Development of a pancreas-intestine crosstalk organ on chip to mimic and test the influence of specific diets on mice

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Context and justification of the project

During the first meeting of Sorbonne Université's new interdisciplinary Initiative on Alimentation on February 14th 2025 (where we also presented the ASU-GHI Institute to the community), several national and international experts on nutrition, alimentation and health have clearly demonstrated that metabolic syndrome and obesity are clearly impacted by the patients diets. Prof. Eric Ravussin's and Prof. Karine Clément's exceptional seminars demonstrated this impact with the help of very large scale clinical trials, but it was also clear that there is not a single recipe for success in using special diets, including the use of so-called benefic microbiota to help reduce the negative effects on health of metabolic syndrome. The very broad patient-to-patient response heterogeneity was clearly highlighted and calling for a currently non-existing solution to elucidate the underlying mechanisms behind the positive influence of some changes in alimentation behavior (such as ketogenic diets, intermittent fasting, high/low fat diets, etc.). What is currently clear, however, is that when one of these solutions does work and reduces or even abolishes the deleterious effects of obesity and metabolic disease for a patient (and it was proven in patient-imitating animal tests), it is because it has both deeply impacted the insulin secretion behavior of their pancreatic islets and modified the intestinal immunity and microbiota (which, in turn is also known to influence pancreatic islets positively).

On the other hand, Dr. Marcia Hiriart from UNAM, our strategic partner, has presented their latest results using animal models (mice and rats) and showing very interesting results on the rapid effects of specific high-fat or high-sucrose diets on them. Interested in our current human-based *in vitro* models, she also highlighted the need for an improved in vitro model using mice and rats cells, to elucidate the mechanisms at play in such drastic results and hence better understand the impact of sweet and fat on the animal systemic health.

To understand the mechanisms of what is happening *in vivo*, clinicians and biologists concur that it is thus **imperative to develop new *in vitro* models capable (a) of imitating the dynamic and pancreas-intestine communication modulated by alimentation and (b) of enabling the easy access to the exchange molecules between the two organs while placed in different diets conditions** (using specific associated molecules and cells form different animal models fed with the different diets).

An excellent option to develop such models is the **current development of organ-on-chip (OoC) systems**, offering the possibility to culture primary cells for long periods of times inside microfluidic chips where all the necessary biological assays typically addressed inside conventiona l*in vitro* models can be performed on multicellular tissues that recapitulate the organs with the greatest fidelity. In such systems, fresh primary cells are even used today to imitate a specific patient's or animal condition's biology *in vitro*, for personalised drug efficacy and safety tests, as well as mechanistic assays that elucidate possible cellular and molecular mechanisms. Recently, an international effort is being made to **interconnect OoCs for the precise restoration and monitoring of organ-to-organ crosstalk at the tissular, cellular and molecular levels**. For this to work properly, several groups demonstrated that the interconnection needs to be via true blood vessels, as the role of endothelial cells is crucial for the efficient homeostasis and organ-to-organ communication. Since 2024, our 2 groups have elaborated an ambitious project proposal of a vascularized multiorgan obesity model, responding to the 2025 MedOoC PEPR call; **this PhD project focuses on the key intestine-pancreas axis**.

Objectives

The objective of this project is thus to develop the first vascularised multi-OoC system of intestine and pancreas, using perfusable blood vessels as interconnection. The PhD student will first learn how to set up and then optimize individually two existing OoC *in vitro* platforms (developed in collaboration between our two groups since 2022). In parallel, a perfusable blood capillary vessel model currently developed by Team 1 will be modified to vascularize each model independently, and then in series, using recently reported anastomosis approaches currently tested by Team 1 with promising results. This model will finally be optimised for the use of freshly obtained animal cells, obtained routinely by Team2, to offer the **ideal technological tool** of dynamic interorgan communication.

Summarized methodology, preliminary results and expected results

Development methodology: To develop the first iteration of such a technological model, two OoC *in vitro* platforms currently developed between our teams will be used and optimised: (1) a microfluidic chip with stiffness and flow control designed for long-term (2 weeks) culture of fresh, primary pancreatic islets and (2) a multicellular intestine model made of mouse cells. The first OoC platform has demonstrated excellent results using fresh mice cells and is in the process of functional insulin secretion validation using perifusion assays. The second prototype, currently developed by Team 2, will be modified to culture animal cells dynamically under flow and on the correct physiological stiffness using an on-demand chip, based on an existing 2-channel design already developed and used by Team 1 and perfectly adapted to this aim. Finally, (3) the current perfusable capillary-on-chip device using primary endothelial cells will be used to vascularise each chip: individually first, and then in the form of a new integral setup that will be engineered to interconnect the two organs together in a sequential or recirculation mode. First, easily available healthy mouse cells will be used for optimisation, before cells from diet-specific animals will be employed (all cells are routinely obtained by Team 2, with all the permits).
Validation assays: routine tests of cell survival, preservation and functionality will first validate the longevity in all 3 models under flow, independently and after interconnections; then perfused vascularisation will be validated using our current imaging assays; finally gene expression and transcriptome assays will be performed. To prove the efficacy of the model, the effect on several molecules recapitulating specific diet-associated metabolites will be tested in the final model and compared with existing results from Team 2 and UNAM's team.

International partnership: a complementary and interdisciplinary collaboration

The 2 SU co-directors of this project have been collaborators for the past 3 years, with promising preliminary results. Dr. Chloé Amouyal is a clinical endocrinologist, expert in metabolic disease in the context of obesity and in charge of a research sub-group at UMRS1269 developing *in vitro* intestine and pancreas models with animal cells, and animal testing. Prof. M. Hautefeuille is an expert in on-demand OoC development and currently focuses on vascularisation. Since the PhD project is aimed at OoC development, he will be the leading co-supervisor. Since 2024, we have also started a solid collaboration with Dr. Marcia Hiriart from UNAM, who is a world expert in metabolic disease and works with animal models and is interested in testing the models for specific high-sugar and high-fat diets. As no co-direction is currently available between our entities, she will be co-supervisor and will host the PhD student and visit our laboratory.

All technical and scientific expertise from both teams will support PhD student progression, benefiting from our existing collaboration. Funding for the project will be provided from our team's existing projects; we expect substantial funding increase in 2025 (PEPR project proposal, ANR proposals accepted in 2nd round and ranked last year; SU Emergence proposal).