The pancreatic endocrine β-cells are the only cells in the body that can produce insulin, a hormone responsible for controlling glucose metabolism, and loss or dysfunction of β-cells leads to diabetes, named by WHO a global epidemic. Over 400 million people worldwide are affected by diabetes and over 1.5 million people die yearly due to diabetes-induced complications. Currently available therapies require external insulin delivery or glucose-lowering drugs together with lifelong glucose monitoring. Therefore, huge efforts are put into the creation of a robust cell source for β-cell replacement therapy where β-cells made in the laboratory from human pluripotent stem cells (hPSCs) could be transplanted into patients. Although tremendous progress has been made recently, hPSCs cannot yet be reliably coaxed into functional β-cells in sufficient purity, numbers, and costs. Ideally, what we need is an expandable population of human differentiated β-cells that maintain their functional capacity over the long-term.

This project aims to discover new signals and mechanisms regulating β-cell proliferation and development. We will focus on early fetal-like β-cells. Early β-cells differ from adult β-cells. Early β-cells show a greater proliferative capacity than adult β-cells but do not yet have a well-developed ability to secrete insulin in response to glucose stimulation. The β-cells developed in the process of directed differentiation from hPSCs are more proliferatively and physiologically similar to early fetal-like β-cells than to adult ones. Some studies suggest that the increase in human β-cell proliferation is linked to their diminished function, colloquially speaking, proliferate or secrete. These observations raise the important question as to whether proliferation and functionality are mutually exclusive states in β-cells.

The differences in proliferative capacity and functionality between early and adult β-cells may be explained at least in part by signals from the surrounding microenvironment, the extracellular matrix (ECM). The ECM is more than just a passive support system and reservoir of macromolecules such as growth factors or cytokines. The ECM represents a dynamic microenvironment as its organization varies significantly between developing and mature tissues. Based on literature and our preliminary data, we hypothesize that ECM components play a pivotal role in β-cell proliferation, differentiation, and function and the ECM composition is crucial for the balance between functional maturity and proliferative capacity of β-cells.

Here, we focus on the ECM component - SPOCK2 found to be expressed in the newborn β-cells and their progenitors. Based on our preliminary data we hypothesize that SPOCK2 may engage in different processes in pancreatic β-cell, 1) SPOCK2 can regulate proliferation and function of human early fetal-like pancreatic β-cells and 2) SPOCK2 can play a role in β-cell formation *in vitro*.

To address these issues, we will:

1) determine the SPOCK2 role in human early pancreatic β-cell proliferation and function; as preliminary data we identified SPOCK2, as a putative inhibitor of human β-cell proliferation, here we will study to an extent the SPOCK2 effect on β-cell proliferation and physiology.

2) determine how does SPOCK2 controls the expansion of human early pancreatic β-cells; we will investigate the molecular mechanism associated with the β-cell proliferation orchestrated by SPOCK2 inhibition.

3) determine the SPOCK2 role during subsequent stages of human β-cell development *in vitro*; we will gain insight into functional changes of SPOCK2-deficient β-cells and SPOCK2 significance for human pancreatic development.

Using advanced microscopy techniques, single cell RNA sequencing and mouse models, we will characterize newly generated human β-cells by comparing them to adult β-cells and elucidate the key role of SPOCK2 in the β-cell formation and proliferation. Our research will contribute to the improvement of procedures for generating human β-cells, which may be an attractive platform for basic research, for diabetes modelling, as well as for high-throughput drug testing or, finally, may be used in regenerative medicine in cell therapies for patients with diabetes.