This read-me document is a guide to build an FPGA-board based ECG-gating system for multi-photon microscopy of the in-vivo mouse brain, as described in Streich et. al, Nat. Meth. 2021.

The material contains two parts: 1. General considerations; 2. A demonstration which was used in our microscopy system. For users who have experience with FPGA programming and PCB design, we provide all original codes and designs for their reference. Modification and setting changes are available depending on their applications and hardware. For users with less or none experience in these two techniques, we provide a demonstration which can be duplicated in their labs.

General considerations

An FPGA board can provide real-time peak detection of cardiac waves through digital processing. However, most commercially available FPGA boards only accept input analog signals within a specific range, while different physiological monitoring systems provide different output signal ranges. Therefore, a signal conditioning module must be introduced between these two instruments. Usually, a signal conditioning module can be integrated into a single printed circuit board which contains steps of signal decoupling, bandpass filtering, rescaling and bias. The output of the conditioning board should fit the voltage requirement of the A/D converter, for example 0-1.0V, and be robust to the noise.

The A/D conversion should have sufficient resolution (>=12 bit is recommended). The algorithm used for peak detection must automatically adapt to drifting analog signal level. Note: for real imaging applications, accuracy rate is not required to 100%. Errors in peak detection either reduce the duty cycle or introduce motion artifacts in the corresponding image areas. Therefore, a compromise between accuracy rate and algorithm complexity might be considered.

See figure in the “Schematic of FPGA-board based ECG-gating system.pdf”

Demonstration

This demonstration is being used in our adaptive optics three-photon microscope as described in Streich et. al, Nat. Meth. 2021. Note that with different hardware and/or software our ECG-gating might not work reliably.

Hardware

- Physiological monitoring system: ST2 75-1500, Harvard Apparatus
- FPGA board: ZYBO Z7 ZYNQ-7010
- External power supply to the conditioning board: +/- 12V
- Conditioning PCB board design: bandpass filter(1Hz – 3 kHz); potential meter for re-scaling (0-20kΩ); potential meter for bias (0-1kΩ); inverting stage (optional) is only useful when the input cardiac wave is reversed.

Software

- Copy the BOOT.bin file into a micro-SD card and insert into the slot of the FPGA board. This file was compiled from the VIVADO software.
- Run the Matlab (we also provide a GUI) scripts to set parameters of FPGA codes. Manually test and find proper parameters by changing the values in the scripts.

Wire connection

- Connect a mini-USB cable between the FPGA board and host computer (set parameters).
- Connect external power supply (+/- 12V) to J1 (pin 1, 3) on the conditioning board.
- Connect J_poti on the conditioning board to bias potential meter.
- Connect J5 on the conditioning board to re-scaling potential meter.
- Connect ECG signals from ST2 75-1500 to J_IN on the conditioning board.
- Connect J_OUT on the conditioning board to the XADC Pmod port (+, pin1; GND, pin 7).
- Connect Trigger out to the High-speed Pmod port JD port (+, pin3; GND, pin 5).

NOTE: These pin settings on the FPGA are for the provided BOOT.bin file. For users who can programming FPGA using VIVADO, the pin settings can be modified.