

**Location:** Institut Pierre-Gilles de Gennes (IPGG)/ Institut Curie, 75005, Paris  
**Team/Lab:** Quantitative Developmental Biology & IPGG Technology platform  
**Theme:** Nano and Microfabrication / spinning-disc confocal live-microscopy / Numerical simulations  
**Duration:** 6 months  
**Desired starting date:** no later than January 2023

## Project & Internship proposal

### Context

Temperature impacts all biochemical reactions inside a cell. For developing multicellular organisms, temperature fluctuations pose particular challenges because morphogenetic events depend on both, spatially and temporally coordinated cellular decisions. Despite this, most multicellular systems show a surprising degree of robustness with respect to temperature changes within certain temperature limits. With climate change pushing organisms more and more often towards those limits, it is important to understand how those limits are set, what molecular mechanisms harmonize overall development of an organism and whether there are ways to control these mechanisms.

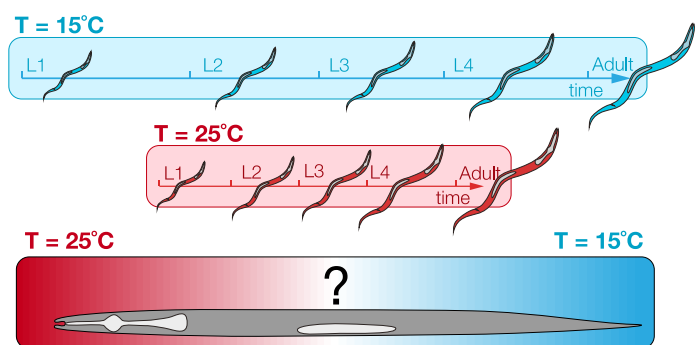
In this project, we address these questions with a very unusual approach: by placing a small developing multicellular organism (nematode worm, called *C. elegans*) in a **linear temperature gradient** (Fig. 1) in a microfluidic system and following its development through live-

microscopy. Using fluorescent reporters of developmental gene expression as well as stem cell markers, we will investigate whether and how compensatory mechanisms help the animal counteract the effects of this extremely unnatural environmental condition. The nematode *C. elegans* larva is only about ~0.5mm long, and we would like a temperature difference of 5-10 degrees between its head and its tail. To obtain this very steep temperature gradient, we overlay a micro-patterned array of transparent resistors (*Selva et al. 2009*) on a microfluidics device that immobilizes the worm along its anteroposterior axis (Fig. 2). This allows us to apply the gradient AND perform high-resolution live imaging at the same time.

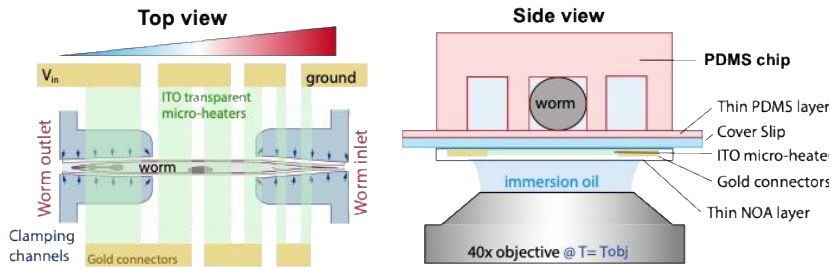
### Goals of the internship:

When the future intern will be joining the team, we will already be able to run experiments with the temperature gradient along the worms. Thanks to the interdisciplinarity of the project, this internship could match with different expectations depending on the student profile:

- Working on COMSOL to simulate the expected temperature profile that depends on many parameters (ITO thickness, resistance, applied voltage, temperature of the objective...etc)
- Optimizing the parameters, the fabrication process and the setup to control as much as possible the temperature gradient that the animals are facing
- Developing a new resistor pattern to obtain a step temperature instead of a linear temperature gradient



**Figure 1. Graphical illustration of the project idea.** The *C. elegans* larva robustly develops through the four larval stages (L1-L4), almost twice as fast at 25°C compared to 15°C. The goal of the project is to uncover compensatory mechanisms that enable this adaptivity of development, by challenging *C. elegans* with a linear temperature gradient and observing gene expression dynamics and cell division timings in the hypodermis before, during, and after perturbation.



**Figure 2. Schematics of the proposed microfluidics device, trapping *C. elegans* larvae inside a channel in a temperature gradient.** (A) Bottom view (B) Side view (not to scale). Stable animal confinement is obtained with a channel whose width can be tuned by pressurizing neighboring “clamping channels”. The gradient of temperature is obtained through a pattern of transparent ITO micro-heaters (light green), etched on the coverslip surface. Width of the microheaters and input voltage determine the shape and

- Developing a new microfluidic device to image *C. elegans* throughout its entire post-embryonic development
- Perform spinning-disc confocal live-microscopy to track developmental gene expressions and skin stem cell divisions with and without the temperature gradient

All experiments will be performed at the platform at Institut Pierre-Gilles de Gennes (IPGG), taking advantage of its well-equipped cleanroom. Working on different parts is obviously feasible and we would be happy to adapt the proposal with your expectations. If

you need additional information, please do not hesitate to contact [eliot.schlang@curie.fr](mailto:eliot.schlang@curie.fr).

### References:

#### *About the linear temperature gradient:*

B. Selva, J. Marchalot, and M. C. Jullien, “An optimized resistor pattern for temperature gradient control in microfluidics,” *J. Micromechanics Microengineering*, vol. 19, no. 6, 2009, doi: 10.1088/0960-1317/19/6/065002.

#### *About the microfluidic chip to immobilize *C. elegans* larvae along its anteroposterior axis:*

S. Berger, S. Spiri, A. deMello, and A. Hajnal, “Microfluidic-based imaging of complete *Caenorhabditis elegans* larval development,” *Dev.*, vol. 148, no. 18, 2021, doi: 10.1242/DEV.199674.

#### *About oscillations of gene expressions and skin stem cell divisions during larval stages:*

G. J. Hendriks, D. Gaidatzis, F. Aeschmann, and H. Großhans, “Extensive Oscillatory Gene Expression during *C. elegans* Larval Development,” *Mol. Cell*, vol. 53, no. 3, pp. 380–392, 2014, doi: 10.1016/j.molcel.2013.12.013.

Gritti, N., Kienle, S., Filina, O. et al., « Long-term time-lapse microscopy of *C. elegans* post-embryonic development” *Nat Commun* 7, 12500, 2016, doi:10.1038/ncomms12500

#### *About the mathematical model:*

B. Novák and J. J. Tyson, “Design principles of biochemical oscillators,” *Nat. Rev. Mol. Cell Biol.*, vol. 9, no. 12, pp. 981–991, 2008, doi: 10.1038/nrm2530.