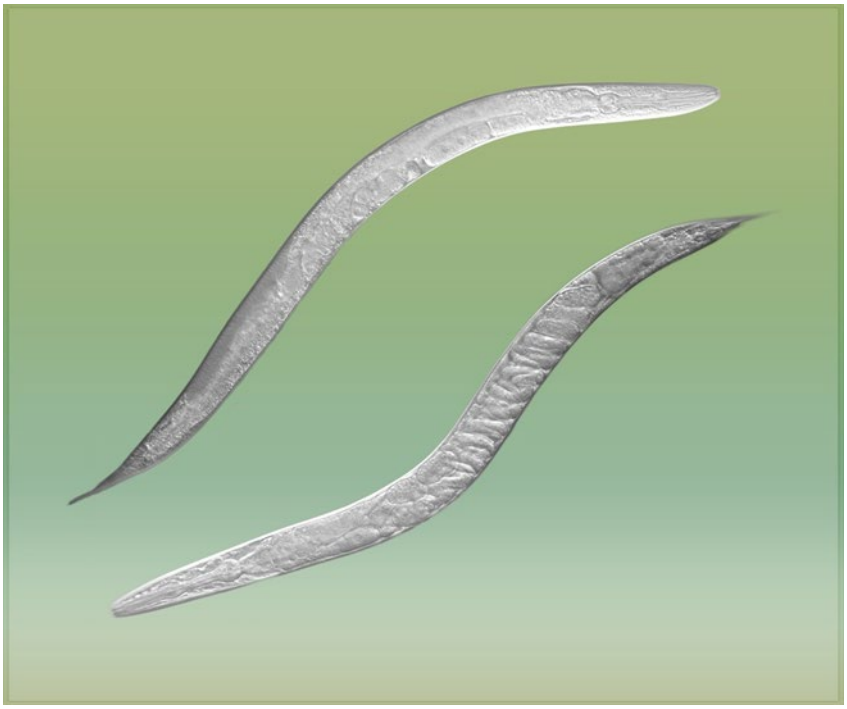
How do nematodes think?

Manuel Zimmer has been a professor of neurobiology at the University of Vienna since October 2018. He and his group are currently still located at the IMP. Zimmer is enthusiastic about the future BioCenter St. Marx and its potential to develop into a unique Lifescience Hub with an international reputation. He is looking forward to cooperations on the multidisciplinary level and expects promising synergies.

How would you briefly outline the fundamental research questions in the neurosciences and in your own work?

The architecture and structure of nervous systems and brains are highly complex and present a major challenge for science. Our approach deliberately focuses on the nematode worm *Caenorhabditis elegans* (*C. elegans*): the idea is to investigate an animal with a relatively small nervous system as a model organism. Nematodes have a very stereotypical bodyplan and have only 302 nerve cells, whereby each exhibits a specific identity (eutely). Moreover, the same nerve cells are in exactly the same position in each and every individual. This enables studying patterns in a reproducible manner and provides an opportunity to draw general conclusions about more complex nervous systems. The basic questions we are pursuing based on *C. elegans* are: How do nervous systems collect information and how are sensory inputs perceived, processed and translated into behaviors?



The Nobel laureate Sydney Brenner established the nematode C. elegans as a model organism in 1960 and studied its organ development and the development of the nervous system.
Picture of C. elegans with the kind permission of Susanne Skora

What does "information processing" by a worm really mean?

The worm can smell, taste, feel and distinguish between light and dark. It can even be considered to be a specialist when it comes to differentiating different odors. As opposed to humans, for example, it can smell oxygen and carbon dioxide. *C. elegans* can also distinguish chemicals and exhibits a well-developed tactile sense. In our research we use oxygen, carbon dioxide and organic scents to stimulate the nematode's sensory nerve cells.

Is it possible to "read the worm's mind"?

The nervous system of *C. elegans* was already "mapped" about 30 years ago using electron microscopy. It is very small but nonetheless quite complex: the nerve cells are linked together in a complicated manner. Although this network map is very useful, it represents a static construct. What has been missing to date is the possibility to study the network dynamics on different levels. We therefore developed a method in which we stain the individual nerve cells with a calcium-dependent pigment (a modified green fluorescent protein). We then use a microscope that has been specially modified to meet our requirements with regard to speed and sensitivity: this enables us to scan the entire nervous system and highlight its neuronal activity (see link to YouTube video on Manuel Zimmer's website). This yields gigabytes of data that are then analyzed using a tailor-made computer program.

The worm's head is immobilized and we then observe its brain and neuronal activities in **real time** under the microscope. This experiment was first successfully conducted in 2013. To date, with the exception of *C. elegans*, neuronal activities across the entire brain have been studied in real time only in zebrafish larvae. Our work with *C. elegans* revealed that most active neurons in the brain (when the worm is awake) coordinate themselves in order to form collective, global activity patterns, even in the absence of sensory stimuli. Brain states with characteristic neuronal activity patterns are cyclically repeated: these patterns are not randomly structured. Nerve cell complexes coordinate and organize themselves into highly complex entities that communicate with each other. The only exception to these dynamic states is when the worm falls asleep. In that case, a large part of the nervous system becomes inactive, with the exception of the so-called sleep neurons. This means we are also investigating the function of sleep in *C. elegans*. A toolbox of computation procedures enables us to visualize and quantify these processes – we are actually "reading the worm's thoughts."

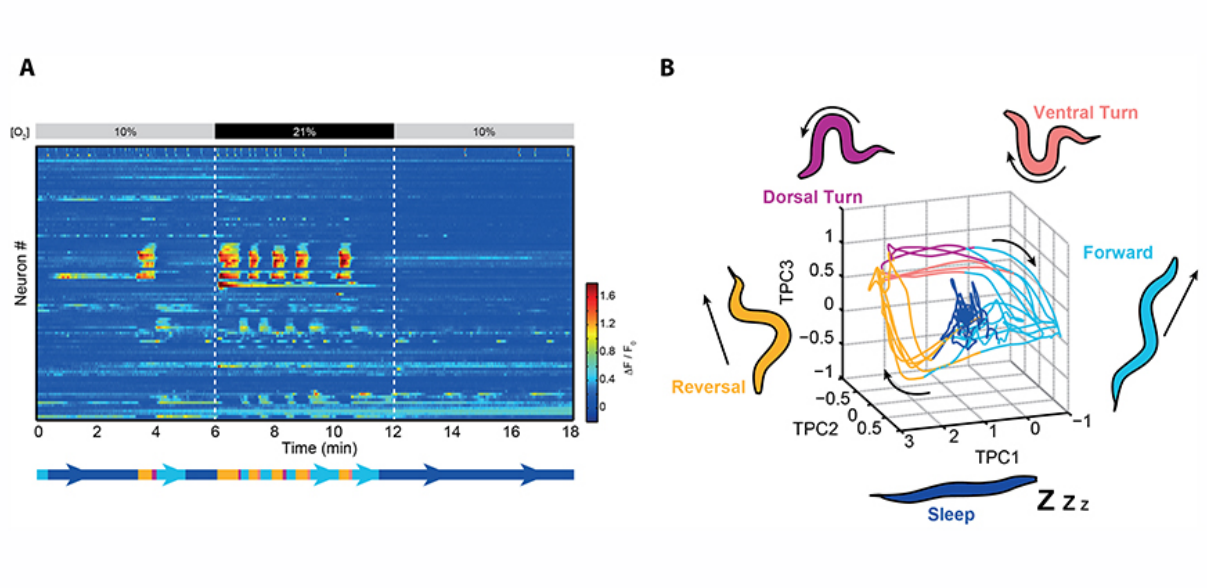


Fig. A: Each row oft he „heatmap" represents the activity of a neuron over time; rows are sorted by correlation. Colors indicate relative change in fluorescence intensities.
Fig. B: Time evolution of the brain state from the above recording represented by a phase plot in principal components space.

Can you predict the worm's behavior based on its neuronal patterns?

Yes, a series of further experiments have helped us to thoroughly characterize these activity patterns. We are actually able to predict what the worm will be doing next, for example in which direction it will crawl. We are now developing our method a step further: this will allow us to go beyond focusing on the brain region of *C. elegans* to encompass the entire neuronal nerve system – for example including what might be called a "mini-brain" located at the tip of the tail. In addition, we want to observe the worm under conditions that are as natural as possible, i.e. not immobilized in a holder but rather in an arena that closely simulates its natural habitat and enables free movement. The main hypothesis that we seek to test is that this animal has evolved to reach independent, curiosity-driven decisions, for example when confronted with stimuli such as food. This involves issues of ethology and quantitative behavior analyses. We apply image-processing methods for such quantifications. This reveals behavior patterns which themselves can again be analyzed statistically and mathematically.

The multidisziplinary research group of Manuel Zimmer, from left to right: Ulrich Rey, Luka Železnik, Richard Latham, Kerem Uzel, Julia Riedl, Harris Kaplan, Niklas Khoss, Oriana Salazar, Annika Nichols, Manuel Zimmer, Lukas Hille, Mara Andrione, Charles Fieseler, Anton Parinov, Daniel Correira

What are your next goals?

My group is a multidisciplinary team that combines advanced microscopy techniques for imaging the entire brain and nervous system with quantitative behavior, molecular genetics and computational neuroscience. Our research objectives continue to focus on brain dynamics, which are crucial for the function of the nervous system. We are striving to understand the information flow from the sensory input to the decision-making process and to the behavioral outputs. We also want to determine why

such a system requires a sleep-wake cycle. Realistically simulating brain dynamics and behavior will yield new results and insights. This holistic approach is currently possible only when applied to such a small animal. Nonetheless, it will enable us to better understand the functional principles behind nervous systems in general – principles which we believe can be translated to larger organisms as well.

Box:

In 1960, the Nobel Laureate Sydney Brenner established the nematode *C. elegans* as a model organism and studied its organ development along with the ontogeny of its nervous system. The simple handling of the animals on agar plates, with bacteria serving as food (*E. coli* strain: OP50 and HB101), and the features of its developmental biology (among others, eutely, simple structure, transparency) favored its success as a laboratory animal. Manuel Zimmer and his team are working on the laboratory strain of *C. elegans.* (Source: Wikipedia)

Manuel Zimmer's website: https://www.imp.ac.at/groups/manuel-zimmer/

Selected publications:

- Nichols, ALA., Eichler, T., Latham, R., Zimmer, M. (2017). A global brain state underlies *C. elegans* sleep behavior. Science. 356(6344)
- Kato, S., Kaplan, HS., Schrödel, T., Skora, S., Lindsay, TH., Yemini, E., Lockery, S., Zimmer, M. (2015). Global Brain Dynamics Embed the Motor Command Sequence of *Caenorhabditis elegans*. Cell. 163(3):656-69