Our lab is studying embryonic development of dopaminergic (DA) neurons in the zebrafish brain. Dopaminergic neurons produce the brain chemical dopamine, whose role is to relay information between nerve cells of different brain tissues that control a variety of brain activities including control of movement, neuro-endocrine and cognitive functions. These neurons have been specifically associated with body homeostasis, drug addiction, schizophrenia, Tourette’s syndrome, and Parkinson’s disease (PD). Therefore, uncovering basic mechanisms that govern DA cell fate is relevant for the advancement of therapies aimed to inhibit or prevent DA deterioration in human neurodegenerative diseases. By studying how DA neurons are regulated during normal brain development we may also begin to understand the genetic basis for neuropsychiatric disorders such as schizophrenia and Tourette’s syndrome and find new ways to alleviate these neurological impairments.

However, little is known about the molecules that instruct immature DA neurons to proliferate, survive and innervate their target tissues mainly due to: 1) lack of early markers that are specifically expressed in DA progenitors 2) the difficulty in studying the migration and specification of DA progenitors in mammalian embryos 3) limited knowledge of signals that control progenitor specification. To overcome these hurdles we chose to study DA neural specification by employing zebrafish as a model organism with a relatively simple DA system.

Our main research goal is to identify the assembly of extrinsic and intrinsic signals that govern DA cell fate and organization in the brain. To uniquely address these complex issues we exploit the amenability of zebrafish to genetic manipulations. Zebrafish DA system develops within 1-4 days post fertilization and the organism’s optical transparency allows in vivo analysis of DA neurons and their circuits (Fig. 1). The validity of our model organism to human neurological disorders is supported by reports of a fair degree of conservation in DA circuits across fish, mice and humans. Thus, application of various neurotoxins that are known to impair DA function to goldfish and zebrafish induces a specific reduction in DA cell number and elicits Parkinsonian-like symptoms. Moreover, it has been shown that zebrafish DA neurons that project into the striatum correspond to the mammalian ascending midbrain DA system (Fig. 2).

We have undertaken a genetic approach to study zebrafish mutants that display altered DA phenotypes. For example, the zebrafish mutant too few (tof) fails to develop hypothalamic DA and serotonergic (5HT) neurons. The observed phenotype is highly specific since other groups of DA and 5HT, as well as noradrenergic neurons, are not affected in tof. Using a positional cloning approach, we have identified the tof zebrafish mutant gene, which encodes a forebrain specific zinc finger transcription repressor homologous to the mammalian fezl. We revealed that tof/fezl controls the development of DA neurons by regulating the secretion of an unknown telencephalic signal, which emanates from the telencephalon.

One research direction is to elucidate the unknown physiological signal that is impaired in tof. To this end, we are employing combination of biochemical, functional genomics and genetic methods to seek for genes that are regulated by Fezl (termed ‘Fezl target genes’). A parallel effort is being made to gain information about gene function and gene order in the tof/fezl pathway. This is done by microarray analysis of tof- mediated alteration in gene transcription in Fezl-expressing cells, which are isolated from wild type and tof embryos.

At the cellular level, the interactions between DA neurons and other brain cells are crucial for proper development, maintenance and functionality of these neurons. Moreover, a major part of DA neurogenesis is the formation of axonal trajectories and synapses. To study these aspects of development, we exploit the fact that zebrafish embryos are transparent and develop externally. Thus we use transgenic lines that express fluorescent reporter proteins in DA progenitors to track the lineage of DA cells by real-time imaging and to monitor the formation of DA neural circuits during development.
Using this methodology we aim to expand our earlier efforts to uncover physiological signals that control different aspects of DA specification.

Selected publications

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Fig. 1 An example of a zebrafish embryo stained with an antibody directed against Tyrosine hydroxylase (TH). The antibody detects the following groups of neurons: hypothalamic dopaminergic (Hy DA)-, locus coeruleus (LC) noradrenergic (NA)-, as well as AAC- and gut sympathetic- neurons.

Fig. 2 In zebrafish, the basal (ventral, encircled in blue) and alar (dorsal, encircled in red) diencephalic dopaminergic neurons are considered to be analogous to mammalian hypothalamic A11–A15 and mesencephalic A8-A10 DA neuron groups, respectively.