Internship proposal – 2025

Location: Matière et Systèmes Complexes, CNRS-Université Paris Cité,
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Thesis possibility after internship: YES **Funding**: ANR

Nuclear mechanics and cancer cell invasion

Context

When cancer cells migrate to form metastases, they must deform and go through a confined environment formed by other cells and the extracellular matrix (ECM). The nucleus, the cell's largest and stiffest organelle, may play a critical role in migration through confined environments (1). A soft nucleus should thus be an advantage for a cancer cell to invade the surrounding tissues. The stiffness of the nucleus is determined by both chromatin and lamins, intermediate filament proteins present in the nuclear envelope (2). Inside the nucleus, chromatin adopts specific positions relative to the nuclear periphery, with a dense layer of rigid heterochromatin located beneath the nuclear lamina (3). These heterochromatin lamina-associated domains (LADs) are characterized by low gene density, high content of repetitive DNA sequences and enrichments in repressive histone modifications, trimethylated histone H3 lysine 9 (H3K9me3) established by the lysine methyltransferases (KMTs) SETDB1 and SUV39H. This dense layer of rigid heterochromatin together with lamins strongly influences nuclear stiffness (4). Our collaborators have shown that an interplay between the H3K9 KMTs SETDB1 and SUV39H1 in the A549 lung cancer epithelial cell model modulates H3K9me3 levels specifically at the nuclear periphery (at LADs). Their data suggest that altering the balance between SETDB1 and SUV39H1 leads to modulation of H3K9me3 levels at LADs, and has an impact on nuclear mechanics.

Objectives

The first objective of the project is to measure the mechanical properties of the nucleus following modulation of H3K9me3 levels, using an experimental set-up based on optical tweezers (see Figure) (6). We will measure the nucleus rigidity in the different cell lines developed by our collaborators. We will seek to establish a correlation between the rigidity of the nucleus and the level and spatial organization of H3K9me3 in the different cell lines.

The second objective of the project is to quantitatively characterize the *in vitro* migration capacity of the different cell lines. We will use the lens-free Cytonote device (Iprasense), which enables fast and efficient reconstruction of the phase images of migrating cells over a large field of view (7). We will quantify the motility of cells migrating in different confined environments, including 3D or quasi-2D viscoelastic gels mimicking the extracellular matrix. We will look for a correlation between cell migration capacity and nuclear rigidity.

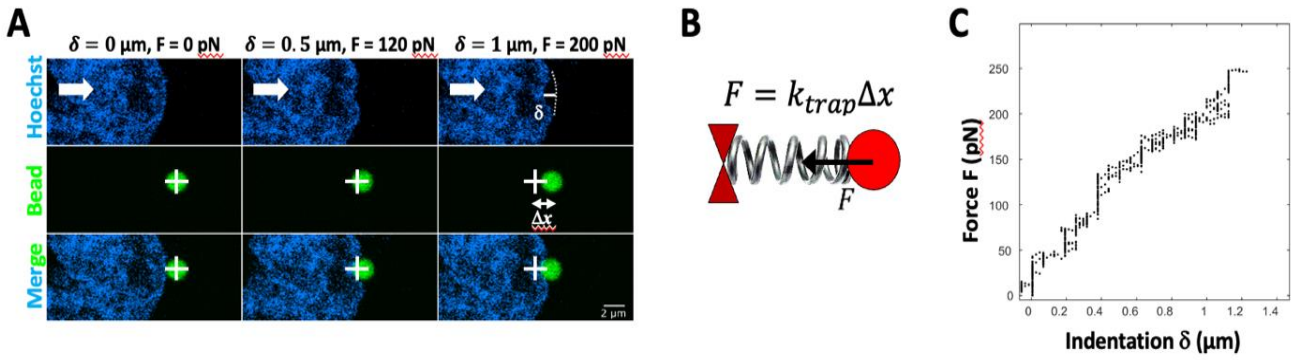

Figure: *Measurements of the nucleus viscoelasticity by indentation in living cells with optical tweezers. (**A**) Images showing a typical nuclear indentation experiment. The white cross represents the centre of the optical tweezers in which the 2 µm-diameter bead is trapped (green). The nucleus (blue, Hoechst) is indented by moving the cell to the right (white arrow) which moves the bead away from the centre of the trap centre by a distance Δx. The indentation δ of the nucleus is measured by image analysis. (**B**) Scheme of the bead in the optical trap. The force F exerted by the optical trap on the bead is similar to the force exerted by a spring of stiffness k_{trap}. (**C**) Force-indentation curve showing force F as a function of indentation δ in the experiment shown in (A).*

References

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