Fiche à nommer selon le format Nom_Prénom_ProjetED2021, à enregistrer en format PDF et à renvoyer à l'adresse : edcdv@sorbonne-universite.fr

Nom et prénom du directeur de thèse (et si besoin du co-directeur) : Marie-Emilie Terret  
Le directeur de thèse et le co-directeur doivent impérativement avoir l'HDR ou équivalent

Coordonnées   Tel : 01 44 27 16 92   e-mail : marie-emilie.terret@college-de-france.fr

Nom et prénom du co-encadrant (non HDR) (s'il y a lieu) : Sandra Touati

Coordonnées   Tel : 0144272575   e-mail : sandra.touati@sorbonne-universite.fr

Y-a-t-il un candidat déjà identifié pour le projet: OUI

Nom et prénom du responsable de l'équipe : Marie-Emilie Terret et Marie-Hélène Verlhac

Intitulé de l'équipe : Oocyte Mechanics and Morphogenesis

Nombre de chercheurs et enseignants-chercheurs statutaires de l’équipe titulaires d’une HDR (ou équivalent) : 2

Nom et prénom du responsable d'UMR ou de département: Marie-Hélène Verlhac

Intitulé et N° d'UMR ou de département : CIRB, UMR 7241, U 1050

Titre du projet de thèse : Cortical tension, a diagnostic tool of oocyte quality

Signature du directeur d'UMR ou de département (vaut avis favorable pour le dépôt du projet) :

(date de l’avis : 08/03/2021.

Spécialité : Biologie cellulaire, Biologie du développement, Biophysique,

Résumé du projet de thèse (1 page maximum, en anglais)
This thesis will be realized in partnership between Sandra Touati from Katja Wassmann’s team (SU) and ours (CDF).

Meiosis produces gametes, essential for sexual reproduction. Two successive meiotic divisions terminate meiosis, allowing the formation of one big haploid oocyte in females. Meiosis in human females is error-prone, producing a high basal rate of poor-quality oocytes that increases with maternal age and as such is the leading cause of miscarriage and congenital syndromes such as trisomies. It is a fundamental public health problem in our societies where women tend to post-pone child bearing reflecting their investment in their career and leading to increased use of Assisted Reproductive Technologies (ART). The quality of the oocyte, key to its developmental potential, determines the chances of producing a healthy embryo after fertilization. Human and mouse oocytes quality is accurately predicted by mechanical properties: if they are too soft or too stiff, they won’t develop after fertilization. Strikingly, aberrant cortical tension is a rather frequent defect in oocytes. Using multidisciplinary approaches, we have discovered how cortical tension is regulated in mouse oocytes and one-cell embryos, and showed that modifying their stiffness alters the geometry of division\textsuperscript{1,2,3}. Our latest work shows that a too low cortex tension causes chromosome segregation errors and aneuploidy in mouse oocytes\textsuperscript{4}. However, the phenotype of stiff oocytes is still unknown. In this project, we want to understand and predict the impact of cortical mechanics on oocyte and one-cell embryo quality, and develop new technologies to sort them according to their cortical tension for personalized reproductive medicine. In the past years, our lab developed innovative tools, in particular state-of-the-art imaging, biophysical approaches, biophysical modeling, combined to mouse genetics. Using these tools, in collaboration with Clément Campillo (Evy University), Sandra Touati (Sorbonne University) and Elsa Labrune (Hospices civils de Lyon, specialized in ART), we now propose to:

**Aim 1: Analyze how oocyte quality is impacted by a too high cortical tension**

If extra-soft oocytes represent the most common defect among human oocytes (36%), stiff oocytes are also frequent (19%). We will characterize oocyte defects induced by a too high cortical tension by engineering stiff oocytes, using chemical inhibitors and manipulation of F-actin cortical network. We will analyze cortical tension with micropipette aspiration\textsuperscript{1,2,3,4} (Campillo lab), shape, spindle morphogenesis and positioning, actin organization, chromosome segregation and fertilization in live cells. This aim will be performed in collaboration with Sandra Touati from Katja Wassmann’s lab (Sorbonne University), which studies in depth the control of cell cycle and chromosome segregation at meiosis exit in mouse oocytes (Gryaznova EMBO J 2021, Karasu JCB 2019, Vallot Curr Biol 2018, El Yakoubi Nat Commun 2017, Touati Nat Commun 2015). Indeed, a previous study suggests that stiff oocytes could have defects in meiosis exit.

**Aim 2: Characterize oocytes coming from natural populations of murine and human oocytes**

The goal here is to generalize our findings observed on artificially induced extra-soft and stiff oocytes to naturally soft and stiff ones by measuring systematically in unmanipulated cells all the parameters that we found\textsuperscript{1,2,3,4} or will find (Aim 1) as being altered. For that, we will use Atomic Force Microscopy (AFM) coupled to live fluorescent imaging (Campillo lab), as AFM offers a high throughput and allows screening oocyte populations compared to micropipette aspiration. We will evaluate cortical tension variability between oocytes coming from the same animal, but also between animals, and test the effect of maternal age on cortical tension (comparing young and old mice), age being an unfavorable criterion for oocyte quality and women fertility. We will then apply this technique to human oocytes (Labrune lab) to test if some unfavorable fertility features (such as age, some pathologies...) correlate with aberrant oocyte cortical tension in human.

**Aim 3: Develop a microfluidic device to sort oocytes and one-cell embryos according to their cortical tension**

This aim will provide a tool that we plan to develop for future medical application. We fabricated a user-friendly minimally invasive microfluidic device to sort oocytes/one-cell embryos based on their cortical tension and thus their developmental potential. We believe it will have a significant social impact, since few fertility centers have the expertise to properly measure cortical tension with micropipette aspiration or AFM. We are setting up this device in the mouse and we will adapt it to human oocytes and one-cell embryos (Labrune lab), to predict their developmental potential for clinical use.

In the end, our comprehensive project will bring new fundamental advances on why oocyte formation is error-prone but also exploit this knowledge to deliver a potential device for oocyte and one-cell embryo quality diagnostic. Our microfluidic technique could have a strong societal impact with a clear medical relevance by changing the daily practices in fertility centers to improve ART rate of success.


In bold previous PhD students of the PhD supervisor. Note that Agathe Chaîgne obtained prestigious awards, among them the prize for young researchers Bettencourt-Schuller 2015, the prize Le Monde de la Recherche Universitaire 2015 and the 2015 Lindau Nobel Laureate Meeting. Isma Bennabi was also recipient of the prize for young researchers Bettencourt-Schuller in 2019.
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**Faisabilité du projet de thèse (1/2 page maximum, en anglais)**

Our project is ambitious and innovative, requiring a unique interdisciplinary set of skills at the interface between cell biology labs (Terret and Wassmann labs), a biophysics lab (Campillo lab) and a medical lab (Labrune lab). We believe that we are the best consortium to successfully conduct it:

- First, we have shown our ability to direct projects (management of PhDs and post docs, securing funding, publishing results in high impact journals, invitations at international conferences).

- Second, we have all the skills to pursue it since it relies on tools, expertise and collaborations that were successfully established (see references 1 to 4 from previous section). It should be noted that Marie-Emilie Terret and Katja Wassmann collaborated successfully in the past (Terret, Wassmann et al Curr Biol 2003, Med Sci 2008), and that Marie-Emilie Terret was part of Sandra Touati’s PhD jury in 2014 when she was in Katja Wassmann’s lab (she was recruited by the CNRS in Katja Wassmann’s lab in 2019 after a post doc in the UK in Franck Uhlmann’s lab).

- Third, all the equipment necessary for the success of the project is already in place and functioning. The Terret and Wassmann labs have all the equipment to study mouse oocytes (microinjection apparatus, spinning disk confocal microscope…). The Terret group is located in the Centre for Interdisciplinary Research in Biology, CIRB, in the Collège de France, which has several state-of-the-art facilities (animal, imaging, including a cutting-edge bio-AFM). A contract has been signed between the CIRB and the Institut Pierre-Gilles de Gennes (IPGG) to use their facilities for microfluidic systems fabrication.

Our three aims require expertise in mouse oocytes (Terret and Wassmann), state-of-the art imaging (Terret and Wassmann), biophysics (Campillo), microfluidics (Campillo) and human oocytes (Labrune). These aims do not depend on each other for their feasibility, as they are independent but interrelated. We plan to develop our project in 36 months. We recruited in 2021 two research engineers, Lucie Barbier and Rose Bulteau, for 1 and 2 years respectively, thanks to two grants obtained by the consortium in 2020. They will train the PhD student in Atomic Force Microscopy (AFM), micropipette aspiration and microfluidics.

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**Thèses actuellement en cours dans l’équipe**

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<td>CROZET Flora</td>
<td>Marie-Emilie TERRET</td>
<td>2017</td>
<td>515</td>
<td>Bourse MENESR de 3 ans financée par le Fonds France-Canada pour la Recherche (FFCR), 4ème année FRM.</td>
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**Trois publications récentes du directeur de thèse** (du co-directeur ou du co-encadrant s’il y a lieu). Mettre en gras le nom du directeur de thèse.

**Marie-Emilie TERRET** :


**Sandra TOUATI** :

1. Cdc14 and PP2A Phosphatases Cooperate to Shape Phosphoproteome Dynamics during Mitotic Exit. 

2. Phosphoproteome dynamics during mitotic exit in budding yeast. 

3. Mouse oocytes depend on BubR1 for proper chromosome segregation but not for prophase I arrest. 
**Ecole Doctorale COMPLEXITE DU VIVANT – Fiche Projet CONCOURS**

**Docteurs encadrés par le directeur de thèse** ayant soutenu entre la date de dépôt de ce dossier et il y a 5 ans et publications relatives à leur sujet de thèse. Mettre en gras le nom du directeur de thèse et celui du docteur.

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<td><strong>Flora Crozet</strong></td>
<td>10 septembre 2021</td>
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**Publications :**

1. Communication by physical contact between the oocyte and the surrounding somatic cells shapes the oocyte quality. 

2. Myosin-X is dispensable for spindle morphogenesis and positioning in the mouse oocyte. 
   **Crozet F**, Da Silva C, Verlhac MH*, **Terret ME**. Development In Press.

3. Artificially decreasing cortical tension generates aneuploidy in mouse oocytes. 

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**Publications :**

1. [Cortical tension of the oocyte and euploidy: the right balance].

2. Artificially decreasing cortical tension generates aneuploidy in mouse oocytes. 

3. A computational model of the early stages of acentriolar meiotic spindle assembly. 

4. Shifting meiotic to mitotic spindle assembly in oocytes disrupts chromosome alignment. 

5. Meiotic spindle assembly and chromosome segregation in oocytes. 

**Isma Bennabi** was recipient of the prize for young researchers Bettencourt-Schueller in 2019.