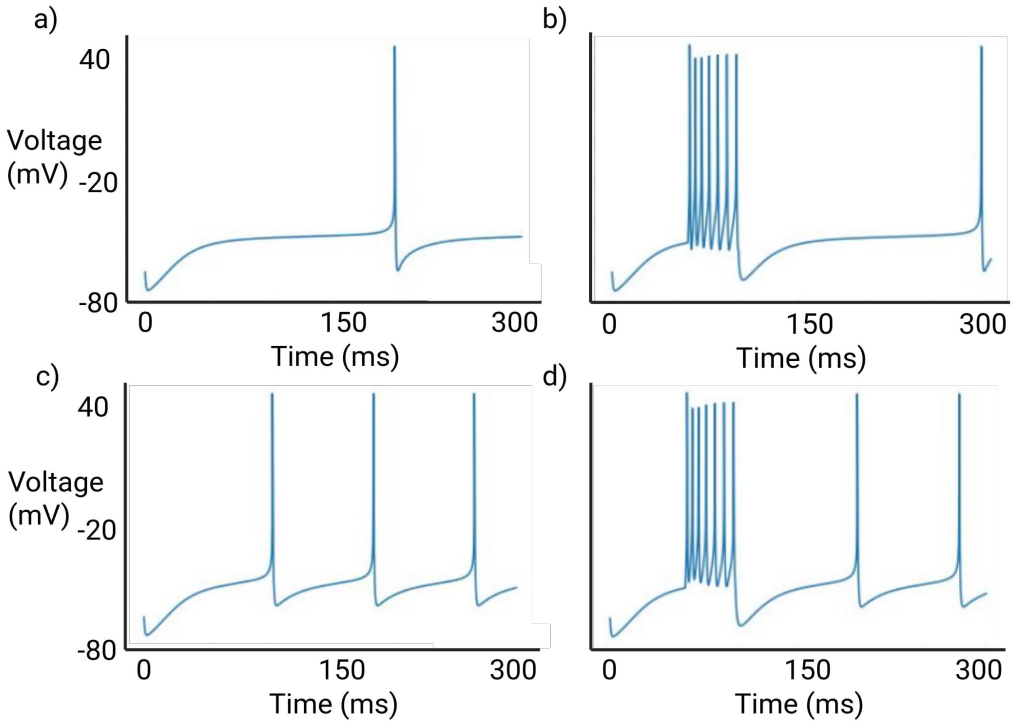


Figure 3.3: Adding calcium channels leads to a higher firing rate in the dIN. a, b) Two different neurons without calcium channels. c, d) The same neurons as in a and b, this time with calcium channels.

**Strengthening dIN–dIN gap junctions.** Increasing $g^{\text{gap}}$ between two dINs produced (i) spikelets and then full action potentials in the *unstimulated* dIN when its partner was driven (Fig.3.4a), (ii) shorter latency in the follower cell (Fig.3.4b), and (iii) higher spike-time correlation (tighter left–right premotor locking) (Fig.3.4c). These effects mirror the role of electrical coupling in stabilizing the dIN population and sustaining locomotor rhythm [215].

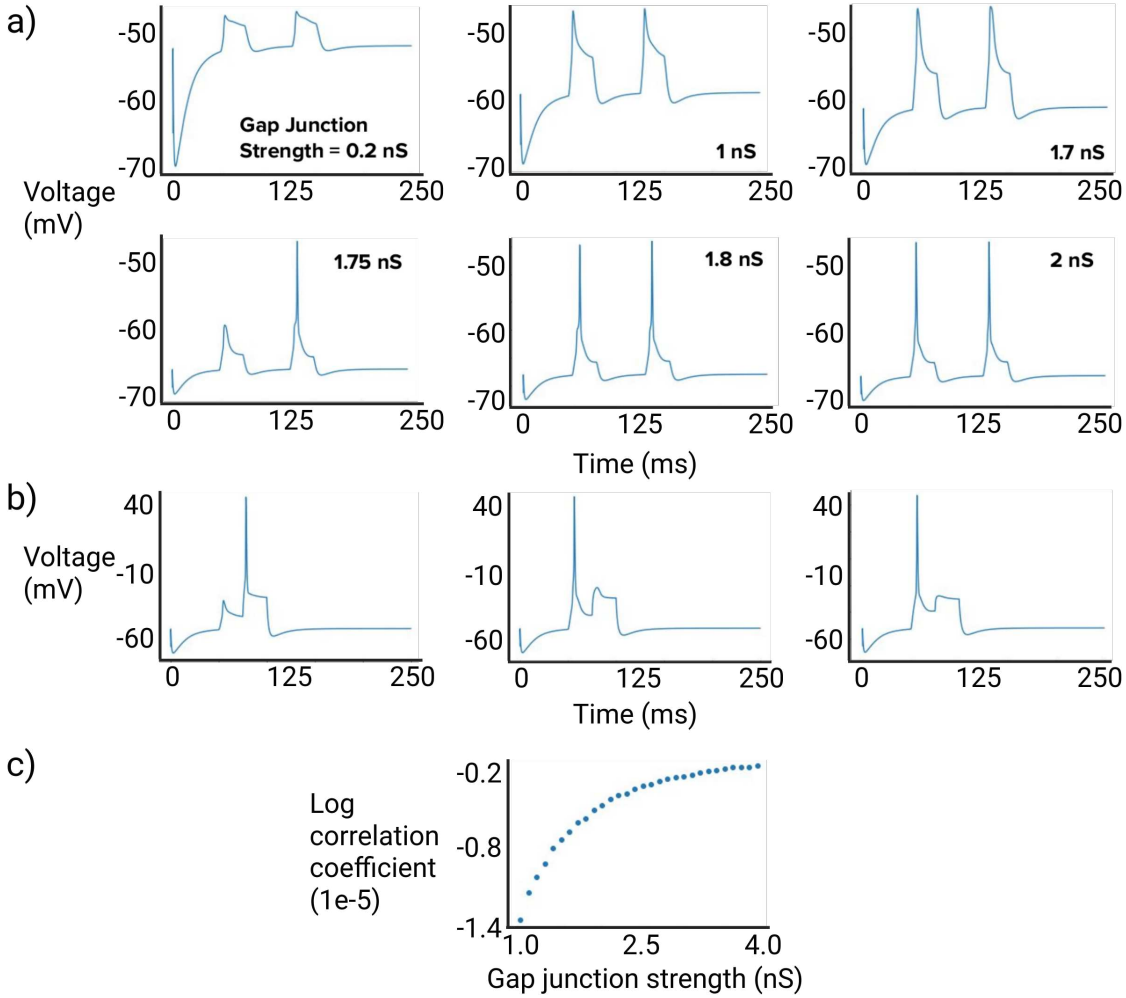


Figure 3.4: Increasing gap junction strength between two dINs leads to: a) action potentials in the unstimulated dIN when the other is stimulated. Top row, from left to right action potential strength is 0.2nS, 1nS and 1.7nS. No action potential is produced. Bottom row, from left to right action potential strength is 1.75nS, 1.8nS and 2nS. At least one action potential is produced. b) Faster action potentials in the unstimulated dIN. From left to right increasing gap junction strength: 1nS, 2nS and 4nS. Action potential time with respect to that in the stimulated neuron from left to right 24.5ms, 1.8ms, 0.6ms. c) Increasing correlation between the two dINs.

### 3.3.4 Assembling an eight-cell bilateral network

With class templates in hand, I assembled a minimal bilateral network: four cells on each side (dIN, aIN, cIN, MN) for a total of eight neurons 3.5. Connectivity followed the canonical tadpole scaffold: dINs and aINs respectively excite and inhibit all neurons on the same side of the spinal cord; cINs inhibit all neurons on the opposite side of the spinal cord; dINs are electrically coupled [272].

Even at this scale the network produced alternating bouts of activity and silence across sides: when one side was active, the other was suppressed; after inhibition decayed and rebound accumulated, roles switched (Fig.3.5). This is the expected half-center logic for left–right alternation in hatchlings [323, 322, 219].

## 3.4 Summary

This HH network met its goal as a proof of concept: it reproduced class-typical excitability (tonic vs. transient), PIR-based dIN recruitment, electrical coupling effects, and emergent left–right alternation. However, HH models are parameter-rich, meaning that further development and analysis of the model, such as adding more neuron classes and performing parameter sweeps, quickly becomes unfeasible. Furthermore, these models exhibit limited robustness. Their large number of parameters must be precisely tuned, and even minor perturbations in these values can cause the model to break down. Since the overarching aim was to establish a tractable platform for hypothesis-driven experiments, I transitioned to a simpler integrate-and-fire (I&F) formulation with far fewer parameters, making it easier to scale the network, add heterogeneity, and perform systematic analysis while retaining the key dynamical mechanisms identified here.

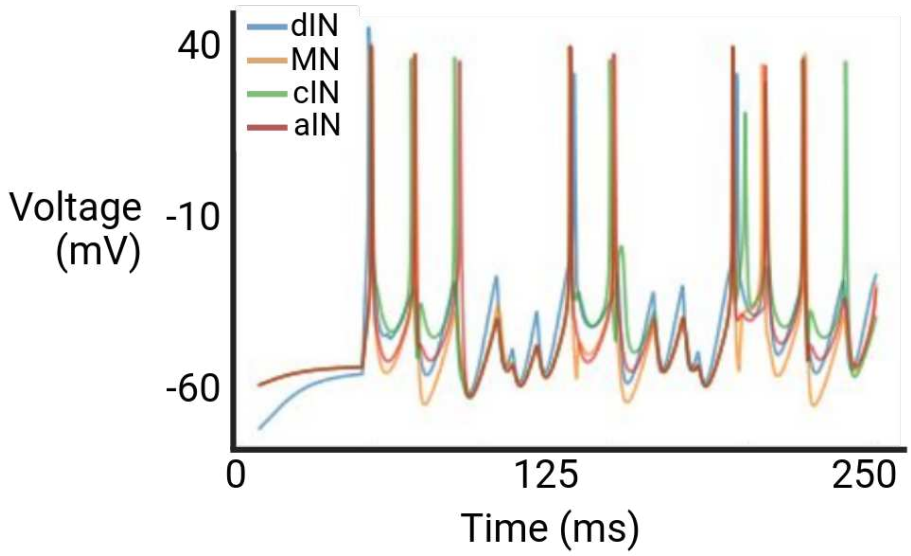


Figure 3.5: Example voltage traces from all four types of CPG neurons from one side of the spinal cord when the neurons are connected. Alternating periods of activity and silence indicate that the two sides are alternating activity like in the spinal cord.

# 4 An Integrate–and–Fire Model of the *Xenopus* Tadpole Spinal Cord

## 4.1 Overview

After establishing a biophysically detailed Hodgkin–Huxley (HH) proof of concept (Chapter 3), I transitioned to a simpler integrate–and–fire (I&F) formulation to enable faster parameter exploration and network scaling while retaining key dynamical properties (tonic firing for aIN/cIN/MN; phasic/rebound for dINs).

This shift is important for both interpretability and experimental alignment. The I&F reduction preserves the causal levers uncovered with HH (e.g., the role of PIR-like reset dynamics, the timing of inhibitory motifs, and the impact of coupling on synchronization) while eliminating many biophysical degrees of freedom that are difficult to constrain experimentally. In practice, this enables efficient exploration of how network structure and a small set of physiologically anchored parameters shape emergent patterns, supports robust fitting to intracellular recordings via simulation-based inference, and facilitates reproducible sensitivity analyses. Crucially, the simplified model lowers the barrier to scaling toward segmental chains and left–right networks at the population level, where questions about alternation stability, frequency control, and rostrocaudal delays can be posed cleanly.

## 4.2 Methods

### 4.2.1 Experimental Targets and Datasets

To constrain model behavior at the appropriate developmental stage (st. 37–42), I used physiological recordings of each class (aIN, cIN, MN, dIN) at the same stage as modeled provided by Wenchang Li from St Andrews (Fig.4.1. Figure4.1 shows a section of a voltage trace from one of each type of CPG neuron. These traces served as targets for single–cell fitting: spike count and timing under step currents, inter–spike interval (ISI) statistics, latency to first spike, after–hyperpolarization (AHP) amplitude, sag/rebound signatures, and (for dINs) the phasic single–spike response to sustained depolarization together with post–inhibitory rebound.

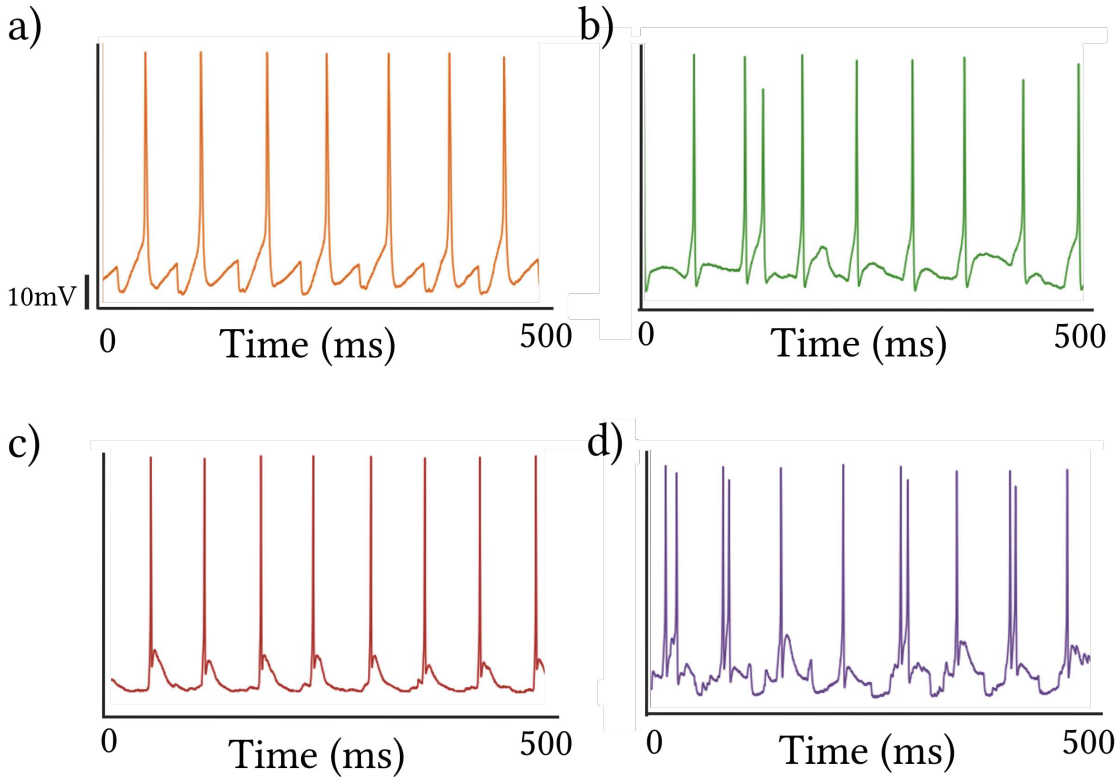


Figure 4.1: Example voltage traces from physiological recordings obtained from Wenchang Li of all four types of CPG neurons in the *Xenopus* tadpole. a) dIN, b) aIN, c) cIN, d) MN. The recordings are made during fictive swimming so the neurons are connected: this is why the dIN also fires repetitively.

### 4.2.2 Neuron Model and Parameterization

I employed a two–dimensional adaptive integrate–and–fire neuron (AdEx–style), which balances interpretability with few parameters:

$$C\,\frac{dV}{dt} = g_L\,(E_L - V) + g_L \Delta_T \exp\!\Big(\frac{V - V_T}{\Delta_T}\Big) - w + I_{\text{app}} + I_{\text{syn}}, \tag{4.1}$$

$$\tau_w\,\frac{dw}{dt} = a\,(V - E_L) - w, \tag{4.2}$$

with spike/reset

$$\text{if } V \geq V_{\text{spike}} \;\Rightarrow\; \begin{cases} V \leftarrow V_r, \\ w \leftarrow w + b. \end{cases}$$

Here $V$ is membrane potential, $w$ an adaptation current; $(a, b, \tau_w)$ control subthreshold and spike–triggered adaptation, $\Delta_T$ sets spike onset sharpness, and $(E_L, V_T, V_r)$ define leak, threshold, and reset. Synaptic/electrical coupling enter via $I_{\text{syn}}$ (see below).

**Class grouping.** I fit a tonic parameter set shared by aINs, cINs, and MNs (distinct synaptic signs/projection patterns; same intrinsic template), and a separate phasic/rebound parameter set for dINs. This reflects experimental observations that aINs/cINs/MNs readily fire repetitively under steady depolarization, whereas dINs are phasic and strongly PIR–enabled [322, 219, 272].

### 4.2.3 Simulation–Based Inference for Parameter Optimisation

To match the model's voltage responses to the experimental traces, I used simulation–based inference (SBI) to appropriately tune $(C, g_L, E_L, V_T, \Delta_T, V_r, a, b, \tau_w)$ per class (Fig.4.2). Fig. 4.2a shows how SBI works: Simulation-based inference (SBI) provides a principled framework for estimating the parameters of mechanistic neuron models without requiring an analytically tractable likelihood function. In our case, SBI is used to tune the parameters of the standard Adaptive Exponential Integrate-and-Fire (AdEx) model so that its simulated output matches biological recordings. The procedure begins by specifying a prior distribution over the model parameters—here a uniform prior over a biologically-realistic range. SBI then samples parameter sets from this prior and runs repeated simulations of the AdEx model, generating synthetic voltage traces and spike patterns. These simulated outputs are compared to experimental data from Wenchang Li, and the algorithm learns a statistical mapping between the high-dimensional parameter space and the observed

physiological features. By iteratively incorporating information from many simulations, SBI produces a posterior distribution describing which parameter combinations are most consistent with the biological recordings. This provides not only a set of well-fitting parameters, but also an explicit quantification of uncertainty and parameter degeneracy—advantages that traditional hand-tuned or gradient-based calibration approaches lack. In doing so, SBI offers a robust and data-driven method for constraining single-neuron models and reducing the fragility associated with classical tuning of Hodgkin–Huxley-type or AdEx-type systems. Once optimised, samples can then be drawn from the posterior distribution and compared to the real data. Fig 4.2b shows an example of an output parameter space, where each row is a cross section through a different dimension, and to the right, an example voltage trace from a sample of parameters taken from the optimised space.

Each candidate parameter vector was simulated under the same current protocols as the recordings; a feature vector summarized the output (spike count, ISI mean/CV, latency, AHP depth, rebound amplitude, voltage RMSE). The loss combined feature deviations and trace error. Separate optimization runs produced the tonic template for the aINs, cIN and MNs, and the phasic/rebound dIN template.

## 4.3 Results

### 4.3.1 Single-cell Optimisation

**Tonic (aIN/cIN/MN).** The optimised tonic neurons had low–to–moderate adaptation $(a, b)$, modest $\Delta_T$, and a reset $V_r$ that supported stable repetitive firing to step currents. With constant $I_{\text{app}}$, the tonic neuron fires regular trains, the firing frequency scales with current, and inhibition pauses but does not qualitatively alter the regime. (Fig.4.3a,c) The metrics for the simulated neuron matched the experimental metrics very well (Fig.4.3b).

**Phasic dIN.** The optimised dINs had stronger subthreshold adaptation $(a)$, higher $b$ and longer $\tau_w$, and a threshold/reset configuration close to a stable fixed point under constant depolarization—yielding a single spike to a sustained step and robust rebound after brief inhibitory episodes. With constant $I_{\text{app}}$, the dIN spikes once and then remains subthreshold (accommodation) (Fig.4.4a). If the current is briefly removed and reapplied, it spikes again (Fig.4.4b). Likewise, a transient inhibitory input (from a cIN) elicits an extra spike via post–inhibitory rebound, consistent with in vivo reports [322, 219, 215] (Fig.4.4d). When a model cIN is connected to a dIN, each cIN spike can prime a subsequent dIN spike, producing multiple dIN spikes over a constant depolarizing background—again matching the qualitative experimental

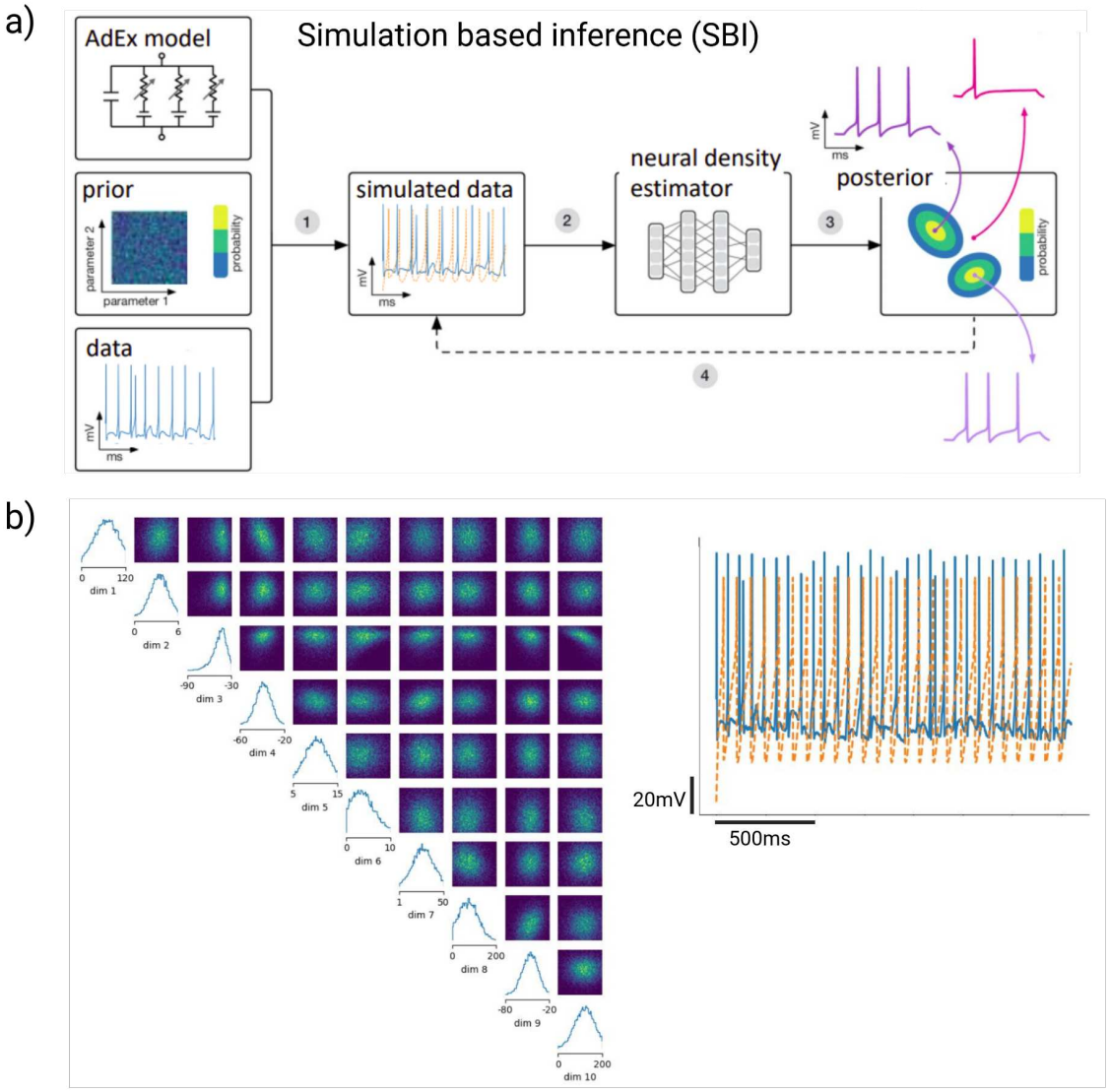


Figure 4.2: a) Diagram illustrating the use of simulation-based inference (SBI) to tune parameters of the AdEx neuron model. SBI draws parameters from a uniform prior, simulates the model, and compares the output to experimental recordings (here from Wenchang Li). By learning the relationship between parameters and observed activity, SBI infers the posterior distribution of parameter values that best reproduce the biological data. b) Optimised parameter space (left) obtained with simulation-based inference for a repetitively spiking AdEx neuron model, and an example simulated voltage trace (right, orange) compared to the corresponding experimental recording (blue). The parameters correspond to $(C, g_L, E_L, V_T, \Delta_T, V_r, a, b, \tau_w)$ and $I_{\text{inp}}$, the input current. The close match between model and data illustrates that the inferred parameters capture the characteristic firing pattern of the neuron.

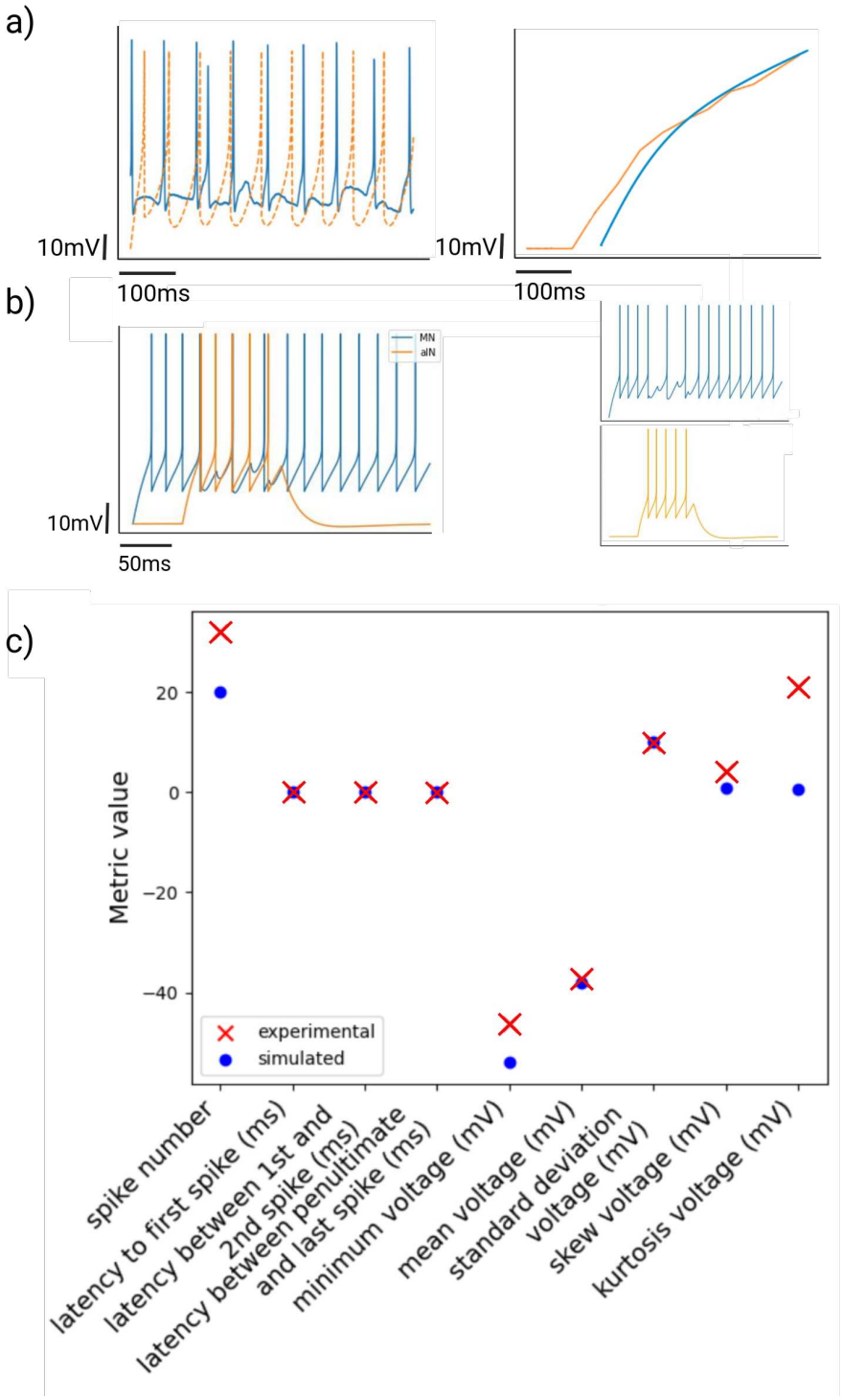


Figure 4.3: a) Voltage trace of an experimental (blue) and simulated (orange) AdEx repetitively-spiking neuron (left) and F-I curve (right). b) Experimental versus simulated metrics for this neuron. c) Connecting an aIN to a MN leads to reduced firing in the MN whilst the aIN is firing. Left: Traces from the MN and aIN plotted together. Right: Just the MN trace (top in blue) and just the aIN trace (bottom in orange).

picture. Again, the metrics for the simulated dIN match the experimental metrics very well (Fig.4.4c).

### 4.3.2 Phase–Plane Tuning of dIN Dynamics

A phase diagram (phase plane) plots the nullclines and trajectories of the two state variables $(V, w)$. For AdEx, the $V$–nullcline is curved due to the exponential term, while the $w$–nullcline is approximately a straight line $w = a\,(V - E_L)$. Their intersection is a fixed point; the reset condition adds a vertical jump (from $V_{\text{spike}}$ to $V_r$) and an increment in $w$.

**Tonic regime.** In the tonic template, the unstable fixed point under depolarization lies between the nullclines, which do not cross; trajectories settle onto a stable limit cycle (periodic spiking) (Fig.4.5a).

**Phasic regime (dIN).** For the dIN, parameters were tuned so that under constant depolarization the fixed point is stable and is at the intersection of the nullclines (Fig.4.5b). A brief perturbation (inhibitory pulse or current removal) lowers the value of $V$ so briefly displaces the state, generating a single spike before trajectories fall back to the stable fixed point. Increasing $b$ (spike–triggered adaptation) and $\tau_w$ prevents immediate re–entry into the spiking loop, enforcing the one–spike response to a sustained step. This "near–threshold stable fixed point + reset geometry" ensured the model could escape the fixed point transiently (to spike) and then be captured again—precisely the phasic behavior required for dINs.

### 4.3.3 Scaling and connectivity

I scaled the network to match the number of cells of each class used by [117] at the same stage and connected populations using their reported weight/probability matrix as a template. With modest tonic drive to dINs and intact cross–inhibition via cINs, the full model produced robust left–right alternating population bursts—i.e., swimming–like activity—without further fine–tuning, in line with the canonical scaffold (Fig.4.6) [272, 323, 322, 219]. To move beyond minimal motifs and toward a circuit that reproduces the tadpole's swim pattern, I scaled the model to the population sizes reported for the same developmental stage and used the connection probabilities and synaptic weights from [117] as a template for all pairwise interactions. I assigned

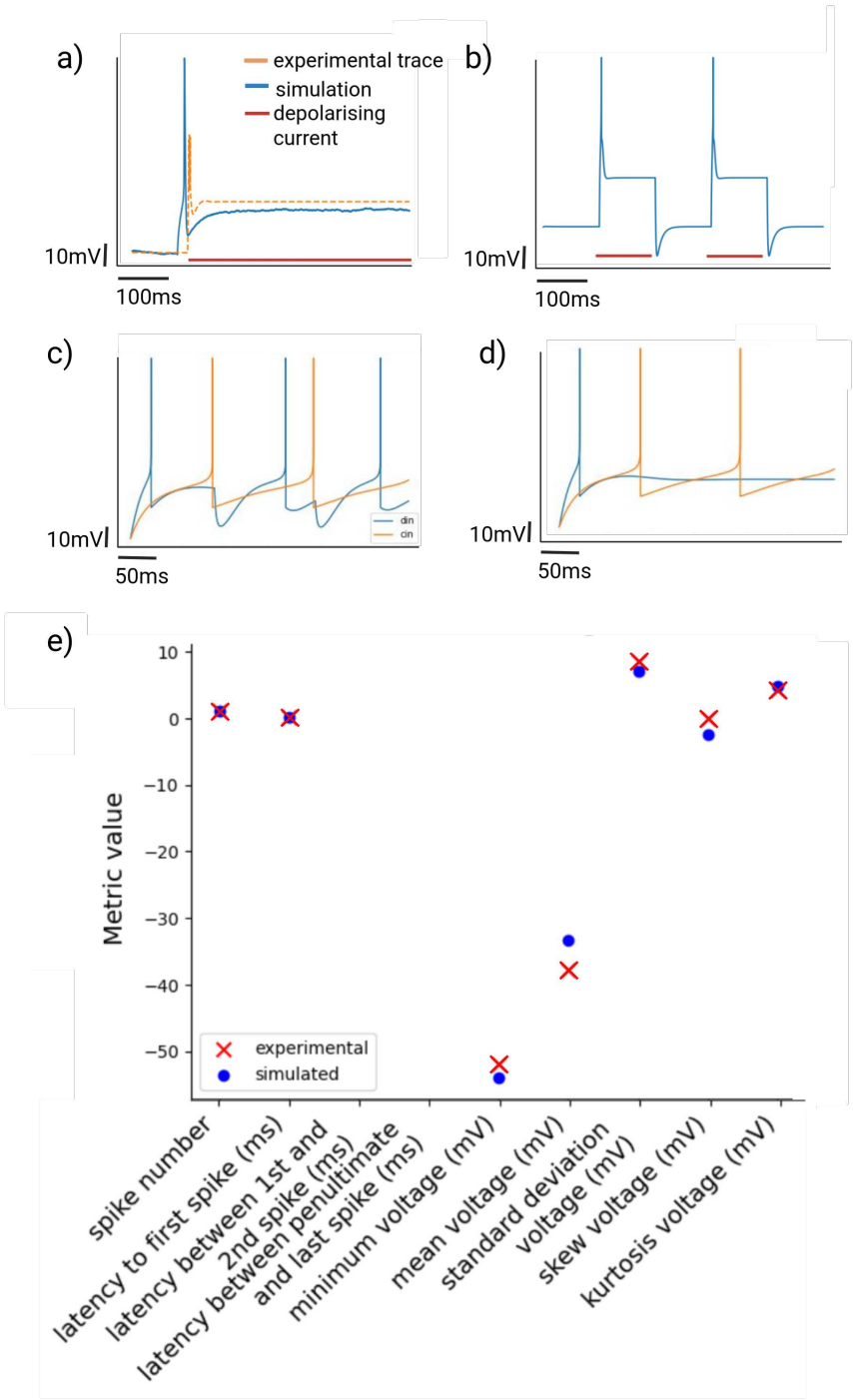


Figure 4.4: a) Example experimental (blue) and simulated AdEx (orange) dIN voltage trace for application of a constant current. The red line shows when current was applied. b) The simulated dIN fires again if a depolarising current is applied between 100-200ms and again between 300-400ms. c. Experimental versus simulated metrics for this neuron. c. cIN activity allows a dIN to spike more than once. Left cIN connected to the dIN versus right unconnected.

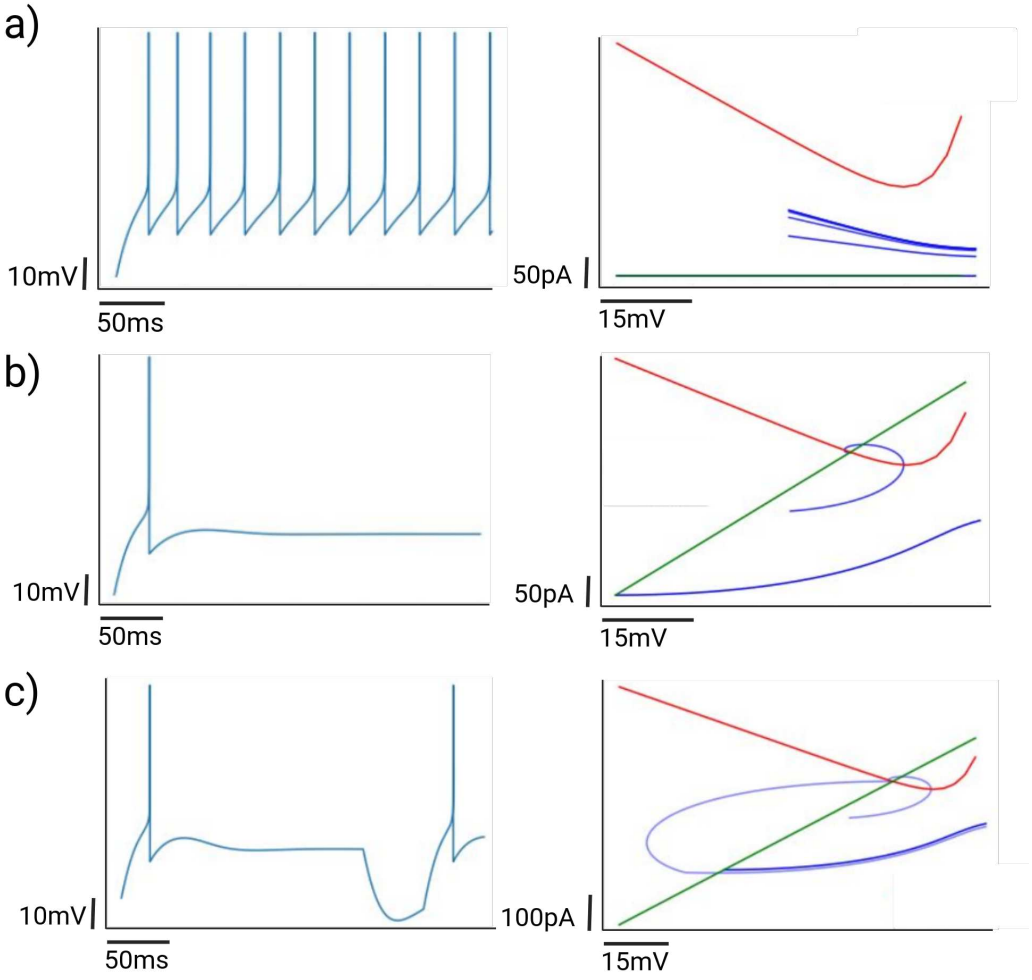


Figure 4.5: a. Voltage trace and phase plane trajectory of a tonically-firing AdEx neuron. b. Voltage trace and phase plane trajectory of a transiently-spiking AdEx neuron. c. A brief input of negative current moves the trajectory away from the fixed point so the neuron can spike again.

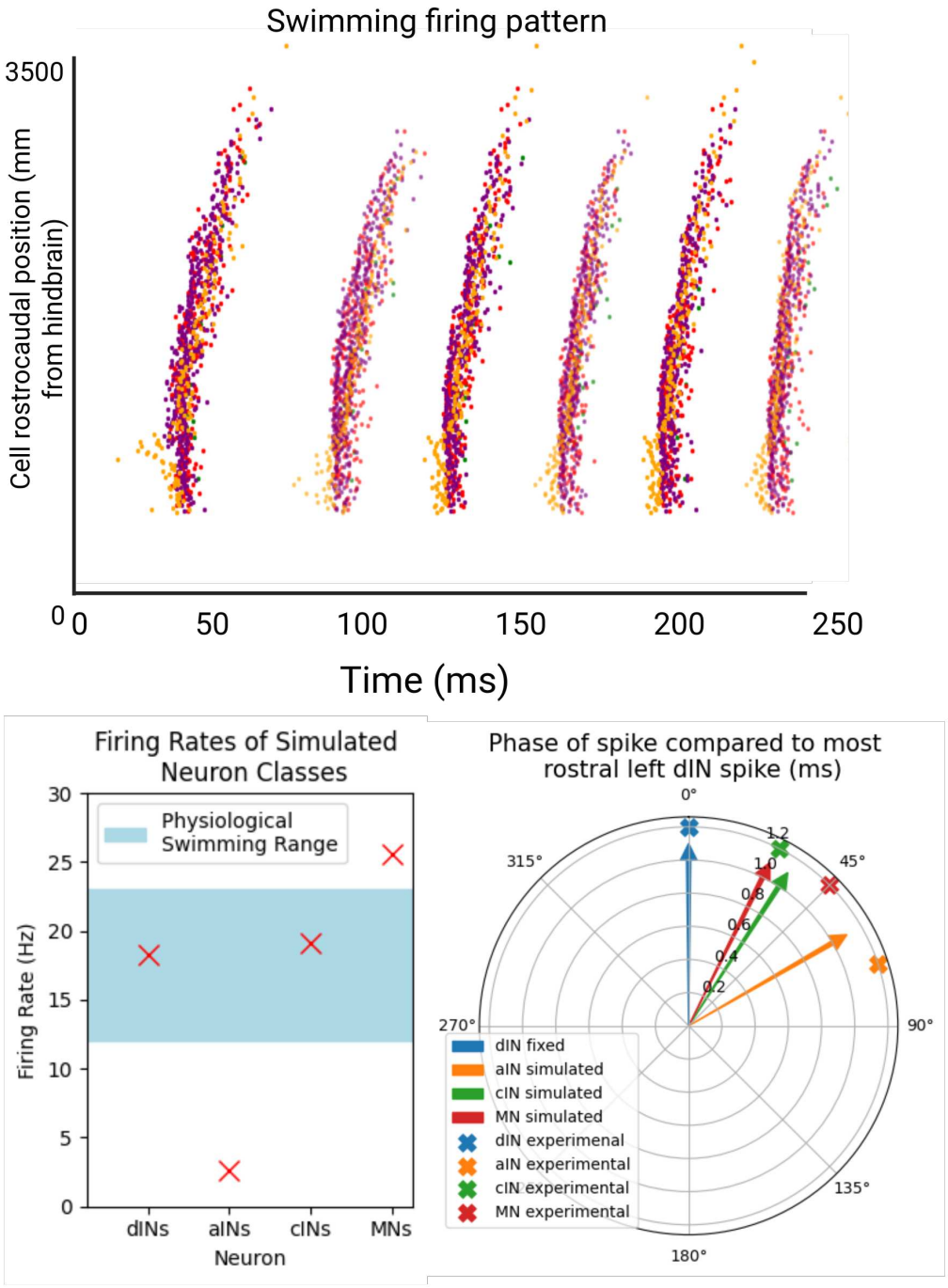


Figure 4.6: Raster plot of the full model network firing in a swimming-like pattern, showing alternating left–right activity and a rostro-caudal delay characteristic of tadpole locomotion. Below, summary metrics quantify this activity and are used to compare the model neuron's firing properties to experimental recordings, allowing assessment of how well the model reproduces key features of tadpole swimming. Left: the physiological firing rate of the CPG neurons is between 12-23Hz, however the aINs are only sparsely recruited during swimming. This is reflected in my model (red crosses). Right: The crosses show the phase of the spike for each CPG neuron in the real tadpole, taking the rostral left dIN spike as 0°. The arrows show where in the swim-cycle phase the simulated neurons spike.

each neuron a position along the rostrocaudal axis and connected the neurons according to [117] so that excitation and inhibition propagate caudally with a slight phase lag, as in fictive swimming. Within a hemicord, dINs project to each other and to MNs, and receive fast, phasic inhibition from aINs. Across the midline, cINs project to the opposing hemicord's dINs and MNs, establishing the mutual inhibition that enforces left–right alternation [323, 322, 219]. The resulting connectivity, when instantiated with the [117] weight and probability matrix, yields a network that mirrors the canonical scaffold for tadpole swimming [272].

With only modest tonic depolarization to dINs and no additional parameter tuning, the integrated network settles into robust alternating population bursts that recruit MNs in antiphase on the two sides, producing a sustained swim-like rhythm (Fig.4.6). The period and duty cycle fall within the range expected for hatchling tadpoles, and the rostrocaudal delay emerges from the connectivity rather than requiring explicit timing control. dINs fire a single spike on each cycle and reliably re-engage following brief inhibitory transients, consistent with post-inhibitory rebound; aINs contribute cycle-phased inhibition that sharpens burst onsets and truncates late spikes; cINs deliver the mid-cycle contralateral inhibition that terminates the ongoing burst and enables the next burst on the opposite side, reproducing the hallmark left–right alternation described experimentally [323, 322, 219]. Importantly, alternation persists over a finite range of tonic drives to dINs and modest variations in synaptic weight, indicating that the solution is structurally stable rather than finely tuned.

### 4.3.4 Examination of the Individual Firing Patterns

At the scaled network level, MNs and cINs occasionally exhibited *two* spikes per cycle (a bimodal ISI distribution around the swim period) (Fig.4.7). This could reflect one or a combination of (i) residual depolarization after the first spike (synaptic tails overlapping with the next cycle), (ii) insufficient adaptation or refractory protection in those classes, and (iii) timing mismatch between decay of contralateral inhibition and renewal of ipsilateral excitation. I tested several remedies—lengthening inhibitory decay, increasing $b$ or shortening $\tau_w$ in MNs/cINs, lowering $V_r$, tightening refractoriness, and slightly weakening recurrent excitation—but none eliminated doublets across the full parameter range while preserving stable alternation.

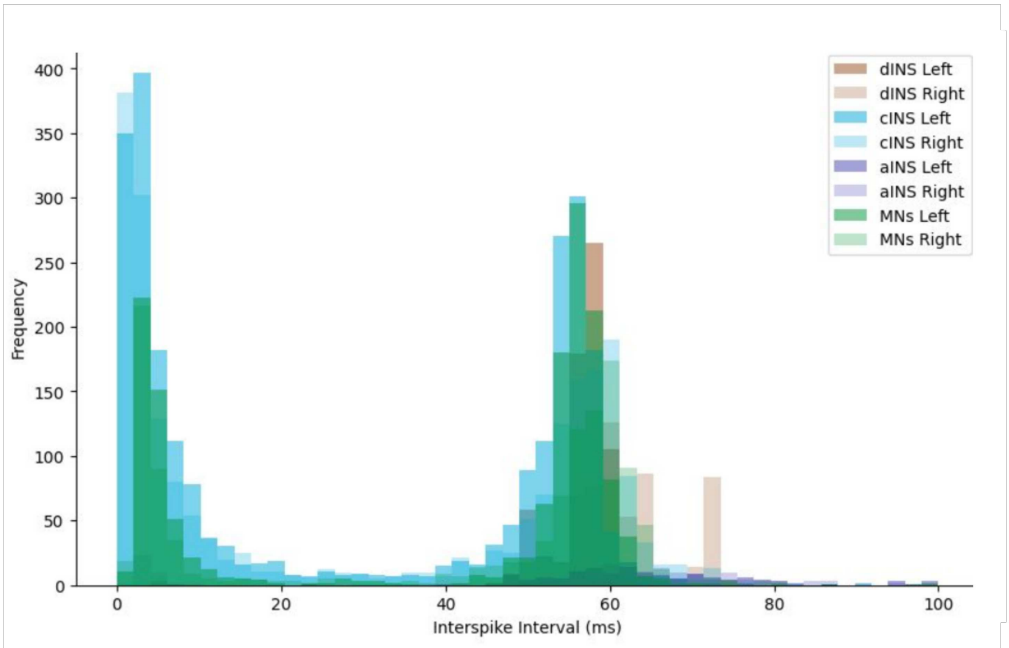


Figure 4.7: A histogram of inter-spike interval distribution shows that the MNs and and cINs are firing more than one spike per swimming cycle, not in line with experimental findings.

## 4.4 Summary

Using a compact AdEx–type I&F formulation fit to stage–matched physiology, I separated the intrinsic dynamics into (i) a tonic template for aIN/cIN/MN and (ii) a phasic/rebound template for dINs. Phase–plane analysis ensured the dIN's single–spike–to–step and PIR behaviors. When scaled and wired with literature–based connectivity, the network produced swimming–like left–right alternation. Residual double–spike artifacts in MNs/cINs were not removed by any adjustments tested.

Together these results indicate that, after scaling cell numbers and adopting empirically guided connectivity from [117], the model expresses the core dynamical motifs of the tadpole spinal CPG: dIN-driven excitation, ipsilateral cycle-phased inhibition, midline commissural inhibition, left–right alternation, and a physiologically plausible rostrocaudal delay. In other words, the network no longer simply produces alternation in principle; it now reproduces the organizational logic of the larval tadpole swim circuit with minimal assumptions and without extensive fine-tuning, providing a platform for systematic perturbations and data-constrained comparisons to the canonical scaffold [272, 323, 322, 219].

# 5 Synfire chains as a minimal model for patterned propagation

## 5.1 Overview

Building on the integrate-and-fire model of the tadpole spinal cord, I next turn to a synfire chain framework to examine how structured feedforward connectivity can generate robust, precisely timed propagating activity. The synfire chain hypothesis proposes that precisely timed volleys of spikes can propagate through layered feedforward networks, forming a reliable substrate for temporal patterning in neural circuits [1]. In current-based leaky integrate-and-fire (LIF) implementations, the fate of an input volley (a "pulse packet") is determined by two key parameters: its **amplitude** (number of coincident spikes) and its **temporal dispersion** (standard deviation of spike times). Diesmann and colleagues demonstrated a sharp boundary in this amplitude–dispersion plane separating self-sustaining propagation from decay [97].

My goal in this chapter was twofold. First, to reproduce the classic synfire-chain results with LIF neurons to create a simple, controllable substrate for patterned activity. Second, to explore how introducing inhibitory coupling between chains yields competitive dynamics reminiscent of central pattern generator (CPG) alternation, and to outline next steps toward a tadpole-like CPG using this framework [274, 219, 215, 117].

## 5.2 Methods

### 5.2.1 Model neurons and pulse packets

Each model neuron was a standard current-based LIF unit,

$$\tau_m \frac{dV}{dt} = -(V - E_L) + RI(t),$$

with instantaneous spike emission at $V \geq V_{\text{th}}$ and reset to $V_r$ followed by a refractory period. Synaptic input $I(t)$ was modeled as the sum of exponentially decaying postsynaptic currents elicited by presynaptic spikes with fixed delays; for a spike at time $t_k$ the synaptic conductance followed

$$g(t) = w\, e^{-\frac{t-t_k}{\tau_s}}\, H(t - t_k),$$

and the current was

$$I(t) = g(t)\,\big(V(t) - E_{\text{rev}}\big),$$

where $H$ is the Heaviside step function, $w$ the synaptic weight, $\tau_s$ the synaptic time constant, and $E_{\text{rev}}$ the reversal potential [135].

Following [97], the input to the first layer was a pulse packet: $N_0$ spikes with times drawn from a normal distribution $\mathcal{N}(t_0, \sigma_0)$, where $N_0$ is the packet amplitude and $\sigma_0$ its temporal spread. Downstream layers were fully (or densely) feedforward connected with identical LIF neurons and homogeneous synapses; all spikes from layer $\ell$ targeted layer $\ell + 1$ with delay $d$ and weight $w$. For each layer, I measured the number of emitted spikes and the standard deviation (jitter) of their times to track whether packets sharpened and amplified (successful propagation) or broadened and attenuated (failure), as in the amplitude–dispersion analysis introduced by [97, 283].

## 5.3 Results

### 5.3.1 Reproducing the synfire boundary

Systematically scanning $(N_0, \sigma_0)$ reproduced the classical transition reported by [97]: tight, high-amplitude packets ($\sigma_0$ small, $N_0$ large) propagated stably across layers, often sharpening; broader or weaker packets decayed within a few layers (Fig.5.2). Plotting the trajectory of each packet in the amplitude–dispersion plane revealed the characteristic flows toward an attractor (robust propagation) or toward extinction (no propagation) [97, 283, 205] (not shown). This validated the implementation and provided a calibrated operating regime for subsequent manipulations.

### 5.3.2 Adding inhibition within layers

To move into the direction of building a spinal cord model, I converted 20% of neurons in each layer to inhibitory units, keeping the projection pattern the same so that 20% of the connections were inhibitory. Under otherwise unchanged parameters, this fraction of inhibition typically abolished propagation: inhibitory feedback broadened packets and reduced effective drive to the next layer so that amplitude fell below the synfire threshold (Fig.5.3). This is consistent with the general result that inhibition (without compensatory adjustments) disrupts the synchrony required for synfire transmission [205]. Restoring propagation required either strengthening $w$, tightening the input packet, or partially segregating inhibitory targets to reduce timing jitter.

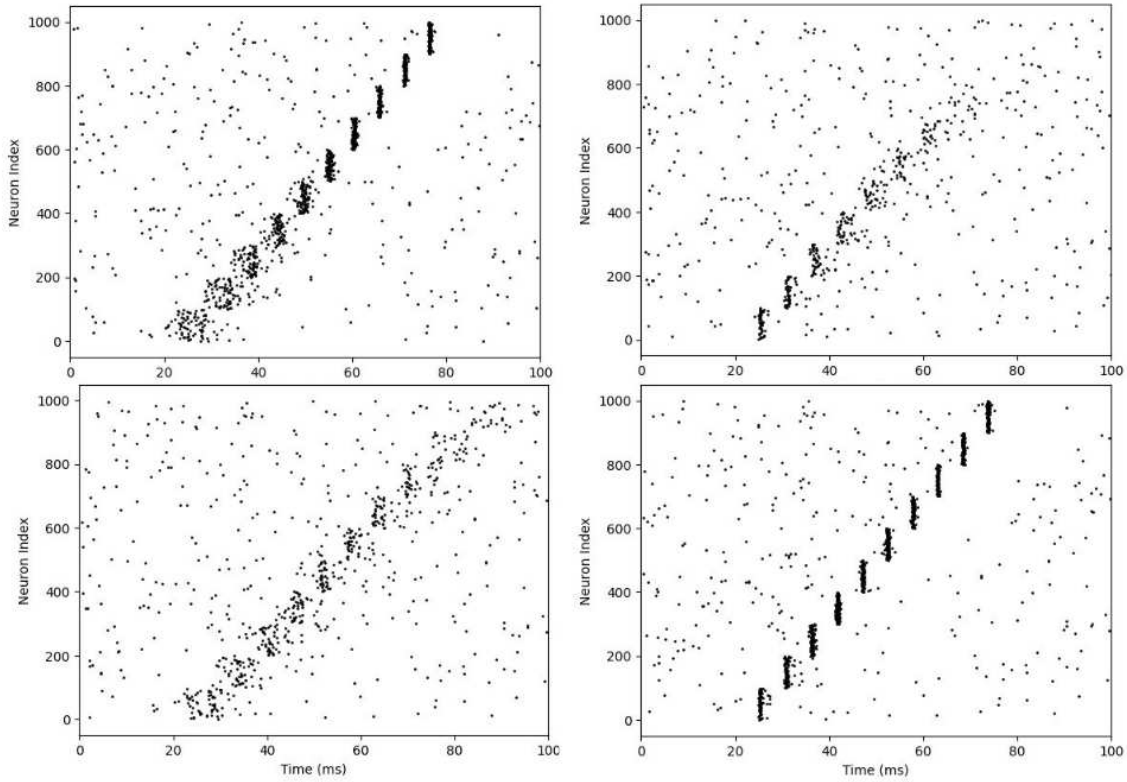


Figure 5.1: Varying the number of input spikes (a) and standard deviation of the input spike packet (sigma) determines whether the spikes propagate to the end of the chain. Clockwise from top left: sigma=3, a=85; sigma=0, a=45; sigma=3, a=70; sigma=0, a=60.

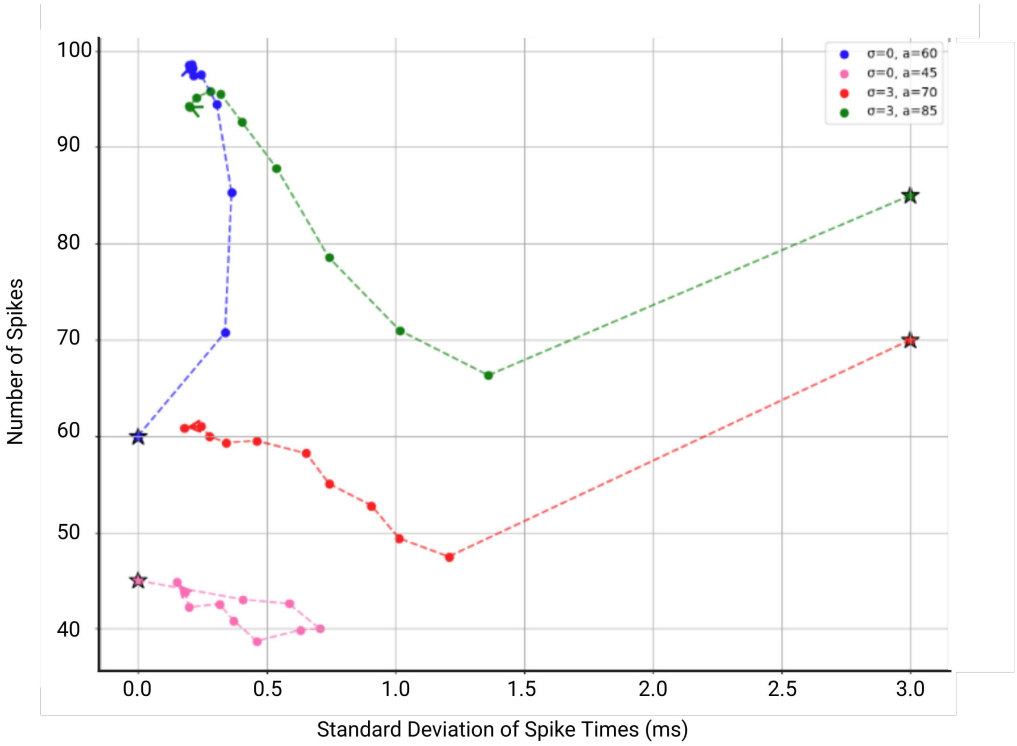


Figure 5.2: This plot shows the evolution and fate of spike packets with different starting values of the number of spikes in a packet and the standard deviation ($\sigma$) of the spike times. With fewer spikes, the standard deviation must be lower for the spikes to successfully propagate through the entire network. The colours correspond to the different starting configurations represented in the previous figure. Starting values of $\sigma$ and number of spikes (a) are as follows: blue trace $\sigma$=0 and a=60; pink trace $\sigma$=0 and a=45; red trace $\sigma$=3 and a=70; green trace $\sigma$=3 and a=85. Only the spikes from the blue and green starting configurations propagate successfully whilst those of the red and pink dye out (see previous figure).

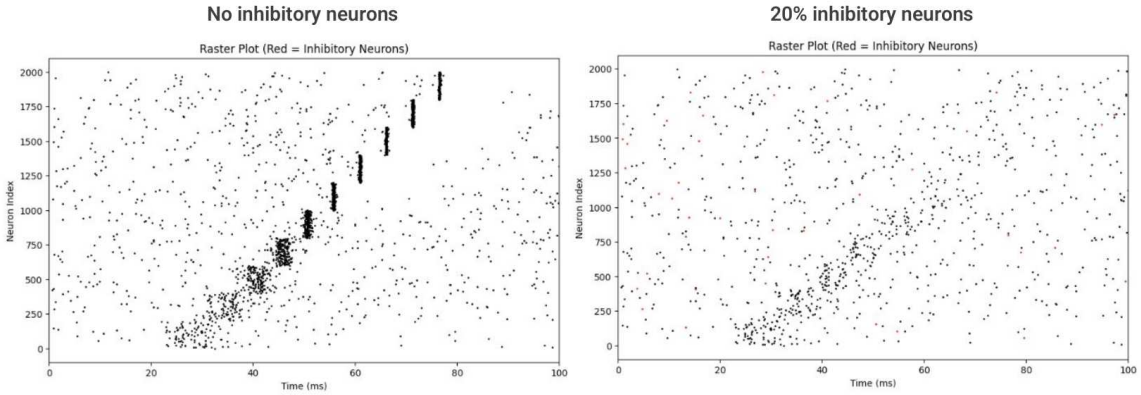


Figure 5.3: Adding 20% inhibitory neurons stops propagation.

### 5.3.3 Two chains with inhibitory cross-coupling

I next instantiated two parallel synfire chains (Chain 1, Chain 2), each composed solely of excitatory LIF layers (no within-layer inhibition, which would represent the aINs, for simplicity), and coupled them via cross-inhibitory projections between corresponding layers (Chain 1 layer $k$ inhibiting Chain 2 layer $k$, and vice versa). When I drove the two chains with matched pulse packets that were offset in time by $\sim$5 ms (Chain 2 lagging Chain 1), the network produced alternating packets across chains: Group 1 in Chain 1 fired, briefly suppressed the homologous group in Chain 2; as inhibition decayed, Group 1 in Chain 2 fired; the same alternation repeated for Group 2, and so on (Fig.5.4). The result was a left–right-like sequence of volleys (*1L, 1R, 2L, 2R,* ...) emerging from mutual inhibition and a small input phase offset—an abstract analog of half-center alternation often invoked for spinal CPGs [51, 274]. Adjusting cross-inhibitory strength tuned the depth and stability of alternation, while too-strong inhibition could quench both chains.

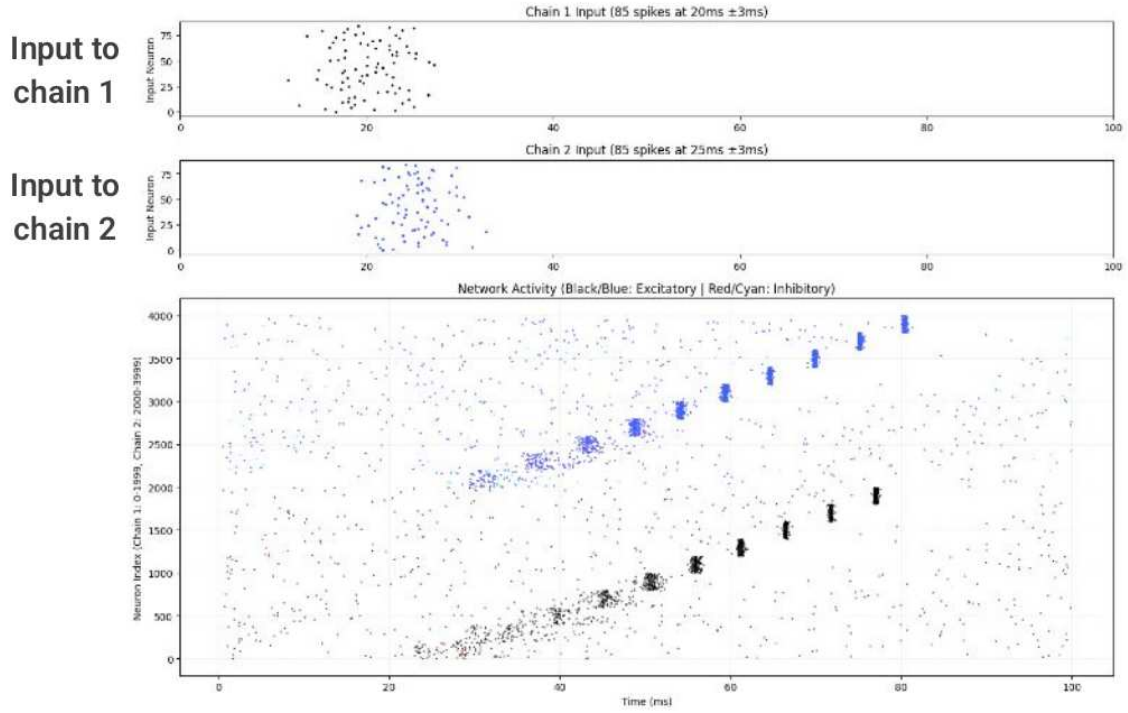


Figure 5.4: When two chains are connected with inhibitory cross-chain connections, two waves of activity can propagate at different times if one chain is given a slightly delayed (+5ms, both a=85 and $\sigma$=3) input to the other chain.

## 5.4 Summary

In summary, I reproduced synfire propagation with LIF neurons and verified the dependence on input amplitude and dispersion [97]. Adding even modest within-layer

inhibition disrupted propagation without compensatory adjustments. Two chains coupled by reciprocal inhibition yielded robust alternation when driven with slightly offset inputs, offering a simple route to left–right CPG-like dynamics. Extending this architecture with segmental modules, commissural and ipsilateral inhibition, and calibrated delays provides a clear roadmap toward a tadpole-inspired CPG built from synfire elements [274, 219, 215, 117].

This progression is important for two reasons. First, it establishes that precise spike timing, rather than detailed biophysics, can be sufficient to generate key organizational principles of spinal pattern generation: reliable feedforward transmission, competition via inhibition, and robust alternation. By isolating the control parameters of synfire dynamics—packet amplitude and temporal dispersion—alongside a small set of inhibitory motifs, the model clarifies which features are necessary for propagation and alternation and which are merely descriptive. Second, the synfire scaffolding offers a tractable bridge between abstract timing-based theories and data-driven vertebrate CPG models. Because the architecture is modular, one can incrementally incorporate biologically grounded elements—segmental delays, commissural pathways enforcing mid-cycle inhibition, and ipsilateral in-cycle shaping—while retaining analytical leverage over packet evolution and stability. This makes it possible to pose testable hypotheses about how changes in synchrony, inhibitory strength, or conduction delays shift a network between propagation failure, stable alternation, and pathological co-activation. In practical terms, the framework provides a compact, computationally efficient substrate on which to map the canonical *Xenopus* motifs and to explore how development, neuromodulation, or mutation might tune timing and inhibition to preserve swimming-like output [274, 219, 215, 117].

## 5.5 Outlook

To move this synfire framework toward the functional behavior of the *Xenopus* tadpole spinal CPG [274, 219, 215, 117]—while retaining LIF-type simplicity—next steps would include the following:

**Map synfire groups to segmental modules.** Represent each axial segment (or short segmental block) by one feedforward group per hemicord. Stagger layer-to-layer delays to reproduce the rostrocaudal phase lag observed during swimming [346, 274].

**Half-center architecture via cross inhibition.** Match the commissural inhibition (cIN-like) to the cross-coupling between left/right groups at the same segmental level of real tadpoles, with IPSC kinetics calibrated to mid-cycle inhibition in tadpole circuits [322, 219].

**In-cycle ipsilateral inhibition (aIN-like).** Add fast, in-phase ipsilateral inhibition onto excitatory groups and motoneurons to regulate cycle period and sharpen packets, in line with the aIN role [219, 274].

**dIN-like excitatory drive within a synfire layer.** Keep excitatory groups tightly synchronized (the "pulse packet" is the dIN volley), optionally assisted by mild gap-junction-inspired synchrony (implemented as brief common input or fast shared conductance) to mimic dIN electrical coupling [215, 264].

**Speed control.** Modulate excitatory weight $w$, tonic bias, or packet dispersion to scale cycle frequency; compare with experimentally observed frequency changes [274, 117].

**Motor readout.** Include motoneuron groups as the terminal layer on each side; ensure phasic, one-spike-per-cycle firing and left–right alternation consistent with fictive swimming [278, 274].

**Validation against data.** Quantify inter-burst interval distributions, phase relations, and rostrocaudal delays; compare with *in vitro* recordings [274, 117].

This plan preserves the conceptual clarity of synfire propagation (timing set by packet synchrony and delays) while incorporating the minimal inhibitory motifs (ipsilateral and commissural) known to be critical for tadpole swimming. It thus complements more biophysically detailed models by offering a tractable mesoscopic description of pattern generation driven by spike timing [97, 205].

# 6 Conclusion

This thesis set out to examine how different modeling levels—ranging from biophysically detailed Hodgkin–Huxley (HH) neurons to compact integrate–and–fire (I&F) abstractions, including synfire modules, can help us to understand how left–right alternation is generated in the *Xenopus laevis* tadpole spinal cord. The guiding idea throughout was that a small set of neurons can reproduce the core features of fictive swimming, and that moving between model scales helps to separate essentials from details. The aim was to create a model that is amenable to analysis and further development.

The first modeling section (Chapter 3) partly reproduced the existent HH model, but with only the four CPG neuron classes: ipsilateral excitatory dINs that provide premotor drive, commissural inhibitory cINs that enforce reciprocal inhibition and mid–cycle IPSPs, ipsilateral inhibitory aINs that deliver in–phase feedback and sensory gating, and motoneurons as readouts of premotor activity. The aim of this part was to create a comparison to the I&F models, to be able to pinpoint what level of detail is necessary to create the key properties of the tadpole spinal cord. These include dIN rebound spiking and the necessity of contralateral inhibition for rhythm maintenance.

With this physiological context, the HH network provided a biophysical proof–of–concept. Class templates were tuned so that aINs, cINs and MNs expressed tonic firing, whereas dINs expressed phasic, rebound–prone excitability. Introducing a low–threshold inward drive and appropriate inactivation kinetics yielded dINs that spiked once to sustained depolarization and fired in rebound after brief hyperpolarization. Adding calcium conductances increased dIN responsiveness, and strengthening gap junctions promoted spikelets, shortened latencies and tighter spike–time locking. When assembled into a minimal bilateral circuit, even a small network (four cells per side) exhibited alternating bouts of activity and silence: the active side suppressed its counterpart; as inhibition decayed and intrinsic rebound accumulated, activity switched sides. This confirmed that canonical half–center logic can emerge from realistic ionic mechanisms, and motivated the transition to simpler models for scaling, parameter exploration and systematic validation.

Replacing HH neurons with adaptive I&F formulations preserved the essential dynamics while enabling efficient inference and analysis. Two AdEx templates—one tonic (shared by aIN/cIN/MN) and one phasic/rebound (dIN)—were calibrated against stage–matched recordings using simulation–based inference so that voltage traces, firing thresholds and adaptation envelopes matched experimental data. Phase–plane

analysis clarified how a near–threshold fixed point together with spike–triggered adaptation enforces the "single–spike–to–step plus rebound" signature of dINs, whereas the tonic template produced regular trains with realistic f–I scaling. At network scale, the I&F model reproduced robust left–right alternation with dIN–driven premotor timing. The bimodal firing rate distribution of MNs/cINs—highlighted where synaptic kinetics, refractory protection or adaptation envelopes may require further refinement to ensure clean one–spike–per–cycle responses without compromising alternation.

The synfire–chain approach provided a complementary, timing–first perspective. Reproducing the classical amplitude–dispersion boundary with current–based LIF neurons showed that the fate of a pulse packet is jointly determined by its amplitude and temporal spread: tight, high–amplitude packets propagate across layers, whereas broad or weak packets decay. Introducing even a small proportion of within–layer inhibition typically abolished propagation by desynchronizing volleys and reducing effective drive. Two excitatory chains coupled by reciprocal inhibition produced alternating sequences when driven with slightly offset inputs, yielding an abstract left–right pattern (*1L, 1R, 2L, 2R*, ...). These results demonstrate that precise spike timing, synaptic delays and minimal inhibitory motifs can generate structured alternation in a feedforward setting, offering a tractable analogy to spinal half–center dynamics.

Together, these strands contribute a unifying narrative. The HH models provide a comparison to the simpler I&F models, and enable us to identify whether any important biological properties are missing from the I&F models. The bottom-up AdEx models show that these mechanisms can be captured with a small set of interpretable parameters that support fast fitting and large–scale exploration. The top-down synfire approach reveals how synchrony and delay can serve as control dials for spatiotemporal patterning. The inter–model agreement increases confidence that the core ingredients—rebound–prone ipsilateral excitation, precisely timed contralateral inhibition, modest electrical coupling and appropriate synaptic delays—are sufficient to account for the essential features of tadpole swimming.

There are, however, limitations to be considered going forwards. Compressing rich ionic mechanisms into effective I&F parameters inevitably leaves some biophysical nuances—such as calcium–dependent resonance or Ih–mediated sag—outside the model's direct control. The synaptic descriptions were intentionally simple and homogeneous, whereas experimental data indicate meaningful diversity in inhibitory kinetics, excitatory components and rostrocaudal delay gradients. The observed double–spike episodes in scaled I&F networks indicate interactions among refractory windows, adaptation and lingering synaptic currents that merit more precise calibration. It remains to be seen whether the same issue would be encountered in the synfire approach when aINs and MNs are added.

Future work should incorporate data–constrained synaptic waveforms and segment–specific delays to reproduce measured phase lags; reintroduce explicit electrical coupling among dINs in the AdEx framework to test rhythm robustness under heterogeneity; and modularize the network segmentally so that left/right units at each level interact via commissural inhibition and in–phase ipsilateral feedback. Extending the models with simple forms of plasticity or short–term dynamics may help stabilize alternation across frequencies, linking development, sensory modulation and adaptation. Finally, quantitative comparisons to *in vitro* recordings of inter–burst intervals, phase relations and rostrocaudal propagation will be essential for closing the loop between model predictions and experiment.

In conclusion, this thesis shows that a small, coherent set of mechanisms can generate reliable left–right alternation in the tadpole spinal cord and that these mechanisms can be expressed at multiple levels of abstraction without losing their functional essence. By moving fluidly between detailed HH models, bottom-up AdEx models and top-down synfire models, the work provides both mechanistic insight and practical tools. The proposed extensions position this framework as a fast, testable substrate for future, data–driven hypotheses about how spinal circuits coordinate alternating patterns, how they remain robust to perturbations and how their computations evolve as locomotion grows in complexity during development.

# Bibliography


[1] Moshe Abeles. *Corticonics: Neural Circuits of the Cerebral Cortex*. Cambridge, UK: Cambridge University Press, 1991. ISBN: 9780521402130.

[2] Victoria E Abraira et al. "The Cellular and Synaptic Architecture of the Mechanosensory Dorsal Horn". In: *Cell* 168.1 (Jan. 2017), 295–310.e19. ISSN: 0092-8674. DOI: 10.1016/j.cell.2016.12.010. URL: https://doi.org/10.1016/j.cell.2016.12.010.

[3] Victoria E. Abraira and David D. Ginty. "The Sensory Neurons of Touch". In: *Neuron* 79.4 (2013), pp. 618–639. DOI: 10.1016/j.neuron.2013.07.051.

[4] Turgay Akay et al. "Degradation of mouse locomotor pattern in the absence of proprioceptive sensory feedback". In: *Proceedings of the National Academy of Sciences of the United States of America* 111.47 (2014), pp. 16877–16882. DOI: 10.1073/pnas.1419045111. URL: https://doi.org/10.1073/pnas.1419045111.

[5] Abdulrahman Al-Mosawie, James M. Wilson, and Robert M. Brownstone. "Heterogeneity of V2-derived interneurons in the adult mouse spinal cord". In: *European Journal of Neuroscience* 26.11 (2007), pp. 3003–3015. DOI: 10.1111/j.1460-9568.2007.05907.x.

[6] William A. Alaynick, Thomas M. Jessell, and Samuel L. Pfaff. "SnapShot: Spinal Cord Development". In: *Cell* 146.1 (2011), 178–178.e1. DOI: 10.1016/j.cell.2011.06.037.

[7] Simon Alford and Terrence L. Williams. "Endogenous activation of glycine and NMDA receptors in lamprey spinal cord during fictive locomotion". In: *Journal of Neuroscience* 9 (1989), pp. 2792–2800. DOI: 10.1523/JNEUROSCI.09-08-02792.1989.

[8] Mohammed R. Alkaslasi, Andrei Picu, and Niccolò Zampieri. "Motor neuron subtype diversification". In: *Development* 148.12 (2021), dev199734. DOI: 10.1242/dev.199734.

[9] Francisco J. Alvarez et al. "Postnatal phenotype and localization of spinal cord V1 derived interneurons". In: *Journal of Comparative Neurology* 493.2 (2005), pp. 177–192. DOI: 10.1002/cne.20711.

[10] Konstantinos Ampatzis et al. "Pattern of spinal V2a neuron activation controls locomotor speed". In: *Neuron* 83 (2014), pp. 692–707. DOI: 10.1016/j.neuron.2014.06.032.

[11] Leif S. Andersson et al. “Mutations in DMRT3 affect locomotion in horses and spinal circuit function in mice”. In: *Nature* 488 (2012), pp. 642–646. DOI: 10.1038/nature11399.

[12] Ove Andersson and Sten Grillner. “Peripheral Control of the Cat’s Step Cycle. II. Entrainment of the Central Pattern Generators for Locomotion by Sinusoidal Hip Movements during “Fictive Locomotion””. In: *Acta Physiologica Scandinavica* 118.3 (1983), pp. 229–239. DOI: 10.1111/j.1748-1716.1983.tb07267.x.

[13] Futoshi Aoki, Thierry Wannier, and Sten Grillner. “Slow dorsal–ventral rhythm generator in the lamprey spinal cord”. In: *Journal of Neurophysiology* 85.1 (2001), pp. 211–218. DOI: 10.1152/jn.2001.85.1.211.

[14] Silvia Arber et al. “Requirement for the homeobox gene Hb9 in the consolidation of motor neuron identity”. In: *Neuron* 23 (1999), pp. 659–674. DOI: 10.1016/S0896-6273(01)80026-X.

[15] Bart N. Armbruster et al. “Evolving the Lock to Fit the Key to Create a Family of G Protein–Coupled Receptors Potently Activated by an Inert Ligand”. In: *PNAS* 104.12 (2007), pp. 5163–5168. DOI: 10.1073/pnas.0700293104.

[16] François Auclair et al. “The Neural Control of Movement”. In: *The Neural Control of Movement*. Ed. by Patrick J. Whelan and Simon A. Sharples. Academic Press, 2020. ISBN: 978-0-12-816477-8.

[17] Jessica Ausborn, Riyadh Mahmood, and Abdeljabbar El Manira. “Decoding the rules of recruitment of excitatory interneurons in the adult zebrafish locomotor network”. In: *Proceedings of the National Academy of Sciences of the United States of America* 109.52 (2012). ISSN: 00278424. DOI: 10.1073/pnas.1216256110.

[18] Oshri Avraham et al. “Transcriptional Control of Axonal Guidance and Sorting in Dorsal Interneurons by the Lim-HD Proteins Lhx9 and Lhx1”. In: *Neural Development* 4 (2009), p. 21. DOI: 10.1186/1749-8104-4-21.

[19] Julien Bacqué-Cazenave et al. “Temporal relationship of ocular and tail segmental movements underlying locomotor-induced gaze stabilization during undulatory swimming in larval *Xenopus*”. In: *Frontiers in Neural Circuits* 12 (2018), p. 95. DOI: 10.3389/fncir.2018.00095.

[20] Myoung-Ha Baek et al. “Spinal Motor Neurons Regulate Locomotor Rhythm via Retrograde Glutamate Signaling”. In: *Neuron* 96.5 (2017), 1271–1281.e3. DOI: 10.1016/j.neuron.2017.10.029.

[21] S. M. L. Barabino et al. “Inactivation of the zebrafish homologue of Chx10 by antisense oligonucleotides causes eye malformations similar to the ocular retardation phenotype”. In: *Mechanisms of Development* 63 (1997), pp. 133–143. DOI: 10.1016/S0925-4773(97)00036-1.

[22] Mary F. Batista, Jennifer Jacobstein, and Karen E. Lewis. “Zebrafish V2 cells develop into excitatory CiD and Notch-signalling-dependent inhibitory VeLD interneurons”. In: *Developmental Biology* 322.2 (2008), pp. 263–275. DOI: 10.1016/j.ydbio.2008.07.015.

[23] Antonio Benito-Gonzalez and Francisco J. Alvarez. “Renshaw cells and Ia inhibitory interneurons are generated at different times from p1 progenitors and differentiate shortly after exiting the cell cycle”. In: *Journal of Neuroscience* 32.32 (2012), pp. 1156–1170. DOI: 10.1523/JNEUROSCI.3630-12.2012.

[24] Eva M. Berg et al. “Principles Governing Locomotion in Vertebrates: Lessons from Zebrafish”. In: *Frontiers in Neural Circuits* 12 (2018), p. 116. DOI: 10.3389/fncir.2018.00116.

[25] Ari Berkowitz. “Physiology and Morphology of Neurons in the Turtle Spinal Cord Active during Swimming”. In: *Journal of Comparative Physiology A* 188.4 (2002), pp. 255–273. DOI: 10.1007/s00359-002-0302-x.

[26] Ari Berkowitz. “Spinal Interneurons that Are Selectively Activated during Fictive Locomotion in the Turtle”. In: *Journal of Neuroscience* 27.21 (2007), pp. 5425–5432. DOI: 10.1523/JNEUROSCI.4746-06.2007.

[27] Ari Berkowitz. “Spinal Interneurons that Are Selectively Activated during Scratching but Not Swimming”. In: *Journal of Neurophysiology* 99.5 (2008), pp. 2887–2897. DOI: 10.1152/jn.01381.2007.

[28] Ari Berkowitz. “Spinal Interneurons that Are Selectively Active during Fictive Scratching Movements in the Turtle”. In: *Journal of Neuroscience* 25.23 (2005), pp. 5850–5861. DOI: 10.1523/JNEUROSCI.0912-05.2005.

[29] Rüdiger R. Bernhardt, Nhung Nguyen, and John Y. Kuwada. “Growth cone guidance by floor plate cells in the spinal cord of zebrafish embryos”. In: *Neuron* 8.5 (1992), pp. 869–882. DOI: 10.1016/0896-6273(92)90201-N.

[30] J. N. Betley et al. “Stringent specificity in the construction of a GABAergic presynaptic inhibitory circuit”. In: *Cell* 139.1 (2009), pp. 161–174. DOI: 10.1016/j.cell.2009.08.027.

[31] Girish S. Bhumbra and Marco Beato. “Recurrent Excitation between Motoneurones Propagates Spinal Motor Output”. In: *eLife* 7 (2018), e34354. DOI: 10.7554/eLife.34354.

[32] Gurdev S. Bhumbra et al. “The recurrent case for the Renshaw cell”. In: *Journal of Neuroscience* 34.38 (2014), pp. 12919–12932. DOI: 10.1523/JNEUROSCI.0199-14.2014.

[33] Tulsi N Bice and John A Beal. “Quantitative and neurogenic analysis of the total population and subpopulations of neurons defined by axon projection in the superficial dorsal horn of the rat lumbar spinal cord”. In: *Journal of Comparative Neurology* 388.4 (Dec. 1997), pp. 550–564. ISSN: 0021-9967. DOI: https://doi.org/10.1002/(SICI)1096-9861(19971201)388:4<550::AID-CNE4>3.0.CO;2-1. URL: https://doi.org/10.1002/(SICI)1096-9861(19971201)388:4%3C550::AID-CNE4%3E3.0.CO%20http://2-1.

[34] Tulsi N. Bice and John A. Beal. “Quantitative and neurogenic analysis of neurons with supraspinal projections in the superficial dorsal horn of the rat lumbar spinal cord”. In: *Journal of Comparative Neurology* 388.4 (1997), pp. 565–574. ISSN: 00219967. DOI: 10.1002/(SICI)1096-9861(19971201)388:4<565::AID-CNE5>3.0.CO;2-0.

[35] Andrew A. Biewener. “Patterns of mechanical energy change in tetrapod gait: pendula, springs and work”. In: *Journal of Experimental Zoology Part A: Comparative Experimental Biology* 305A (2006), pp. 899–911. DOI: 10.1002/jez.a.334.

[36] Jay B. Bikoff et al. “Spinal inhibitory interneuron diversity delineates variant motor microcircuits”. In: *Cell* 165 (2016), pp. 207–219. DOI: 10.1016/j.cell.2016.01.027.

[37] Zoltán Birinyi et al. “Synaptic targets of commissural interneurons in the lumbar spinal cord of neonatal rats”. In: *Journal of Comparative Neurology* 461.4 (2003), pp. 429–440. DOI: 10.1002/cne.10696.

[38] Zoltán Biró, Russell H. Hill, and Sten Grillner. “The activity of spinal commissural interneurons during fictive locomotion in the lamprey”. In: *Journal of Neurophysiology* 100.2 (2008), pp. 716–722. DOI: 10.1152/jn.90206.2008.

[39] Ericka R. Björnfors and Abdeljabbar El Manira. “Functional diversity of excitatory commissural interneurons in adult zebrafish”. In: *eLife* 5 (2016), e18579. DOI: 10.7554/eLife.18579.

[40] Jake Blacklaws et al. “Sim1 is required for the migration and axonal projections of V3 interneurons in the developing mouse spinal cord”. In: *Developmental Neurobiology* 75.9 (2015), pp. 1003–1017. ISSN: 1932846X. DOI: 10.1002/dneu.22266.

[41] Jonathan A. Blum et al. “Single-cell genomics of the adult mouse spinal cord”. In: *Nature* 596 (2021), pp. 141–147. DOI: 10.1038/s41586-021-03780-6.

[42] Urs L. Böhm et al. “Voltage imaging identifies spinal circuits that modulate locomotor adaptation in zebrafish”. In: *Neuron* 110.7 (2022), 1211–1222.e4. ISSN: 10974199. DOI: 10.1016/j.neuron.2022.01.001.

[43] Roman Borisyuk et al. “To swim or not to swim: A population-level model of *Xenopus* tadpole decision making and locomotor behaviour”. In: *BioSystems* 161 (2017), pp. 3–14. DOI: 10.1016/j.biosystems.2017.07.004.

[44] Joanna Borowska et al. “Functional subpopulations of V3 interneurons in the mature mouse spinal cord”. In: *Journal of Neuroscience* 33.47 (2013), pp. 18553–18565. ISSN: 02706474. DOI: 10.1523/JNEUROSCI.2005-13.2013.

[45] Stéphanie Bourane, Binh Duan, and et al. “Gate Control of Mechanical Pain by Spinal Inhibitory Interneurons”. In: *Science* 350.6260 (2015), pp. 153–156. DOI: 10.1126/science.aac8381.

[46] Laurent J. Bouyer and Serge Rossignol. “The Contribution of Cutaneous Inputs to Locomotion in the Intact and the Spinal Cat”. In: *Annals of the New York Academy of Sciences* 860 (1998), pp. 508–512. DOI: 10.1111/j.1749-6632.1998.tb09109.x.

[47] Joao Braz et al. “Transmitting Pain and Itch Messages: A Contemporary View of the Spinal Cord Circuits That Generate Gate Control”. In: *Neuron* 82.3 (2014), pp. 522–536. DOI: 10.1016/j.neuron.2014.01.018.

[48] James Briscoe and Johan Ericson. “The specification of neuronal identity by graded Sonic Hedgehog signalling”. In: *Seminars in Cell & Developmental Biology* 10.3 (1999), pp. 353–362. DOI: 10.1006/scdb.1999.0295.

[49] James Briscoe et al. “A homeodomain protein code specifies progenitor cell identity and neuronal fate in the ventral neural tube”. In: *Cell* 101 (2000), pp. 435–445. DOI: 10.1016/S0092-8674(00)80853-3.

[50] Oliver Britz et al. “A genetically defined asymmetry underlies the inhibitory control of flexor–extensor locomotor movements”. In: *eLife* 4 (2015), e04718. DOI: 10.7554/eLife.04718.

[51] T. Graham Brown. “The intrinsic factors in the act of progression in the mammal”. In: *Proceedings of the Royal Society B* 84 (1911), pp. 308–319. DOI: 10.1098/rspb.1911.0077.

[52] Robert M. Brownstone and James M. Wilson. “Strategies for delineating spinal locomotor rhythm-generating networks and the possible role of Hb9 interneurones in rhythmogenesis”. In: *Brain Research Reviews* 57.1 (2008), pp. 64–76. DOI: 10.1016/j.brainresrev.2007.06.025.

[53] James T. Buchanan. “Commissural interneurons in rhythm generation and intersegmental coupling in the lamprey spinal cord”. In: *Journal of Neurophysiology* 81.5 (1999), pp. 2037–2045. DOI: 10.1152/jn.1999.81.5.2037.

[54] James T. Buchanan. “Identification of interneurons with contralateral, caudal axons in the lamprey spinal cord: synaptic interactions and morphology”. In: *Journal of Neurophysiology* 47.5 (1982), pp. 961–975. DOI: 10.1152/jn.1982.47.5.961.

[55] James T. Buchanan and Arthur H. Cohen. “Activities of Identified Interneurons, Motoneurons, and Muscle Fibers during Fictive Swimming in the Lamprey and Effects of Reticulospinal and Dorsal Cell Stimulation”. In: *Journal of Neurophysiology* 47.5 (1982), pp. 948–960.

[56] James T. Buchanan and Sten Grillner. “A new class of small inhibitory interneurons in the lamprey spinal cord”. In: *Brain Research* 438.2 (1988), pp. 404–407. DOI: 10.1016/0006-8993(88)91373-X.

[57] James T. Buchanan and Sten Grillner. “Newly identified “glutamate interneurons” and their role in locomotion in the lamprey spinal cord”. In: *Science* 236 (1987), pp. 312–314. DOI: 10.1126/science.3563512.

[58] James T. Buchanan and David McPherson. “The neuronal network for locomotion in the lamprey spinal cord: evidence for the involvement of commissural interneurons”. In: *Journal of the Autonomic Nervous System* 50 (1995), pp. 23–33. DOI: 10.1016/0165-1838(94)00122-U.

[59] James T. Buchanan and David R. McPherson. “The neuronal network for locomotion in the lamprey spinal cord: evidence for the involvement of commissural interneurons”. In: *Journal of Physiology (Paris)* 89.4–6 (1995), pp. 221–233. DOI: 10.1016/0928-4257(96)83638-2.

[60] James T. Buchanan et al. “Identification of excitatory interneurons contributing to generation of locomotion in lamprey”. In: *Journal of Neurophysiology* 62 (1989), pp. 59–69. DOI: 10.1152/jn.1989.62.1.59.

[61] JT Buchanan and S Grillner. “Newly identified ’glutamate interneurons’ and their role in locomotion in the lamprey spinal cord.” In: *Science* 236.4799 (1987), pp. 312–314. URL: https://pubmed.ncbi.nlm.nih.gov/3563512.

[62] Tuan V. Bui et al. “Role of dI3 Interneurons in Functional Recovery Following Spinal Cord Transection”. In: *Journal name unknown* (2016). Fill in exact journal, volume, pages and doi.

[63] Tuan V. Bui, Robert M. Brownstone, et al. “Glutamatergic dI3 Interneurons Gate Cutaneous Afferent Transmission to Motor Neurons”. In: *Journal name unknown* (2013). Fill in exact journal, volume, pages and doi.

[64] Raymond R. Buss and Pierre Drapeau. “Activation of embryonic red and white muscle fibers during fictive swimming in developing zebrafish”. In: *Journal of Neurophysiology* 87.3 (2002), pp. 1244–1251. DOI: 10.1152/jn.00659.2001.

[65] Simon J. B. Butt and Ole Kiehn. "Functional identification of interneurons responsible for left–right coordination of hindlimbs in mammals". In: *Neuron* 38.6 (2003), pp. 953–963. DOI: 10.1016/S0896-6273(03)00353-2.

[66] Vanessa Caldeira et al. "Spinal Hb9::Cre-derived excitatory interneurons contribute to locomotor-like activity in juvenile mice". In: *Scientific Reports* 7 (2017), p. 41369. DOI: 10.1038/srep41369.

[67] Rachel A. Callahan et al. "Spinal V2b neurons reveal a local inhibitory control of motor circuits in vertebrates". In: *Cell Reports* 29.1 (2019), 172–187.e6. DOI: 10.1016/j.celrep.2019.08.091.

[68] Lorenzo Cangiano and Sten Grillner. "Fast and slow oscillations in the lamprey spinal cord: mechanisms and role in pattern generation". In: *Journal of Neurophysiology* 90.5 (2003), pp. 2937–2950. DOI: 10.1152/jn.00402.2003.

[69] Lorenzo Cangiano and Sten Grillner. "Mechanisms of rhythm generation in a spinal locomotor network deprived of crossed connections: The lamprey hemicord". In: *Journal of Neuroscience* 25.4 (2005), pp. 923–935. ISSN: 02706474. DOI: 10.1523/JNEUROSCI.2301-04.2005.

[70] Lorenzo Cangiano and Sten Grillner. "Mechanisms of rhythm generation in a spinal locomotor network deprived of crossed connections: the lamprey hemicord". In: *Journal of Neuroscience* 25.40 (2005), pp. 923–935. DOI: 10.1523/JNEUROSCI.3160-04.2005.

[71] Leping Cheng et al. "Lbx1 and Tlx3 are opposing switches in determining GABAergic versus glutamatergic transmitter phenotypes". In: *Nature Neuroscience* 8.11 (2005), pp. 1510–1515. ISSN: 10976256. DOI: 10.1038/nn1569.

[72] Leping Cheng et al. "Tlx3 and Tlx1 are post-mitotic selector genes determining glutamatergic over GABAergic cell fates". In: *Nature Neuroscience* 7.5 (2004), pp. 510–517. ISSN: 10976256. DOI: 10.1038/nn1221.

[73] Nasim Cheng et al. "Spinal cords: Symphonies of interneurons across species". In: *Frontiers in Neural Circuits* 17 (2023), p. 1146449. DOI: 10.3389/fncir.2023.1146449.

[74] John W. Chopek et al. "Sub-populations of spinal V3 interneurons form focal modules of layered premotor microcircuits". In: *Cell Reports* 25 (2018), 146–156.e3. DOI: 10.1016/j.celrep.2018.08.095.

[75] J. Christenson, K. Lagerbäck, and S. Grillner. "The dorsal cell, one class of primary sensory neuron in the lamprey spinal cord. II. A light- and electron microscopical study". In: *Brain Research* 440.1 (1988), pp. 9–17.

[76] J. D. W. Clarke et al. “Sensory Physiology, Anatomy and Immunohistochemistry of Rohon–Beard Neurones in Embryos of *Xenopus laevis*”. In: *The Journal of Physiology* 348.1 (1984), pp. 511–525. DOI: 10.1113/jphysiol.1984.sp015122.

[77] Alan H. Cohen and Ronald M. Harris-Warrick. “S-N-methyl-D-aspartate and L-aspartate induce locomotor-like rhythmicity in lamprey spinal cord”. In: *Science* 225 (1984), pp. 1022–1024. DOI: 10.1126/science.6474170.

[78] Denis Combes et al. “Developmental segregation of spinal networks driving axial- and hindlimb-based locomotion in metamorphosing *Xenopus laevis*”. In: *Journal of Physiology* 559 (2004), pp. 17–24. DOI: 10.1113/jphysiol.2004.069542.

[79] T. C. Cope, B. D. Clark, and S. C. Bodine. “Motor-unit recruitment in mice: interactions among unit properties, muscle properties and neural drive”. In: *Journal of Neurophysiology* 74.4 (1995), pp. 1568–1579.

[80] Sarah A. Crone et al. “Genetic ablation of V2a ipsilateral interneurons disrupts left–right locomotor coordination in mammalian spinal cord”. In: *Neuron* 60 (2008), pp. 70–83. DOI: 10.1016/j.neuron.2008.08.009.

[81] Liang Cui et al. “Identification of a Spinal Circuit for Mechanical and Cold Pain”. In: *Cell* 167.5 (2016), 1415–1428.e9. DOI: 10.1016/j.cell.2016.10.025.

[82] Stephen P. Currie and Keith T. Sillar. “Developmental changes in spinal neuronal properties, motor network configuration, and neuromodulation at free-swimming stages of xenopus tadpoles”. In: *Journal of Neurophysiology* 119.3 (2018), pp. 786–795. ISSN: 15221598. DOI: 10.1152/jn.00219.2017.

[83] Katherine P. D’Elia and Jeremy S. Dasen. “Evolution of Motor Neuron Diversity and Function”. In: *Current Opinion in Neurobiology* 53 (2018), pp. 1–6. DOI: 10.1016/j.conb.2018.04.007.

[84] Nicholas Dale. “Excitatory amino acid receptors in Xenopus embryo spinal cord and their role in the activation of swimming”. In: *Journal of Physiology* 348 (1984), pp. 527–543. DOI: 10.1113/jphysiol.1984.sp015123.

[85] Nicholas Dale. “Excitatory synaptic drive for swimming mediated by amino acid receptors in the lamprey spinal cord”. In: *Journal of Neuroscience* 6.3 (1986), pp. 2662–2675.

[86] Nicholas Dale and Alan Roberts. “Dual-component amino-acid-mediated synaptic potentials: excitatory drive for swimming in Xenopus embryos”. In: *Journal of Physiology* 363 (1985), pp. 35–59.

[87] Simon M. Danner et al. “Computational modeling of spinal circuits controlling limb coordination and gait patterns in quadrupeds”. In: *eLife* 6 (2017), e31050. DOI: 10.7554/eLife.31050.

[88] Simon M. Danner et al. “Spinal V3 Interneurons and Left–Right Coordination in Mammalian Locomotion”. In: *Frontiers in Cellular Neuroscience* 13.November (2019), pp. 1–23. ISSN: 16625102. DOI: 10.3389/fncel.2019.00516.

[89] Jeremy S. Dasen. “Hox regulation of vertebrate motor neuron diversity”. In: *Current Opinion in Neurobiology* 74 (2022), p. 102542. DOI: 10.1016/j.conb.2022.102542.

[90] Jeremy S. Dasen and Thomas M. Jessell. “Hox networks and the origins of motor neuron diversity”. In: *Current Topics in Developmental Biology* 88 (2009), pp. 169–200. DOI: 10.1016/S0070-2153(09)88006-X.

[91] Jeremy S. Dasen and Thomas M. Jessell. “Hox networks and the origins of motor neuron diversity”. In: *Current Topics in Developmental Biology* 88 (2009), pp. 169–200. DOI: 10.1016/S0070-2153(09)88006-X.

[92] Christopher A Del Negro et al. “Breathing matters”. In: *Nat Rev Neurosci.* 19.6 (2019), pp. 351–367. DOI: 10.1038/s41583-018-0003-6.Breathing.

[93] Ana Del Pozo et al. “Behavioral characterization of dmrt3a mutant zebrafish reveals crucial aspects of vertebrate locomotion through phenotypes related to acceleration”. In: *eNeuro* 7.3 (2020), pp. 1–13. ISSN: 23732822. DOI: 10.1523/ENEURO.0047-20.2020.

[94] Julien Delile et al. “Single cell transcriptomics reveals spatial and temporal dynamics of gene expression in the developing mouse spinal cord”. In: *Nature Communications* 10 (2019), p. 2733. DOI: 10.1038/s41467-019-10536-2.

[95] Denise Deska-Gauthier et al. “Locomotor rhythm generation linked to the output of spinal V3 interneurons”. In: *Cell Reports* 31.10 (2020), p. 107784. DOI: 10.1016/j.celrep.2020.107784.

[96] Jared Diamond. “The Mauthner Cell”. In: *Fish Physiology, Volume 5: Sensory Systems and Electric Organs*. Ed. by W. S. Hoar and D. J. Randall. Vol. 5. New York and London: Academic Press, 1971, pp. 265–346. ISBN: 1546-5098. DOI: 10.1016/S1546-5098(08)60050-3. URL: https://www.sciencedirect.com/science/article/pii/S1546509808600503.

[97] Markus Diesmann, Marc-Oliver Gewaltig, and Ad Aertsen. “Stable propagation of synchronous spiking in cortical neural networks”. In: *Nature* 402.6761 (1999), pp. 529–533. DOI: 10.1038/990101.

[98] Qian Ding et al. “BARHL2 Transcription Factor Regulates the Ipsilateral/Contralateral Subtype Divergence in Postmitotic dI1 Neurons of the Developing Spinal Cord”. In: *Proceedings of the National Academy of Sciences of the USA* 109.5 (2012), pp. 1566–1571. DOI: 10.1073/pnas.1112392109.

[99] Kimberly J. Dougherty and Ole Kiehn. “Functional organization of V2a-related locomotor circuits in the rodent spinal cord”. In: *Annals of the New York Academy of Sciences* 1198 (2010), pp. 85–93. DOI: 10.1111/j.1749-6632.2010.05439.x.

[100] Kimberly J. Dougherty and Ole Kiehn. “Functional organization of V2a-related locomotor circuits in the rodent spinal cord”. In: *Annals of the New York Academy of Sciences* 1198 (2010), pp. 85–93. DOI: 10.1111/j.1749-6632.2010.05520.x.

[101] Kimberly J. Dougherty et al. “Locomotor rhythm generation linked to the output of spinal Shox2 interneurons”. In: *Neuron* 80.4 (2013), pp. 920–933. DOI: 10.1016/j.neuron.2013.08.015.

[102] B. Duan et al. “Identification of Spinal Circuits Transmitting and Gating Mechanical Pain”. In: *Cell* 159.6 (2014), pp. 1417–1432. DOI: 10.1016/j.cell.2014.11.003.

[103] Jason Dyck, Guillermo M. Lanuza, and Simon Gosgnach. “Functional characterization of dI6 interneurons in the neonatal mouse spinal cord”. In: *Journal of Neurophysiology* 107.12 (2012), pp. 3256–3266.

[104] J C Eccles, P Fatt, and S Landgren. “CENTRAL PATHWAY FOR DIRECT INHIBITORY ACTION OF IMPULSES IN LARGEST AFFERENT NERVE FIBRES TO MUSCLE”. In: *Journal of Neurophysiology* 19.1 (Jan. 1956), pp. 75–98. ISSN: 0022-3077. DOI: 10.1152/jn.1956.19.1.75. URL: https://doi.org/10.1152/jn.1956.19.1.75.

[105] Örjan Ekeberg and Sten Grillner. “Simulations of neuromuscular control in lamprey swimming”. In: *Philosophical Transactions of the Royal Society B: Biological Sciences* 354.1385 (1999), pp. 895–902. DOI: 10.1098/rstb.1999.0441.

[106] Emelie Eklöf Ljunggren and et al. “Optogenetic activation of V2a neurons drives locomotion in zebrafish”. In: *Frontiers in Neural Circuits* 8 (2014), p. 76. DOI: 10.3389/fncir.2014.00076.

[107] Emelie Eklöf-Ljunggren et al. “Origin of excitation underlying locomotion in the spinal circuit of zebrafish”. In: *Proceedings of the National Academy of Sciences USA* 109 (2012), pp. 5511–5516. DOI: 10.1073/pnas.1115377109.

[108] A. El Manira, J. Tegnér, and S. Grillner. "Intrinsic Membrane Properties of Neurons in the Lamprey Spinal Locomotor Network". In: *Brain Research* 769.2 (1997), pp. 179–187. DOI: 10.1016/S0006-8993(97)00861-3.

[109] Adriana Escalante and Rüdiger Klein. "Spinal Neuron Subtype Diversification from Shared Precursors: Mechanisms and Functional Implications". In: *Cell Reports* 31.10 (2020), p. 107739. DOI: 10.1016/j.celrep.2020.107739.

[110] Melanie Falgairolle and Michael J. O'Donovan. "Optogenetic activation of V1 interneurons reveals the multimodality of spinal locomotor networks in the neonatal mouse". In: *Journal of Neuroscience* 41.41 (2021), pp. 8545–8561. DOI: 10.1523/JNEUROSCI.0875-21.2021. URL: https://doi.org/10.1523/JNEUROSCI.0875-21.2021.

[111] Melanie Falgairolle and Michael J. O'Donovan. "V1 interneurons regulate the pattern and frequency of locomotor-like activity in the neonatal mouse spinal cord". In: *PLOS Biology* 17.9 (2019), e3000447. DOI: 10.1371/journal.pbio.3000447. URL: https://doi.org/10.1371/journal.pbio.3000447.

[112] Michaël Falgairolle and Michael J. O'Donovan. "Motoneurons regulate the central pattern generator during drug-induced locomotor-like activity in the neonatal mouse spinal cord". In: *eLife* 6 (2017), e26622. DOI: 10.7554/eLife.26622.

[113] A.G Feldman and G.N. Orlovsky. "Activity of interneurons mediating reciprocal 1a inhibition during locomotion". In: *Brain Research* 84.2 (1975), pp. 181–194.

[114] B. Fernández-López et al. "The glutamatergic neurons in the spinal cord of the sea lamprey, *Petromyzon marinus*". In: *PLOS ONE* 7.6 (2012), e38566.

[115] Blanca Fernández-López et al. "The Glutamatergic Neurons in the Spinal Cord of the Sea Lamprey: An In Situ Hybridization and Immunohistochemical Study". In: *PLoS ONE* 7.10 (2012). ISSN: 19326203. DOI: 10.1371/journal.pone.0047898.

[116] Andrea Ferrario et al. "From decision to action: Detailed modelling of frog tadpoles reveals neuronal mechanisms of decision-making and reproduces unpredictable swimming movements in response to sensory signals". In: *PLoS Computational Biology* 17.12 (2021), e1009654. DOI: 10.1371/journal.pcbi.1009654.

[117] Andrea Ferrario et al. "Mechanisms Underlying the Recruitment of Inhibitory Interneurons in Fictive Swimming in Developing *Xenopus laevis* Tadpoles". In: *Journal of Neuroscience* 43.8 (2023), pp. 1387–1404. DOI: 10.1523/JNEUROSCI.0520-22.2022.

[118] Andrea Ferrario et al. “Structural and functional properties of a probabilistic model of neuronal connectivity in a simple locomotor network”. In: *eLife* 7 (2018), e33281. DOI: 10.7554/eLife.33281. URL: https://doi.org/10.7554/eLife.33281.

[119] Kevin Fidelin and Claire Wyart. “Locomotion modulates early spontaneous motor neuron activity in zebrafish embryos”. In: *Current Biology* 25.14 (2015), pp. 1780–1787. DOI: 10.1016/j.cub.2015.05.057.

[120] Andrew J. P. Fink et al. “Presynaptic inhibition of spinal sensory feedback ensures smooth movement”. In: *Nature* 509 (2014), pp. 43–48. DOI: 10.1038/nature13276.

[121] Hans Forssberg. “Stumbling corrective reaction: A phase-dependent compensatory reaction during locomotion”. In: *Journal of Neurophysiology* 42 (1979), pp. 936–953. DOI: 10.1152/jn.1979.42.4.936.

[122] Hans Forssberg, Sten Grillner, and Serge Rossignol. “Phase dependent reflex reversal during walking in chronic spinal cats”. In: *Brain Research* 85 (1980), pp. 103–107. DOI: 10.1016/0006-8993(80)90642-8.

[123] Hans Forssberg, Sten Grillner, and Serge Rossignol. “Phasic Gain Control of Reflexes from the Dorsum of the Paw during Spinal Locomotion”. In: *Brain Research* 132.1 (1977), pp. 121–139. DOI: 10.1016/0006-8993(77)90711-6.

[124] Hans Forssberg et al. “The locomotion of the low spinal cat. II. Interlimb coordination”. In: *Acta Physiologica Scandinavica* 108 (1980), pp. 283–295.

[125] Evan Foster, Christina Beattie, and et al. “GABAergic Neurons in Laminae III–IV of the Spinal Cord Regulate Mechanical Sensitivity via Inhibition of Cutaneous Afferents”. In: *Neuron* 85.6 (2015), pp. 1319–1331. DOI: 10.1016/j.neuron.2015.02.028.

[126] Clément Francius et al. “Identification of multiple subsets of spinal V1 interneurons”. In: *Development* 140 (2013), pp. 2159–2168. DOI: 10.1242/dev.091074.

[127] Anselm Friese, Julia A. Kaltschmidt, and et al. “Gamma and alpha motor neurons distinguished by expression of transcription factor Err3”. In: *Proceedings of the National Academy of Sciences USA* 106 (2009), pp. 13588–13593. DOI: 10.1073/pnas.0906809106.

[128] Zipeng Fu et al. “Minimizing Energy Consumption Leads to the Emergence of Gaits in Legged Robots”. In: *Proceedings of the Conference on Robot Learning (CoRL)*. 2021, pp. 1–13. arXiv: 2111.01674. URL: http://arxiv.org/abs/2111.01674.

[129] Matthew I. Gabitto et al. “Bayesian sparse regression analysis documents the diversity of spinal inhibitory interneurons”. In: *Cell* 165 (2016), pp. 220–233. DOI: 10.1016/j.cell.2016.01.026.

[130] David A. Gabriel, Gary Kamen, and Grant Frost. “Neural adaptations to resistive exercise: mechanisms and recommendations for training practices”. In: *Sports Medicine* 41.5 (2011), pp. 365–384.

[131] J. P. Gabriel et al. “Principles governing recruitment of motoneurons during swimming in zebrafish”. In: *Nature Neuroscience* 11 (2008), pp. 533–539. DOI: 10.1038/nn.2097.

[132] Lidia Garcia-Campmany, Floor J. Stam, and Martyn Goulding. “From circuits to behaviour: motor networks in vertebrates”. In: *Current Opinion in Neurobiology* 20.2 (2010), pp. 116–125. DOI: 10.1016/j.conb.2010.01.002.

[133] Giovanni Gatto, Martyn Goulding, et al. “A Functional Topographic Map for Spinal Sensorimotor Reflexes”. In: *Neuron* (2021). Check volume, pages and full author list.

[134] Graziana Gatto et al. “Neuronal diversity in the somatosensory system: bridging the gap between cell type and function”. In: *Current Opinion in Neurobiology* 56 (2019), pp. 167–174. ISSN: 18736882. DOI: 10.1016/j.conb.2019.03.002. URL: https://doi.org/10.1016/j.conb.2019.03.002.

[135] Wulfram Gerstner and Werner M. Kistler. *Spiking Neuron Models: Single Neurons, Populations, Plasticity*. Cambridge, UK: Cambridge University Press, 2002. ISBN: 9780521890791.

[136] Stacey M. Glasgow et al. “Ptf1a Determines GABAergic over Glutamatergic Neuronal Cell Fate in the Spinal Cord Dorsal Horn”. In: *Development* 132.3 (2005), pp. 5461–5469. DOI: 10.1242/dev.02167.

[137] Cynthia Goetz, Chiara Pivetta, and Silvia Arber. “Intrinsic Functional Diversity of Anatomically Defined Spinal Interneuron Populations”. In: *Nature Neuroscience* 18 (2015), pp. 1101–1109. DOI: 10.1038/nn.4042.

[138] Alejandro Gonzalez-Voyer, Svante Winberg, and Niclas Kolm. “Brain structure evolution in a basal vertebrate clade: Evidence from phylogenetic comparative analysis of cichlid fishes”. In: *BMC Evolutionary Biology* 9.1 (2009), pp. 1–12. ISSN: 14712148. DOI: 10.1186/1471-2148-9-238.

[139] Simon Gosgnach et al. “Delineating the diversity of spinal interneurons in locomotor circuits”. In: *Journal of Neuroscience* 37.45 (2017), pp. 10835–10841. ISSN: 15292401. DOI: 10.1523/JNEUROSCI.1829-17.2017.

[140] Simon Gosgnach et al. “V1 spinal neurons regulate the speed of vertebrate locomotor outputs”. In: *Nature* 440 (2006), pp. 215–219. DOI: 10.1038/nature04545.

[141] Martyn Goulding. “Circuits Controlling Vertebrate Locomotion: Moving in a New Direction”. In: *Nature Reviews Neuroscience* 15 (2014), pp. 217–232. DOI: 10.1038/nrn3704.

[142] Martyn Goulding. “Circuits controlling vertebrate locomotion: moving in a new direction”. In: *Nature Reviews Neuroscience* 10 (2009), pp. 507–518. DOI: 10.1038/nrn2608.

[143] Kristen Gowan, Alex S. Helms, et al. “Crossinhibitory Activities of Ngn1 and Math1 Allow Specification of Distinct Dorsal Interneurons”. In: *Neuron* (2001). Check volume and page numbers.

[144] A. Griener et al. “Anatomical and electrophysiological characterization of a population of dI6 interneurons in the neonatal mouse spinal cord”. In: *Neuroscience* 362 (2017), pp. 47–59.

[145] Andrea M. Griener et al. “Extensive axonal branching of developing zebrafish inhibitory interneurons”. In: *Frontiers in Neural Circuits* 9 (2015), p. 50. DOI: 10.3389/fncir.2015.00050.

[146] Sten Grillner. “Human locomotor circuits conform to the primitive vertebrate pattern”. In: *Science* 334.6058 (2011), pp. 912–913. DOI: 10.1126/science.1214778.

[147] Sten Grillner and Abdeljabbar El Manira. “Current principles of motor control, with special reference to vertebrate locomotion”. In: *Physiological Reviews* 100.1 (2020), pp. 271–320. ISSN: 15221210. DOI: 10.1152/physrev.00015.2019.

[148] Sten Grillner and Abdeljabbar El Manira. “Current principles of motor control, with special reference to vertebrate locomotion”. In: *Physiological Reviews* 100 (2020), pp. 271–320. DOI: 10.1152/physrev.00015.2019.

[149] M. K. Gross, M. Dottori, and M. Goulding. “Lbx1 distinguishes two major programs of neuronal differentiation in the dorsal spinal cord”. In: *Neuron* 34.4 (2002), pp. 551–562.

[150] Changhe Guan and et al. “Cholinergic V2a neurons facilitate escape swimming in zebrafish via electrical synapses with motoneurons”. In: *Neuron* 109 (2021), 992–1007.e5. DOI: 10.1016/j.neuron.2021.01.004.

[151] B. Gustafsson and M. J. Pinter. “An investigation of threshold properties among cat spinal $\alpha$-motoneurones”. In: *Journal of Physiology* 357 (1984), pp. 453–483.

[152] B. Gustafsson and M. J. Pinter. “Relations among passive electrical properties of lumbar $\alpha$-motoneurones of the cat”. In: *Journal of Physiology* 356 (1984), pp. 401–431.

[153] Nobue Ha and Kimberly J. Dougherty. “Spinal Shox2 interneuron interconnectivity related to function and development”. In: *eLife* 7 (2018), e42519. DOI: 10.7554/eLife.42519.

[154] Alison Hagevik and Andrew D. McClellan. “Coupling between spinal locomotor networks in larval lamprey revealed by lesioning and pharmacology”. In: *Journal of Neurophysiology* 72.1 (1994), pp. 181–195. DOI: 10.1152/jn.1994.72.1.181.

[155] Daniel Haimson et al. “dI2 Interneurons Coordinate Limb Movements in Avian Spinal Circuits”. In: *Journal name unknown* (2021). Title/journal are approximate placeholders—please replace with the exact citation.

[156] Melinda E. Hale, David A. Ritter, and Joseph R. Fetcho. “A confocal study of spinal interneurons in living larval zebrafish”. In: *Journal of Comparative Neurology* 437 (2001), pp. 1–16. DOI: 10.1002/cne.1267.

[157] Zhao Hao and Ari Berkowitz. “Response Properties of Turtle Spinal Interneurons during Multiple Forms of Scratch Reflex and Swimming”. In: *Journal of Neurophysiology* 106.3 (2011), pp. 1711–1724. DOI: 10.1152/jn.00247.2011.

[158] Fatema Haque et al. “WT1-expressing interneurons in the spinal cord coordinate locomotor behavior in mice”. In: *eLife* 7 (2018), e32555.

[159] Martin Häring et al. “Neuronal atlas of the dorsal horn”. In: *Science* 360 (2018), eaat8464. DOI: 10.1126/science.aat8464.

[160] Nalin Harischandra et al. “Sensory feedback plays a significant role in generating walking gait and in gait transition in salamanders: a simulation study”. In: *Frontiers in Neurorobotics* 5 (2011), p. 3. DOI: 10.3389/fnbot.2011.00003.

[161] C. B. Hart and S. F. Giszter. “Modular premotor drives and unit bursts as primitives for frog motor behaviors”. In: *Journal of Neuroscience* 24.22 (2004), pp. 5269–5282. DOI: 10.1523/JNEUROSCI.5626-03.2004.

[162] Marito Hayashi et al. “Graded Arrays of Spinal and Supraspinal V2a Interneurons Underlie Forelimb Locomotor Control”. In: *Cell Reports* 23 (2018), pp. 125–136. DOI: 10.1016/j.celrep.2018.03.031.

[163] Masaki Hayashi and Martyn Goulding. “Ascending V2a Neurons Enable Coordination of Forelimb Movements”. In: *Nature* 613 (2023). Replace with final volume/pages when available, in press.

[164] Amy W. Helms and Jane E. Johnson. “Specification of Dorsal Spinal Cord Interneurons”. In: *Current Opinion in Neurobiology* 13.1 (2003), pp. 42–49. DOI: 10.1016/S0959-4388(03)00013-3.

[165] James A. Helms and Janet E. Johnson. "Specification of Dorsal Spinal Cord Interneurons". In: *Current Opinion in Neurobiology* 13.1 (2003). Check authorship/initials; sometimes cited as Helms & Johnson, pp. 42–49.

[166] E. Henneman, G. Somjen, and D. O. Carpenter. "Functional Significance of Cell Size in Spinal Motoneurons". In: *Journal of Neurophysiology* 28.3 (1965), pp. 560–580. DOI: 10.1152/jn.1965.28.3.560.

[167] Luis R. Hernández-Miranda, Thomas Müller, and Carmen Birchmeier. "The dorsal spinal cord and hindbrain: From progenitor cells to neuronal circuits". In: *Developmental Biology* 432.1 (2017), pp. 34–42.

[168] Shin Ichi Higashijima, Michael Schaefer, and Joseph R. Fetcho. "Neurotransmitter properties of spinal interneurons in embryonic and larval zebrafish". In: *Journal of Comparative Neurology* 480.1 (2004), pp. 19–37. ISSN: 00219967. DOI: 10.1002/cne.20279.

[169] Shin-ichi Higashijima, Michael Schaefer, and Joseph R. Fetcho. "Neurotransmitter properties of spinal interneurons in embryonic and larval zebrafish". In: *Journal of Comparative Neurology* 480.1 (2004), pp. 19–37. DOI: 10.1002/cne.20279.

[170] Shin-ichi Higashijima et al. "Engrailed-1 expression marks a primitive class of inhibitory spinal interneuron". In: *Journal of Neuroscience* 24.25 (2004), pp. 5827–5839. DOI: 10.1523/JNEUROSCI.5342-03.2004.

[171] Kevin L. Hilde et al. "Satb2 Is Required for the Development of a Spinal Exteroceptive Microcircuit That Modulates Limb Position". In: *Neuron* 91.4 (2016), pp. 763–776. DOI: 10.1016/j.neuron.2016.07.023.

[172] R. Hill, G.A. Wyse, and M. Anderson. *Animal Physiology*. Sunderland, Massachusetts: Sinauer, 2016.

[173] Christopher A. Hinckley and Lea Ziskind-Conhaim. "Electrical coupling between locomotor-related excitatory interneurons in the mammalian spinal cord". In: *Journal of Neuroscience* 26.33 (2006), pp. 8477–8483. DOI: 10.1523/JNEUROSCI.0934-06.2006.

[174] Christopher A. Hinckley and Lea Ziskind-Conhaim. "Persistent sodium current contributes to induced voltage oscillations in locomotor-related Hb9 interneurons in the mouse spinal cord". In: *Journal of Neurophysiology* 103.5 (2010), pp. 3407–3420. DOI: 10.1152/jn.01076.2009.

[175] Christopher A. Hinckley et al. "Locomotor-like rhythms in a genetically distinct cluster of interneurons in the mammalian spinal cord". In: *Journal of Neurophysiology* 93.3 (2005), pp. 1439–1449. ISSN: 00223077. DOI: 10.1152/jn.00647.2004.

[176] Craig A. Hinckley et al. “Locomotor-Like Rhythmicity Is Generated by Synaptic Glutamatergic Actions in the Mammalian Spinal Cord”. In: *Scientific Reports* 5 (2015), p. 9511. DOI: 10.1038/srep09511.

[177] A. L. Hodgkin and A. F. Huxley. “A quantitative description of membrane current and its application to conduction and excitation in nerve”. In: *The Journal of Physiology* 117 (1952), pp. 500–544. DOI: 10.1113/jphysiol.1952.sp004764.

[178] Mikael Huss. “Computational Modeling of the Lamprey CPG: From Subcellular to Network Level”. PhD thesis. Umeå, Sweden: Umeå University, 2007. URL: http://umu.diva-portal.org/smash/record.jsf?pid=diva2:12226.

[179] Ana Belén Iglesias González et al. “Single Cell Transcriptomic Analysis of Spinal Dmrt3 Neurons in Zebrafish and Mouse Identifies Distinct Subtypes and Reveal Novel Subpopulations Within the dI6 Domain”. In: *Frontiers in Cellular Neuroscience* 15.December (2021), pp. 1–19. ISSN: 16625102. DOI: 10.3389/fncel.2021.781197.

[180] Auke Jan Ijspeert et al. “From swimming to walking with a salamander robot driven by a spinal cord model”. In: *Science* 315.5817 (2007), pp. 1416–1420. DOI: 10.1126/science.1138353.

[181] Bruce C. Jayne and George V. Lauder. “How swimming fish use slow and fast muscle fibers: implications for models of vertebrate muscle function”. In: *Journal of Experimental Biology* 194 (1994), pp. 141–153.

[182] Thomas M. Jessell. “Neuronal specification in the spinal cord: inductive signals and transcriptional codes”. In: *Nature Reviews Genetics* 1 (2000), pp. 20–29. DOI: 10.1038/35049541.

[183] H. Jung et al. “Global Control of Motor Neuron Topography Mediated by the Repressive Actions of a Single Transcription Factor”. In: *Neuron* 98.5 (2018), 945–962.e6. DOI: 10.1016/j.neuron.2018.04.025.

[184] Andre Kardon et al. “Dorsal Horn Neurons Expressing TRPV1 Control Itch”. In: *Nature Neuroscience* 17 (2014), pp. 804–811. DOI: 10.1038/nn.3719.

[185] Harvey J. Karten. “Vertebrate brains and evolutionary connectomics: On the origins of the mammalian ‘neocortex’”. In: *Philosophical Transactions of the Royal Society B: Biological Sciences* 370.1684 (2015). ISSN: 14712970. DOI: 10.1098/rstb.2015.0060.

[186] Aruna Karunaratne et al. “GATA2 and GATA3 regulate markers of V2b inhibitory interneuron identity in the developing spinal cord”. In: *Development* 129 (2002), pp. 3713–3724.

[187] Keisuke Kawano et al. “Spinal V0v neurons regulate speed-dependent head stabilization and diagonal coupling during swimming in zebrafish”. In: *Nature Communications* 13 (2022), p. 1994. DOI: 10.1038/s41467-022-29658-0.

[188] Ole Kiehn. “Decoding the organization of the spinal locomotor central pattern generator”. In: *Trends in Neurosciences* 39.3 (2016), pp. 158–169. DOI: 10.1016/j.tins.2016.01.002.

[189] Ole Kiehn. “Locomotor circuits in the mammalian spinal cord”. In: *Annual Review of Neuroscience* 29 (2006), pp. 279–306. DOI: 10.1146/annurev.neuro.29.051605.112910.

[190] Yasushi Kimura, Yukiko Okamura, and Shin-ichi Higashijima. “V1 neurons and locomotor control in zebrafish”. In: *Science Advances* 5.7 (2019), eaav6934. DOI: 10.1126/sciadv.aav6934.

[191] Yukiko Kimura, Yasushi Okamura, and Shin-ichi Higashijima. “alx, a zebrafish homolog of Chx10, marks ipsilateral descending excitatory interneurons that participate in the regulation of locomotion”. In: *Journal of Neuroscience* 26 (2006), pp. 5684–5697. DOI: 10.1523/JNEUROSCI.4993-05.2006.

[192] Yukiko Kimura et al. “Hindbrain V2a neurons in the initiation of locomotion in zebrafish”. In: *Journal of Neuroscience* 33 (2013), pp. 1164–1173. DOI: 10.1523/JNEUROSCI.3759-12.2013.

[193] S. Kishore et al. “Orderly compartmental mapping of premotor inhibition inputs in developing spinal cord”. In: *Science* 370.6519 (2020), pp. 431–436. DOI: 10.1126/science.abb6809.

[194] S. Kishore et al. “Orderly compartmental mapping of premotor inhibition inputs in developing spinal cord”. In: *Science* 370.6519 (2020), pp. 431–436. DOI: 10.1126/science.abb6809.

[195] Shailesh Kishore, Martha W. Bagnall, and David L. McLean. “Systematic shifts in the balance of excitation and inhibition coordinate the activity of axial motor pools at different speeds of locomotion”. In: *Journal of Neuroscience* 34.4 (2014), pp. 14046–14054. DOI: 10.1523/JNEUROSCI.0514-14.2014.

[196] Olaf Kjaerulff and Ole Kiehn. “Crossed phasic synaptic input to motoneurons during selective activation of the contralateral spinal locomotor network”. In: *Journal of Neuroscience* 17.24 (1997), pp. 9433–9447. DOI: 10.1523/JNEUROSCI.17-24-09433.1997.

[197] Laura D. Knogler and Pierre Drapeau. “Sensory gating by spinal inhibitory neurons in zebrafish”. In: *Frontiers in Neural Circuits* 8 (2014), p. 35. DOI: 10.3389/fncir.2014.00035.

[198] Jeremie Knüsel et al. “A Salamander’s Flexible Spinal Network for Locomotion, Modeled at Two Levels of Abstraction”. In: *Integrative and Comparative Biology* 53.2 (2013), pp. 269–282. DOI: 10.1093/icb/ict067.

[199] Sebastian C. Koch, Sarah T. Acton, and Martyn Goulding. “Spinal Circuits for Touch, Pain, and Itch”. In: *Annual Review of Physiology* 80 (2018), pp. 189–217. DOI: 10.1146/annurev-physiol-022516-034303.

[200] Sebastian C. Koch et al. “ROR$\beta$ Spinal Interneurons Gate Sensory Transmission during Locomotion to Secure Gait Rhythm”. In: *Neuron* 96.6 (2017), 1419–1431.e5. DOI: 10.1016/j.neuron.2017.11.011.

[201] Lina M. Koronfel et al. “Elimination of glutamatergic transmission from Hb9 interneurons does not impact treadmill locomotion”. In: *Scientific Reports* 11 (2021), p. 16008. DOI: 10.1038/s41598-021-95143-y.

[202] Stella Koutsikou et al. “A simple decision to move in response to touch reveals basic sensory memory and mechanisms for variable response times”. In: *The Journal of Physiology* 596.24 (2018), pp. 6219–6233. DOI: 10.1113/JP276356.

[203] A. K. Kozlov et al. “A hemicord locomotor network of excitatory interneurons: A simulation study”. In: *Biological Cybernetics* 96.2 (2007), pp. 229–243. ISSN: 03401200. DOI: 10.1007/s00422-006-0132-2.

[204] Andrei Kozlov and et al. “Gating of NMDA currents by short-term plasticity of inhibition shapes locomotor frequency in the lamprey spinal cord”. In: *Journal of Neurophysiology* 101 (2009), pp. 225–235. DOI: 10.1152/jn.90697.2008.

[205] Arvind Kumar, Stefan Rotter, and Ad Aertsen. “Spiking activity propagation in neuronal networks: reconciling different perspectives on neural coding”. In: *Nature Reviews Neuroscience* 11.9 (2010), pp. 615–627. DOI: 10.1038/nrn2886.

[206] Hoang C. Lai, Rebecca P. Seal, and Jane E. Johnson. “Making Sense out of Spinal Cord Somatosensory Development”. In: *Neuron* 92.2 (2016), pp. 253–255. DOI: 10.1016/j.neuron.2016.10.009.

[207] Yiran Lang et al. “Multisite simultaneous neural recording of motor pathway in free-moving rats”. In: *Biosensors* 11.12 (2021). ISSN: 20796374. DOI: 10.3390/bios11120503.

[208] Anders Lansner et al. “Computer simulation of the segmental neural network generating locomotor activity in the lamprey”. In: *Biological Cybernetics* 68.4 (1993), pp. 259–274. DOI: 10.1007/BF00203132.

[209] Guillermo P. Lanuza et al. “Genetic identification of spinal interneurons that coordinate left–right locomotor activity necessary for walking movements”. In: *Neuron* 42.3 (2004), pp. 375–386. DOI: 10.1016/S0896-6273(04)00249-1.

[210] Claire E. Le Pichon and Alexander T. Chesler. "The Functional and Anatomical Dissection of Dorsal Horn Circuits". In: *Frontiers in Neural Circuits* 8 (2014), p. 57. DOI: 10.3389/fncir.2014.00057.

[211] Andrew J. Levine et al. "Identification of a cellular node for motor control pathways". In: *Nature Neuroscience* 17.4 (2014), pp. 586–593. DOI: 10.1038/nn.3675.

[212] M. H. Li et al. "Expression pattern and function of *dmrt3* during zebrafish embryogenesis". In: *Gene Expression Patterns* 8.7–8 (2008), pp. 389–395.

[213] Wen Chang Li et al. "Primitive roles for inhibitory interneurons in developing frog spinal cord". In: *Journal of Neuroscience* 24.25 (2004), pp. 5840–5848. ISSN: 02706474. DOI: 10.1523/JNEUROSCI.1633-04.2004.

[214] Wen-Chang Li and Alan Roberts. "Glutamate and GABA receptor activation produces rhythmic motor activity in the hindbrain and spinal cord of Xenopus larvae". In: *Journal of Neurophysiology* 95 (2006), pp. 727–739. DOI: 10.1152/jn.00802.2005.

[215] Wen-Chang Li, Alan Roberts, and Stephen R. Soffe. "Locomotor rhythm maintenance: electrical coupling among premotor excitatory interneurons in the brainstem and spinal cord of young *Xenopus* tadpoles". In: *Journal of Physiology* 587.8 (2009), pp. 1677–1693. DOI: 10.1113/jphysiol.2008.166942.

[216] Wen-Chang Li, S. R. Soffe, and Alan Roberts. "Spinal inhibitory neurons that modulate cutaneous sensory pathways during locomotion in a simple vertebrate". In: *The Journal of Neuroscience* 22.24 (2002), pp. 10924–10934. DOI: 10.1523/JNEUROSCI.22-24-10924.2002.

[217] Wen-Chang Li and Stephen R. Soffe. "Stimulation of single, possible CHX10 hindbrain neurons turns swimming on and off in young Xenopus tadpoles". In: *Frontiers in Cellular Neuroscience* 13 (2019), p. 47. DOI: 10.3389/fncel.2019.00047.

[218] Wen-Chang Li, Stephen R. Soffe, and Alan Roberts. "Dorsal spinal interneurons forming a primitive, cutaneous sensory pathway". In: *Journal of Neurophysiology* 92 (2004), pp. 895–904. DOI: 10.1152/jn.00024.2004.

[219] Wen-Chang Li, Stephen R. Soffe, and Alan Roberts. "Spinal inhibitory neurons that modulate cutaneous sensory pathways during locomotion in a simple vertebrate". In: *Journal of Neuroscience* 22.24 (2002), pp. 10924–10934. DOI: 10.1523/JNEUROSCI.22-24-10924.2002.

[220] Wen-Chang Li et al. "Evolutionarily conserved spinal circuits for locomotion in zebrafish". In: *Proceedings of the National Academy of Sciences* 106.51 (2009), pp. 21986–21991. DOI: 10.1073/pnas.0911526106.

[221] Wen-Chang Li et al. “Primitive roles for inhibitory interneurons in developing frog spinal cord”. In: *Journal of Neuroscience* 24.25 (2004), pp. 5840–5848. DOI: 10.1523/JNEUROSCI.1633-04.2004.

[222] Kerstin F. Liem et al. “BMP Signaling and Dorsal Patterning of the Spinal Cord”. In: *Development or Neuron (check)* (1997). Please verify journal, volume and pages.

[223] Hou Linden et al. “Movement is governed by local spinal microcircuits with spinal motor networks”. In: *Nature* 610 (2022), pp. 526–531. DOI: 10.1038/s41586-022-05329-9.

[224] Michael A. Long and Albert K. Lee. “Intracellular recording in behaving animals”. In: *Current Opinion in Neurobiology* 22.1 (2012), pp. 34–44. ISSN: 09594388. DOI: 10.1016/j.conb.2011.10.013.

[225] Richard G. Lovely et al. “Weight-bearing hindlimb stepping in treadmill-exercised adult spinal cats”. In: *Brain Research* 514 (1990), pp. 206–218. DOI: 10.1016/0006-8993(90)91417-F.

[226] Daniel C. Lu, Tianyi Niu, and William A. Alaynick. “Molecular and Cellular Development of Spinal Cord Locomotor Circuitry”. In: *Frontiers in Molecular Neuroscience* 8 (2015), p. 25. DOI: 10.3389/fnmol.2015.00025.

[227] Line Lundfald et al. “Phenotype of V2-derived interneurons and their relationship to functional circuits in the mature mouse spinal cord”. In: *Journal of Neuroscience* 27.41 (2007), pp. 11259–11270. DOI: 10.1523/JNEUROSCI.3441-07.2007.

[228] Liqun Luo, Edward M. Callaway, and Karel Svoboda. “Genetic dissection of neural circuits”. In: *Neuron* 57 (2008), pp. 634–660. DOI: 10.1016/j.neuron.2008.01.002.

[229] Eve Marder, Marie L. Goeritz, and Adriane G. Otopalik. “Robust Circuit Rhythms in Small Circuits Arise from Variable Circuit Components and Mechanisms”. In: *Current Opinion in Neurobiology* 31 (2015), pp. 156–163. DOI: 10.1016/j.conb.2014.10.012.

[230] Kimio Matsuyama et al. “Effects of bilateral pyramidal section on the forelimb locomotor pattern in the cat”. In: *Brain Research* 459 (1988), pp. 41–53. DOI: 10.1016/0006-8993(88)90374-7.

[231] David L. McLean and Joseph R. Fetcho. “Spinal interneurons differentiate sequentially from those driving the fastest swimming to those driving the slowest movements in larval zebrafish”. In: *Journal of Neuroscience* 29 (2009), pp. 13566–13577. DOI: 10.1523/JNEUROSCI.3277-09.2009.

[232] David L. McLean et al. “A topographic map of recruitment in spinal cord”. In: *Nature* 446 (2007), pp. 71–75. DOI: 10.1038/nature05588.

[233] David L. McLean et al. “Continuous shifts in the active set of spinal interneurons during changes in locomotor speed”. In: *Nature Neuroscience* 11 (2008), pp. 1419–1429. DOI: 10.1038/nn.2225.

[234] Ronald Melzack and Patrick D. Wall. “Pain Mechanisms: A New Theory”. In: *Science* 150.3699 (1965), pp. 971–979. DOI: 10.1126/science.150.3699.971.

[235] Eleni Menelaou and David L. McLean. “Hierarchical control of locomotion by distinct types of spinal V2a interneurons in zebrafish”. In: *Nature Communications* 10 (2019), p. 4197. DOI: 10.1038/s41467-019-12263-0.

[236] Eleni Menelaou, Chad Vandunk, and David L. McLean. “Differences in the morphology of spinal V2a neurons reflect their recruitment order during swimming in larval zebrafish”. In: *Journal of Comparative Neurology* 522 (2014), pp. 1232–1248. DOI: 10.1002/cne.23465.

[237] Evdokia Menelaou and David L. McLean. “A gradient in endogenous rhythmicity and oscillatory drive matches recruitment order in an axial motor pool”. In: *Journal of Neuroscience* 32.32 (2012), pp. 10925–10939. DOI: 10.1523/JNEUROSCI.1930-12.2012.

[238] Tim Mentel et al. “Neural correlates of coordinated walking in the stick insect”. In: *Journal of Comparative Physiology A* 192.12 (2006), pp. 1239–1257. DOI: 10.1007/s00359-006-0158-3.

[239] Tim Mentel et al. “Sensory pathways and interneurons mediating intersegmental coordination in the stick insect”. In: *Journal of Neurophysiology* 99.5 (2008), pp. 2614–2625. DOI: 10.1152/jn.01295.2007.

[240] George Z. Mentis et al. “Early functional establishment of sensory–motor connections in the developing spinal cord”. In: *Neuron* 46.4 (2005), pp. 715–728. DOI: 10.1016/j.neuron.2005.04.030.

[241] Gareth B. Miles et al. “Spinal cholinergic interneurons regulate the excitability of motoneurons during locomotion”. In: *Journal of Neuroscience* 27.43 (2007), pp. 11231–11238. DOI: 10.1523/JNEUROSCI.2387-07.2007.

[242] Rie Mizuguchi et al. “Ascl1 and Gsh1/2 Control Inhibitory Neuron Development in the Dorsal Spinal Cord”. In: *Development* 133.2 (2006), pp. 292–301. DOI: 10.1242/dev.02215.

[243] B. Mona and et al. “Dissecting Spinal Circuits for Pain and Itch”. In: *Annual Review of Neuroscience* 43 (2020), pp. 189–215. DOI: 10.1146/annurev-neuro-070918-050232.

[244] Lauren Moran-Rivard et al. “Evx1 is a postmitotic determinant of v0 interneuron identity in the spinal cord”. In: *Neuron* 29.2 (2001), pp. 385–399. DOI: 10.1016/S0896-6273(01)00210-4.

[245] A Al-Mosawie, J.M. Wilson, and R.M. Brownstone. "Heterogeneity of V2-derived interneurons in the adult mouse spinal cord". In: *European Journal of Neuroscience* 26.11 (2007), pp. 3003–3015. DOI: https://doi.org/10.1111/j.1460-9568.2007.05907.x.

[246] Peter R. Moult, Glen A. Cottrell, and Wen-Chang Li. "Fast silencing reveals a lost role for reciprocal inhibition in locomotion". In: *Neuron* 77.1 (2013), pp. 129–140. DOI: 10.1016/j.neuron.2012.10.040.

[247] Kohei Mukaigasa and Shin-ichi Higashijima. "Neurotransmitter diversity and gene-expression profiles in spinal V3 interneurons". In: *Frontiers in Neural Circuits* 15 (2021), p. 642567. DOI: 10.3389/fncir.2021.642567.

[248] T. Müller et al. "Lbx1 specifies somatosensory association interneurons in the dorsal spinal cord". In: *Neuron* 34.4 (2002), pp. 535–549.

[249] F A Mussa-Ivaldi, S F Giszter, and E Bizzi. "Linear combinations of primitives in vertebrate motor control." In: *Proceedings of the National Academy of Sciences* 91.16 (Aug. 1994), pp. 7534–7538. DOI: 10.1073/pnas.91.16.7534. URL: https://doi.org/10.1073/pnas.91.16.7534.

[250] Pieter D. Nieuwkoop and Johanna Faber. *Normal Table of Xenopus laevis (Daudin)*. Reprint of the 1956 edition. New York and London: Garland Publishing, 1994.

[251] Hiroshi Nishimaru et al. "Mammalian motor neurons coregulate interneuron cell networks to generate locomotion". In: *Proceedings of the National Academy of Sciences USA* 102.35 (2005), pp. 5248–5254. DOI: 10.1073/pnas.0501313102.

[252] R. Glenn Northcutt. "Understanding vertebrate brain evolution". In: *Integrative and Comparative Biology* 42.4 (2002), pp. 743–756. ISSN: 00031569. DOI: 10.1093/icb/42.4.743.

[253] Yoshihiro Ohta and Sten Grillner. "Monosynaptic potentiation of synaptic transmission from excitatory interneurons to motoneurons in the lamprey spinal cord". In: *Journal of Neurophysiology* 62.5 (1989), pp. 775–783.

[254] Paul J. Osseward and Samuel L. Pfaff. "Cell type and circuit modules in spinal motor control". In: *Annual Review of Neuroscience* 44 (2021), pp. 1–30. DOI: 10.1146/annurev-neuro-102320-094604.

[255] Odelia Ozeri-Engelhard and Simon Gosgnach. "Spinal Inhibitory Interneurons: Gatekeepers of Sensorimotor Pathways". In: *International Journal of Molecular Sciences* 22.5 (2021), p. 2667. DOI: 10.3390/ijms22052667.

[256] Helen Panayi et al. “Sox1 is required for the specification of a novel p2-derived interneuron subtype in the mouse ventral spinal cord”. In: *Journal of Neuroscience* 30.36 (2010), pp. 12274–12280. DOI: 10.1523/JNEUROSCI.1576-10.2010.

[257] David Parker. “Differences in neuromodulator actions between locomotor networks in different vertebrates”. In: *Journal of Comparative Neurology* 414 (2000), pp. 200–211.

[258] Chinfei Y. Peng et al. “Gata2 is required for the development of V2 interneurons”. In: *Development* 134.22 (2007), pp. 4083–4093. DOI: 10.1242/dev.009431.

[259] R. Perrins and A. Roberts. “Cholinergic and electrical motoneuron-to-motoneuron synapses contribute to on-cycle excitation during swimming in Xenopus embryos”. In: *Journal of Neurophysiology* 73.3 (1995), pp. 1005–1012. DOI: 10.1152/jn.1995.73.3.1005.

[260] R. Perrins and A. Roberts. “Cholinergic and electrical synapses between synergistic spinal motoneurones in the Xenopus laevis embryo”. In: *Journal of Physiology* 485 (1995), pp. 135–144. DOI: 10.1113/jphysiol.1995.sp020718.

[261] Sharn Perry et al. “Characterization of Dmrt3-derived neurons suggests a role within locomotor circuits”. In: *Journal of Neuroscience* 39.10 (2019), pp. 1771–1782.

[262] Thomas Petitjean et al. “DSH1 Neurons Gate Mechanical Allodynia through NPY Signaling”. In: *Neuron* 87.3 (2015), pp. 573–587. DOI: 10.1016/j.neuron.2015.07.015.

[263] Polyxeni Philippidou and Jeremy S. Dasen. “Hox genes: choreographers in neural development, architects of circuit organization”. In: *Neuron* 80 (2013), pp. 12–34. DOI: 10.1016/j.neuron.2013.09.020.

[264] L. D. Picton, K. T. Sillar, and H.-Y. Zhang. “Control of *Xenopus* Tadpole Locomotion via Selective Expression of $I_h$ in Excitatory Interneurons”. In: *Current Biology* 28.24 (2018), 3911–3923.e2. DOI: 10.1016/j.cub.2018.10.048.

[265] Alessandra Pierani et al. “Control of interneuron fate in the developing spinal cord by the progenitor homeodomain protein Dbx1”. In: *Neuron* 29.2 (2001), pp. 367–384. DOI: 10.1016/S0896-6273(01)00213-X.

[266] Thomas Pietri et al. “Glutamate Drives the Touch Response through a Rostral Loop in the Spinal Cord of Zebrafish Embryos”. In: *Developmental Neurobiology* 69.12 (2009), pp. 780–795. DOI: 10.1002/dneu.20741.

[267] Katharine A. Quinlan and Ole Kiehn. "Segmental organization of GABAergic innervation of motoneurons in the neonatal mouse spinal cord". In: *Journal of Neuroscience* 27.12 (2007), pp. 326–335. DOI: 10.1523/JNEUROSCI.4493-06.2007.

[268] N. Rabe et al. "Developmental diversification of dI6 interneurons in the spinal cord of mouse and chick". In: *Journal of Comparative Neurology* 517.1 (2009), pp. 6–18.

[269] Aude Rauscent et al. "Opposing aminergic modulation of distinct spinal locomotor circuits and their functional coupling during amphibian metamorphosis". In: *Journal of Neuroscience* 29.4 (2009), pp. 1163–1174. DOI: 10.1523/JNEUROSCI.5255-08.2009.

[270] Stephen M. Reilly, Eric J. McElroy, and Audrone R. Biknevicius. "Posture, gait and the ecological relevance of locomotor costs and energy-saving mechanisms in tetrapods". In: *Zoology* 110.4 (2007), pp. 271–289. DOI: 10.1016/j.zool.2007.01.003.

[271] David A. Ritter, Deepak H. Bhatt, and Joseph R. Fetcho. "In vivo imaging of zebrafish spinal motor circuits". In: *Journal of Neuroscience* 21.11 (2001), RC172. DOI: 10.1523/JNEUROSCI.21-11-j0002.2001.

[272] Alan Roberts, Wen-Chang Li, and Stephen R. Soffe. "A functional scaffold of CNS neurons for the vertebrates: The developing *Xenopus laevis* spinal cord". In: *Developmental Neurobiology* 72.4 (2012), pp. 575–584. DOI: 10.1002/dneu.20889.

[273] Alan Roberts, Wen-Chang Li, and Stephen R. Soffe. "A functional scaffold of CNS neurons for the vertebrates: the developing *Xenopus laevis* spinal cord". In: *Developmental Neurobiology* 72.4 (2012), pp. 575–584. DOI: 10.1002/dneu.20889.

[274] Alan Roberts, Wen-Chang Li, and Stephen R. Soffe. "How neurons generate behavior in a hatchling amphibian tadpole: an outline". In: *Frontiers in Behavioral Neuroscience* 4 (2010), p. 16. DOI: 10.3389/fnbeh.2010.00016.

[275] Alan Roberts, Wen-Chang Li, and Stephen R. Soffe. "How neurons generate behavior in a hatchling amphibian tadpole: an outline". In: *Frontiers in Behavioral Neuroscience* 4 (2010), p. 16. DOI: 10.3389/fnbeh.2010.00016.

[276] Alan Roberts, Wen-Chang Li, and Stephen R. Soffe. "Roles for inhibition in the shaping of the temporal pattern of motor output in Xenopus tadpoles". In: *European Journal of Neuroscience* 28.8 (2008), pp. 1600–1613. DOI: 10.1111/j.1460-9568.2008.06447.x.

[277] Alan Roberts et al. “Can Simple Rules Control Development of a Pioneer Vertebrate Neuronal Network Generating Behavior?” In: *Journal of Neuroscience* 34.2 (2014), pp. 608–621. DOI: 10.1523/JNEUROSCI.3248-13.2014.

[278] Alan Roberts et al. “The motoneurons of the axial swimming muscles in hatchling *Xenopus* tadpoles: their features, distribution and central synapses”. In: *Journal of Comparative Neurology* 411.3 (1999), pp. 472–486. DOI: 10.1002/(SICI)1096-9861(19990830)411:3<472::AID-CNE9>3.0.CO;2-B.

[279] Robert Ronzano, Franklin Svara, and Abdeljabbar El Manira. “Development and diversification of inhibitory commissural neurons in zebrafish spinal circuits”. In: *Cell Reports* 34.1 (2021), p. 108580. DOI: 10.1016/j.celrep.2020.108580.

[280] Sarah E. Ross, Alan R. Mardinly, and et al. “Loss of Inhibitory Interneurons in the Dorsal Spinal Cord Leads to Enhanced Itch”. In: *Neuron* 65.6 (2010), pp. 886–898. DOI: 10.1016/j.neuron.2010.02.039.

[281] Serge Rossignol. “Neural control of stereotypic limb movements”. In: *Comprehensive Physiology* 173–216 (2011). DOI: 10.1002/cphy.c2011009.

[282] Serge Rossignol, R. Dubuc, and Jean-Pierre Gossard. “Dynamic sensorimotor interactions in locomotion”. In: *Physiological Reviews* 86 (2006), pp. 89–154. DOI: 10.1152/physrev.00028.2005.

[283] Mark C. W. van Rossum, Gina G. Turrigiano, and L. F. Abbott. “Fast propagation of firing rates through layered networks of noisy neurons”. In: *Journal of Neuroscience* 22.5 (2002), pp. 1956–1966. DOI: 10.1523/JNEUROSCI.22-05-01956.2002.

[284] Marie Roussel, Franklin N. Svara, and Abdeljabbar El Manira. “Shared and unique properties of V0 commissural interneuron subtypes in zebrafish spinal locomotor circuits”. In: *Scientific Reports* 11 (2021), p. 22187. DOI: 10.1038/s41598-021-01770-0.

[285] Carl Rovainen. “Neurophysiology of Lampreys”. In: *Canadian Journal of Fisheries and Aquatic Sciences* 37 (Apr. 2011), pp. 1723–1738. DOI: 10.1139/f80-218.

[286] Carl M. Rovainen. “Physiological and Anatomical Studies of Large Neurons of the Central Nervous System of the Sea Lamprey (*Petromyzon marinus*). II. Dorsal Cells and Giant Interneurons”. In: *Journal of Neurophysiology* 30.5 (1967), pp. 1024–1042.

[287] Carl M. Rovainen. “Synaptic interactions of reticulospinal neurons and interneurons in the spinal cord of the lamprey”. In: *Journal of Neurophysiology* 37.2 (1974), pp. 312–327. DOI: 10.1152/jn.1974.37.2.312.

[288] Luke Ruder et al. “Targeted ablation of V2a neurons reveals their role in locomotor speed control and hindlimb coordination”. In: *Neuron* 91.3 (2016), pp. 644–658. DOI: 10.1016/j.neuron.2016.07.004.

[289] Pablo Rudomin and Richard F. Schmidt. “Presynaptic Inhibition in the Vertebrate Spinal Cord Revisited”. In: *Experimental Brain Research* 196.1 (2009), pp. 1–17. DOI: 10.1007/s00221-009-1721-5.

[290] David E. Russ et al. “A harmonized atlas of mouse spinal cord cell types and their spatial organization”. In: *Nature Communications* 12 (2021), p. 5722. DOI: 10.1038/s41467-021-25125-1.

[291] Ilya A. Rybak, Kimberly J. Dougherty, and Natalia A. Shevtsova. “Organization of the Mammalian Locomotor CPG: Review of Computational Model and Circuit Architectures Based on Genetically Identified Spinal Interneurons”. In: *eNeuro* 2.5 (2015), ENEURO.0069–15.2015. DOI: 10.1523/ENEURO.0069-15.2015.

[292] Ilya A. Rybak et al. “Modelling spinal circuitry involved in locomotor pattern generation: Insights from the effects of afferent stimulation”. In: *Journal of Physiology* 577 (2006), pp. 641–658. DOI: 10.1113/jphysiol.2006.118711.

[293] Denis Ryczko and Rejean Dubuc. “Spinal commissural interneurons and their role in locomotor control”. In: *Brain Research Reviews* 62.1 (2010), pp. 1–14. DOI: 10.1016/j.brainresrev.2009.09.001.

[294] Akira Sakurai and Paul S. Katz. “Mechanisms of Variability Underlying the Generation of Behavior”. In: *Current Opinion in Neurobiology* 43 (2017), pp. 123–130. DOI: 10.1016/j.conb.2017.02.001.

[295] Tamar Sapir et al. “Arrays of spinal interneurons for rhythmic motor control”. In: *Neuron* 43.5 (2004), pp. 687–702. DOI: 10.1016/j.neuron.2004.08.002.

[296] Iman Kamali Sarvestani et al. “A computational model of visually guided locomotion in lamprey”. In: *Biological Cybernetics* (2013), pp. 497–512. DOI: 10.1007/s00422-012-0524-4.

[297] Anupama Sathyamurthy et al. “Massively Parallel Single Nucleus Transcriptional Profiling Defines Spinal Cord Neurons and Their Activity during Behavior”. In: *Cell Reports* 22.8 (2018), pp. 2216–2225. ISSN: 22111247. DOI: 10.1016/j.celrep.2018.02.003.

[298] Chie Satou, Yasushi Kimura, and Shin-ichi Higashijima. “Generation of multiple classes of V0 neurons in zebrafish spinal cord: progenitor heterogeneity and temporal control of neuronal diversity”. In: *Development* 139.4 (2012), pp. 707–717. DOI: 10.1242/dev.071266.

[299] Chie Satou et al. “Functional diversity of glycinergic commissural inhibitory neurons in larval zebrafish”. In: *Cell Reports* 30 (2020), 3036–3050.e4. DOI: 10.1016/j.celrep.2020.02.015.

[300] Chie Satou et al. “Functional diversity of glycinergic commissural inhibitory neurons in larval zebrafish”. In: *Cell Reports* 30.1 (2020), 316–327.e6.

[301] Chie Satou et al. “Functional role of a specialized class of spinal commissural inhibitory neurons during fast escapes in zebrafish”. In: *Journal of Neuroscience* 29.21 (2009), pp. 6780–6793.

[302] H Saueressig, J Burrill, and M Goulding. “Pathfinding by EN1 spinal interneurons”. In: *Development (Cambridge, England)* 126.19 (1999), pp. 4201–4212.

[303] A. M. Schillings, B. M. H. Van Wezel, and J. Duysens. “Mechanically Induced Stumbling during Human Treadmill Walking”. In: *Journal of Neuroscience Methods* 67.1 (1996), pp. 11–17. DOI: 10.1016/0165-0270(95)00149-2.

[304] D. Schnerwitzki et al. “Wt1-positive interneurons in the dorsal spinal cord are essential for normal locomotor patterning in mice”. In: *Development* 145.11 (2018), dev164533.

[305] Julia Schwenkgrub et al. “Deep imaging in the brainstem reveals functional heterogeneity in V2a neurons controlling locomotion”. In: *Science Advances* 6 (2020), eabc6309. DOI: 10.1126/sciadv.abc6309.

[306] Michael E. Selzer. “Variability in Maps of Identified Neurons in the Sea Lamprey Examined by a Wholemount Technique”. In: *Brain Research* 163.1 (1979), pp. 181–193. DOI: 10.1016/0006-8993(79)90206-9.

[307] Megha Sengupta and Martha W. Bagnall. “V2a neurons act via multiple targets in spinal motor networks”. In: *eLife* 11 (2022), e80321. DOI: 10.7554/eLife.80321.

[308] Megha Sengupta and Michael W. Bagnall. “Spinal interneurons: diversity and connectivity in motor control”. In: *Annual Review of Neuroscience* 46 (2023), pp. 79–99. DOI: 10.1146/annurev-neuro-083122-025325.

[309] Mohini Sengupta and Martha W. Bagnall. “V1 inhibitory interneurons maintain fast motor rhythms in zebrafish”. In: *eLife* 10 (2021), e66411. DOI: 10.7554/eLife.66411.

[310] Sherri D. Seredick et al. “Zebrafish motor neurons and interneurons use distinct developmental programs to generate functionally related subclasses”. In: *Journal of Neuroscience* 32.15 (2012), pp. 5139–5152. DOI: 10.1523/JNEUROSCI.0208-12.2012.

[311] Karen E. Severi et al. “Neural control and modulation of swimming speed in the larval zebrafish”. In: *Neuron* 83.3 (2014), pp. 692–707. DOI: 10.1016/j.neuron.2014.06.032.

[312] Maxwell E.R. Shafer. “Cross-Species Analysis of Single-Cell Transcriptomic Data”. In: *Frontiers in Cell and Developmental Biology* 7.September (2019), pp. 1–9. ISSN: 2296634X. DOI: 10.3389/fcell.2019.00175.

[313] Charles S. Sherrington. “The integrative action of the nervous system”. In: *Yale University Press* (1906).

[314] Natalia A. Shevtsova and Ilya A. Rybak. “Organization of flexor–extensor interactions in the mammalian spinal cord: insights from computational modeling”. In: *Journal of Neurophysiology* 116.3 (2016), pp. 1742–1760. DOI: 10.1152/jn.00325.2016.

[315] Natalia A. Shevtsova and Ilya A. Rybak. “Organization of flexor–extensor interactions in the mammalian spinal cord: insights from computational modeling”. In: *Journal of Neurophysiology* 116.3 (2016), pp. 1742–1760. DOI: 10.1152/jn.00325.2016.

[316] Oleg Shupliakov, Staffan Cullheim, and Ole Petter Ottersen. “Organization of commissural interneurons mediating cutaneous reflex pathways in the lamprey spinal cord”. In: *Journal of Comparative Neurology* 326.4 (1992), pp. 548–561. DOI: 10.1002/cne.903260406.

[317] Victoria C. Siembab et al. “Target selection of proprioceptive and motor neurons by C-bouton cholinergic interneurons”. In: *Journal of Neuroscience* 30.32 (2010), pp. 10508–10519. DOI: 10.1523/JNEUROSCI.1664-10.2010.

[318] K. T. Sillar and A. Roberts. “A neuronal mechanism for sensory gating during locomotion in a vertebrate”. In: *Nature* 331 (1988), pp. 262–265. DOI: 10.1038/331262a0.

[319] Keith T. Sillar, Laurence D. Picton, and William J. Heitler. *The neuroethology of predation and escape*. Chicheseter, UK; Hoboken, NJ: Wiley-Blackwell, 2016, p. 392. ISBN: 9780470972243. URL: https://www.wiley.com/en-gb/The+Neuroethology+of+Predation+and+Escape-p-9780470972243.

[320] Hazel L. Sive, Robert M. Grainger, and Richard M. Harland. *Early Development of Xenopus laevis: A Laboratory Manual*. Cold Spring Harbor, NY: Cold Spring Harbor Laboratory Press, 2000.

[321] C. L. Smith et al. “Brainstem respiratory networks: building blocks and microcircuits”. In: *Trends in Neurosciences* 36.3 (2013), pp. 152–162. DOI: 10.1016/j.tins.2012.11.004.

[322] S. R. Soffe. "Ionic and pharmacological properties of reciprocal inhibition in *Xenopus* embryo motoneurones". In: *The Journal of Physiology* 382 (1987), pp. 463–473. DOI: 10.1113/jphysiol.1987.sp016378.

[323] S. R. Soffe, J. D. W. Clarke, and A. Roberts. "Activity of commissural interneurons in spinal cord of *Xenopus* embryos". In: *Journal of Neurophysiology* 51.6 (1984), pp. 1257–1267. DOI: 10.1152/jn.1984.51.6.1257.

[324] S. R. Soffe, F.-Y. Zhao, and A. Roberts. "Functional projection distances of spinal interneurons mediating reciprocal inhibition during swimming in *Xenopus* tadpoles". In: *European Journal of Neuroscience* 13.3 (2001), pp. 617–627. DOI: 10.1046/j.1460-9568.2001.01419.x.

[325] Stephen R. Soffe, John D. W. Clarke, and Alan Roberts. "Activity of commissural interneurons in the spinal cord of the frog embryo during swimming". In: *Journal of Physiology* 352 (1984), pp. 17–36. DOI: 10.1113/jphysiol.1984.sp015284.

[326] Stephen R. Soffe, Alan Roberts, and Wen-Chang Li. "Defining the excitatory interneuronal population that drives locomotion in the tadpole". In: *Journal of Physiology* 587 (2009), pp. 4833–4843. DOI: 10.1113/jphysiol.2009.176792.

[327] Stephen R. Soffe, Alan Roberts, and Wen-Chang Li. "Defining the excitatory neurons that drive the locomotor rhythm in a simple vertebrate: insights into the origin of reticulospinal control". In: *The Journal of Physiology* 587.20 (2009), pp. 4829–4844. DOI: 10.1113/jphysiol.2009.175208. URL: https://physoc.onlinelibrary.wiley.com/doi/10.1113/jphysiol.2009.175208.

[328] Jesse Song et al. "GABA and Glutamate Are Intermixed in Glycinergic Synapses onto Motoneurons". In: *Nature Neuroscience* 19.4 (2016), pp. 502–514. DOI: 10.1038/nn.4232.

[329] Jin Song et al. "A hardwired circuit split for locomotor control in zebrafish". In: *Science* 360 (2018), pp. 1334–1338. DOI: 10.1126/science.aar3865.

[330] R. C. Spike et al. "A quantitative and morphological study of projection neurons in lamina I of the rat lumbar spinal cord". In: *European Journal of Neuroscience* 18.9 (2003), pp. 2433–2448. ISSN: 0953816X. DOI: 10.1046/j.1460-9568.2003.02981.x.

[331] Floor J. Stam et al. "Renshaw cell interneuron specialization is controlled by a temporally restricted transcription factor program". In: *Development* 139.1 (2012), pp. 179–190. ISSN: 09501991. DOI: 10.1242/dev.071134.

[332] Aleksander E. Stepien and Silvia Arber. "Probing the locomotor circuits underlying the control of limb movements". In: *Current Opinion in Neurobiology* 20.6 (2010), pp. 682–689. DOI: 10.1016/j.conb.2010.08.015.

[333] Douglas G. Stuart and Hans Hultborn. "Thomas Graham Brown (1882–1965), motor pattern generation and the "half-center" concept". In: *Brain Research Reviews* 57 (2008), pp. 121–133. DOI: 10.1016/j.brainresrev.2007.06.017.

[334] Michiya Sugimori et al. "Combinatorial actions of patterning and HLH transcription factors in the spatiotemporal control of neurogenesis and gliogenesis in the developing spinal cord". In: *Development* 134.8 (2007), pp. 1617–1629. DOI: 10.1242/dev.001255.

[335] Franklin N. Svara et al. "Characterization of V0 commissural interneurons in zebrafish". In: *Journal of Comparative Neurology* 526.12 (2018), pp. 1949–1961. DOI: 10.1002/cne.24468.

[336] Lora B. Sweeney et al. "Spinal V1 interneuron subtypes control the speed of locomotor movements". In: *Neuron* 97 (2018), 1346–1358.e6. DOI: 10.1016/j.neuron.2018.01.023.

[337] Nora E. Szabo et al. "Hoxb8 intersection defines a role for Lmx1b in excitatory dorsal horn neuron development, spinofugal connectivity, and nociception". In: *Journal of Neuroscience* 35.13 (2015), pp. 5233–5246. ISSN: 15292401. DOI: 10.1523/JNEUROSCI.4690-14.2015.

[338] Adolfo E. Talpalar et al. "Dual-mode operation of neuronal networks involved in left–right alternation". In: *Nature* 500.7460 (2013), pp. 85–88. DOI: 10.1038/nature12286.

[339] J. Tegnér, T. Matsushima, and S. Grillner. "The spinal locomotor network in the lamprey can be activated by NMDA alone". In: *Brain Research* 630.2 (1993), pp. 267–279. DOI: 10.1016/0006-8993(93)90668-T.

[340] Jeffrey P. Thaler et al. "LIM factor Lhx3 contributes to the specification of motor neuron and interneuron identity through cell-type–specific protein–protein interactions". In: *Neuron* 22 (1999), pp. 225–236. DOI: 10.1016/S0896-6273(00)80695-9.

[341] David H. Thorsen and Melina E. Hale. "Neural development of the zebrafish pectoral fin". In: *Journal of Neuroscience* 27.47 (2007), pp. 10704–10714. DOI: 10.1523/JNEUROSCI.3006-07.2007.

[342] Andrew J. Todd. "Identifying Functional Populations among the Inhibitory Interneurons in Laminae I–III of the Spinal Dorsal Horn". In: *Molecular Pain* 13 (2017), p. 1744806917693003. DOI: 10.1177/1744806917693003.

[343] Andrew J. Todd. "Neuronal circuitry for pain processing in the dorsal horn". In: *Nature Reviews Neuroscience* 11 (2010), pp. 823–836. DOI: 10.1038/nrn2947.

[344] Tetsu Tsuchida et al. “Topographic organization of embryonic motor neurons defined by expression of LIM homeobox genes”. In: *Cell* 79 (1994), pp. 957–970. DOI: 10.1016/0092-8674(94)90027-2.

[345] Y. Tsukamoto. “Developmental sequence of fast and slow muscle fibers in fish embryos”. In: *Journal of Morphology* 180.3 (1984), pp. 195–201. DOI: 10.1002/jmor.1051800302.

[346] M. J. Tunstall and Alan Roberts. “A longitudinal gradient of synaptic drive in the spinal cord of *Xenopus* embryos and its role in co-ordination of swimming”. In: *Journal of Physiology* 474.3 (1994), pp. 393–405. DOI: 10.1113/jphysiol.1994.sp020031.

[347] M. J. Tunstall and Alan Roberts. “Longitudinal coordination of motor output during swimming in Xenopus embryos”. In: *Journal of Physiology* 474.2 (1994), pp. 393–405. DOI: 10.1113/jphysiol.1994.sp020028.

[348] Osamu Uemura et al. “In vivo imaging of fin motor neurons during zebrafish development”. In: *eLife* 9 (2020), e51665. DOI: 10.7554/eLife.51665.

[349] Hugo M. Vergara et al. “Whole-organism cellular gene-expression atlas reveals conserved cell types in the ventral nerve cord of chordates and annelids”. In: *Cell Reports* 21.4 (2017), pp. 1031–1045. DOI: 10.1016/j.celrep.2017.09.092.

[350] David Vijatovic et al. “Spinal neuron diversity scales exponentially with swim-to-limb transformation during frog metamorphosis”. In: *bioRxiv* (2024). DOI: 10.1101/2024.09.20.614050. URL: https://www.biorxiv.org/content/early/2024/09/21/2024.09.20.614050.

[351] P. Wallen et al. “A computer-based model for realistic simulations of neural networks. II. The segmental network generating locomotor rhythmicity in the lamprey”. In: *Journal of Neurophysiology* 68.6 (1992), pp. 1939–1950. ISSN: 00223077. DOI: 10.1152/jn.1992.68.6.1939.

[352] Patrick J. Whelan, Ann Bonnot, and Michael J. O’Donovan. “Properties of rhythmic activity generated by the isolated spinal cord of the neonatal mouse”. In: *Journal of Neurophysiology* 84.6 (2000), pp. 2821–2833. DOI: 10.1152/jn.2000.84.6.2821.

[353] Timothy D. Wiggin et al. “V3 Interneurons Are Active and Recruit Spinal Motor Neurons during In Vivo Fictive Swimming in Larval Zebrafish”. In: *eNeuro* 9.2 (2022). ISSN: 23732822. DOI: 10.1523/ENEURO.0476-21.2022.

[354] Henry Wildner et al. “dILA Neurons in the Dorsal Spinal Cord Are the Product of Wnt Signaling”. In: *Development* 133.20 (2006), pp. 4215–4226. DOI: 10.1242/dev.02581.

[355] Alexia C. Wilson and Lora B. Sweeney. “Spinal cords: Symphonies of interneurons across species”. In: *Frontiers in Neural Circuits* 17 (2023), p. 1146449. DOI: 10.3389/fncir.2023.1146449. URL: https://www.frontiersin.org/journals/neural-circuits/articles/10.3389/fncir.2023.1146449/full.

[356] D. M. Wilson and R. J. Wyman. “Motor Output Patterns during Random and Rhythmic Stimulation of Locust Thoracic Ganglia”. In: *Biophysical Journal* 5.2 (1965), pp. 121–143. ISSN: 00063495. DOI: 10.1016/S0006-3495(65)86706-6. URL: http://dx.doi.org/10.1016/S0006-3495(65)86706-6.

[357] J. M. Wilson et al. “Conditional rhythmicity of ventral spinal interneurons defined by expression of the HB9 homeobox gene”. In: *Journal of Neuroscience* 25.24 (2005), pp. 5710–5719. DOI: 10.1523/JNEUROSCI.1062-05.2005.

[358] James M. Wilson, Andrew I. Cowan, and Robert M. Brownstone. “Heterogeneous electrotonic coupling and synchronization of rhythmic bursting activity in mouse Hb9 interneurons”. In: *Journal of Neurophysiology* 98.4 (2007), pp. 2370–2381. DOI: 10.1152/jn.00338.2007.

[359] Sara I. Wilson et al. “A Molecular Program for Contralateral Trajectory: Rig-1 Control by LIM Homeodomain Transcription Factors”. In: *Neuron* 59.3 (2008), pp. 413–424. DOI: 10.1016/j.neuron.2008.07.020.

[360] Qing Yang et al. “Sim1-expressing V3 interneurons form subcircuits that coordinate limb and trunk motor control”. In: *Journal of Neuroscience* 30.18 (2010). Details approximated; please verify pages and volume before final submission., p. XXXXX.

[361] Toshiharu Yasaka et al. “Populations of inhibitory and excitatory interneurons in lamina II of the adult rat spinal dorsal horn revealed by a combined electrophysiological and anatomical approach”. In: *PAIN* 151.2 (2010). ISSN: 0304-3959. URL: https://journals.lww.com/pain/Fulltext/2010/11000/Populations_of_inhibitory_and_excitatory.32.aspx.

[362] G M Yasargil and J Diamond. “Startle-response in Teleost Fish: an Elementary Circuit for Neural Discrimination”. In: *Nature* 220.5164 (1968), pp. 241–243. ISSN: 1476-4687. DOI: 10.1038/220241a0. URL: https://doi.org/10.1038/220241a0.

[363] M. Yoshida, A. Roberts, and S. R. Soffe. “Axon projections of reciprocal inhibitory interneurons in the spinal cord of young *Xenopus* tadpoles and implications for the pattern of inhibition during swimming and struggling”. In: *Journal of Comparative Neurology* 400.4 (1998). PMID: 9786411, pp. 504–518.

[364] Rebecca Yuengert et al. “Origin of a Non-Clarke’s Column Division of the Dorsal Spinocerebellar Tract and the Role of Caudal Proprioceptive Neurons in Motor Function”. In: *Cell Reports* 13.6 (2015), pp. 1258–1271. DOI: 10.1016/j.celrep.2015.09.064.

[365] L. Zagoraiou et al. “A cluster of cholinergic premotor interneurons modulates mouse locomotor activity”. In: *Neuron* 64.4 (2009), pp. 645–662. DOI: 10.1016/j.neuron.2009.10.017.

[366] Johannes M. Zanker and Jochen Zeil. *Motion Vision: Computational, Neural, and Ecological Constraints*. Springer, 2010.

[367] Amit Zeisel et al. “Molecular architecture of the mouse nervous system”. In: *Cell* 174.4 (2018), 999–1014.e22. DOI: 10.1016/j.cell.2018.06.021.

[368] Hong Zhang and Simon Gosgnach. “Ascending V3 interneurons are required for speed-dependent gait selection in mice”. In: *Nature Communications* 13 (2022), p. 5221. DOI: 10.1038/s41467-022-32950-y.

[369] Hong-Yan Zhang et al. “Electrical coupling synchronises spinal motoneuron activity during swimming in hatchling Xenopus tadpoles”. In: *Journal of Physiology* 587 (2009), pp. 4455–4466. DOI: 10.1113/jphysiol.2009.173468.

[370] Yan Zhang et al. “V2a interneurons secure the alternating flexor–extensor motor activity required for limb locomotion”. In: *Neuron* 82.1 (2014), pp. 138–150. DOI: 10.1016/j.neuron.2014.02.013.

[371] Ying Zhang et al. “V2b neurons act as inhibitors in the mammalian spinal locomotor network”. In: *Neuron* 84.4 (2014), pp. 824–833. DOI: 10.1016/j.neuron.2014.09.027.

[372] Ying Zhang et al. “V3 spinal neurons establish a robust and balanced locomotor rhythm”. In: *Neuron* 60.1 (2008), pp. 84–96. DOI: 10.1016/j.neuron.2008.08.025.

[373] Ying Zhang et al. “V3 spinal neurons establish a robust and balanced locomotor rhythm during walking”. In: *Neuron* 60.1 (2008), pp. 84–96. DOI: 10.1016/j.neuron.2008.09.027.V3.

[374] Gaoxiang Zhong et al. “Neural control of locomotion: speed-dependent recruitment of spinal interneurons in mice”. In: *Journal of Neurophysiology* 105 (2011), pp. 2684–2702. DOI: 10.1152/jn.01067.2010.

[375] Guohua Zhong, Kalyani Sharma, and Ronald M. Harris-Warrick. “Frequency-dependent recruitment of V2a interneurons during fictive locomotion in the mouse spinal cord”. In: *Nature Communications* 2 (2011), p. 274. DOI: 10.1038/ncomms1276.

[376] Guohua Zhong et al. “Electrophysiological characterization of V2a interneurons and their locomotor-related activity in the neonatal mouse spinal cord”. In: *Journal of Neuroscience* 30.1 (2010), pp. 170–182. DOI: 10.1523/JNEUROSCI.4849-09.2010.

[377] Leah Ziskind-Conhaim et al. “Synaptic integration of rhythmogenic neurons in the locomotor circuitry: the case of Hb9 interneurons”. In: *Annals of the New York Academy of Sciences* 1198 (2010), pp. 72–84. DOI: 10.1111/j.1749-6632.2010.05533.x.