Charna Dibner

Scientific activities

Basic research project: Circadian regulation of pancreas gene expression

Due to the emerging importance of the pancreas circadian clock on islet function and type 2 diabetes development in rodents, we aimed to examine circadian gene expression in human islets. The oscillator properties were assessed in intact islets as well as in beta cells. To this end, we established a system for long term bioluminescence recording in cultured human islets, employing lentivector gene delivery of the core clock gene *Bmal1-lucicferase* reporter. Beta cells were stably labelled using a rat insulin2 promoter (RIP) fluorescent construct. Single islet/cell oscillation profiles were measured by combined bioluminescence-fluorescence time-lapse microscopy. Human islets synchronised *in vitro* exhibited self-sustained circadian oscillations of *Bmal1-luciferase* expression at both the population and single islet levels, with period length of 23.6 and 23.9 hours respectively. Endogenous *BMAL1* and *CRY1* transcript expression was circadian in synchronised islets over 48 hours, and antiphasic to *REV-ERB, PER1, PER2, PER3* and *DBP* transcript circadian profiles. *HNF1A* and *PDX1* exhibited weak circadian oscillations, in phase with *REV-ERB* transcript. Dispersed islet cells were strongly oscillating as well, at population and single cell levels. Importantly, beta and non-beta cells revealed oscillatory profiles well synchronised with each other. We provide for the first time compelling evidence for high-amplitude cell autonomous circadian oscillators displayed in human pancreatic islet, and in dispersed human islet cells. Moreover, these clocks are synchronised between beta and non-beta cells in primary human islet cell cultures (Pulimeno et al., Diabetologia, March 2013, in press).

During 2012 first paper on this project was published. In addition, the project got extended to alpha cell studies in mouse and human model (financed by EFSD July 2012), and on the studies of the circadian oscillator in primary human myotube culture from healthy and T2D subjects (financing applications in progress to FNRS and to Arteres Foundations). We will apply the same methodology we have developed for mouse and human islet oscillator studies to the studies of impact of human skeletal muscle clocks on the muscle gene expression and function in health and in type 2 diabetes.