Engineering control solutions for microscopy

EIROforum IWG Workshop on Systems Engineering

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EMBL (Heidelberg)

06.02.2025

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EMBL overview

EMBL-EBI

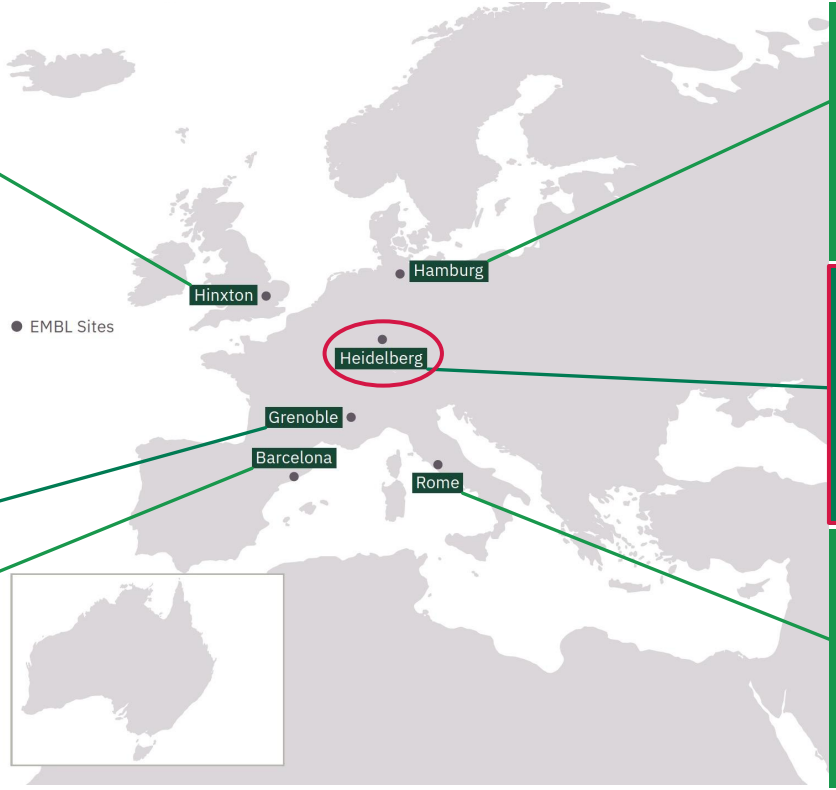
Bioinformatics

Grenoble

Structural biology

Barcelona

Tissue biology and disease modelling

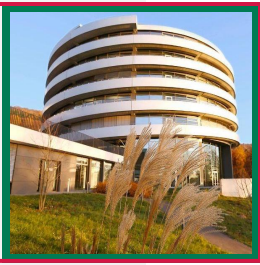


Hamburg

Structural biology

Heidelberg

Life sciences



Rome

Epigenetics and neurobiology

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Prevedel lab: Technologies for imaging life in context

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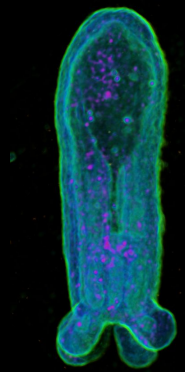
High-speed 3D imaging

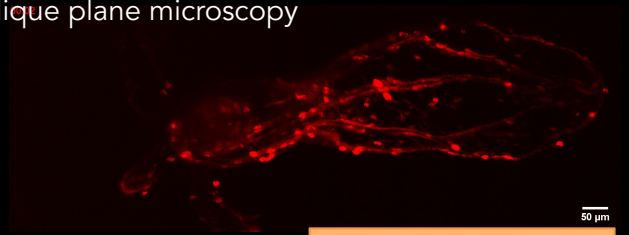
Optical Coherence Microscopy



- Organismal-scale (label-free)

Stokkermans et al., Curr. Bio. 2022
Ruperti et al., Curr. Bio. 2024

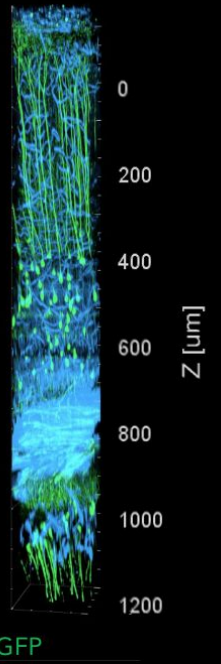
Oblique plane microscopy



- Organismal-scale (fluorescence)

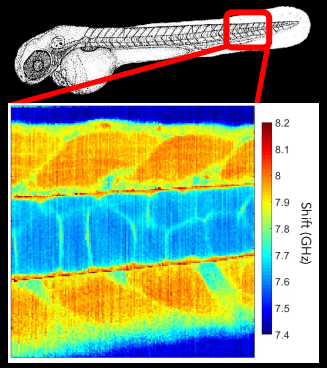
Singh et al., Opt. Exp. 2023
Davis et al., BOE 2024
Paix et al. PNAS., 2022

Deep tissue imaging



Schubert et al., 2024, Nat. Comm.
Streich et al., 2021, Nat.Methods
Morelli et al., 2021, Cell Reports

Biophysical imaging



- Visco-elasticity in 3D

Prevedel et al., Nat. Meth. 2019
Bevilacqua et al., Bio. Opt. Exp. 2019
Chan et al., Comms Bio2021
Bevilacqua et al., Nat. Meth. 2023
Yang et al., Nat. Meth. 2023
Coraggio et al., Nat. Comm. 2024

Controlling a microscope

Microscopy & Biology

Confocal image of a young sea anemone (Nematostella vectensis) and embryos from a fly (Drosophila melanogaster) fluorescently labeled to show mRNA from four genes.

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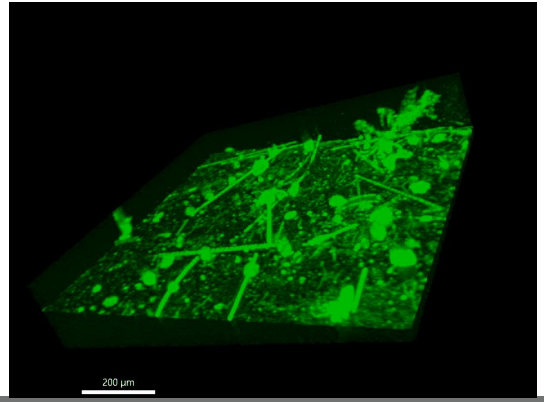
Why do we need to automate a microscope ?



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Tools and Resources

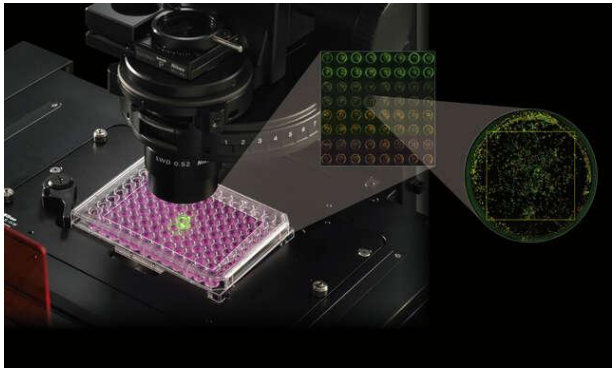
Neuroscience

Rapid whole brain imaging of neural activity in freely behaving larval zebrafish (*Danio rerio*)

Lin Cong, Zeguan Wang, Yuming Chai, Wei Hang, Chunfeng Shang, Wenbin Yang, Lu Bai, Jiulin Du, Kai Wang, Quan Wen

Shanghai Institutes for Biological Sciences, Chinese Academy of Sciences, China; University of Science and Technology of China, China; University of Chinese Academy of Sciences, China

Sep 20, 2017 • https://doi.org/10.7554/eLife.28158



Volumetric (3D) imaging

Tracking dynamic systems

e.g. living organism or single molecules

https://www.microscope.healthcare.nikon.com/applications/life-sciences/high-throughput-imaging

High-throughput system

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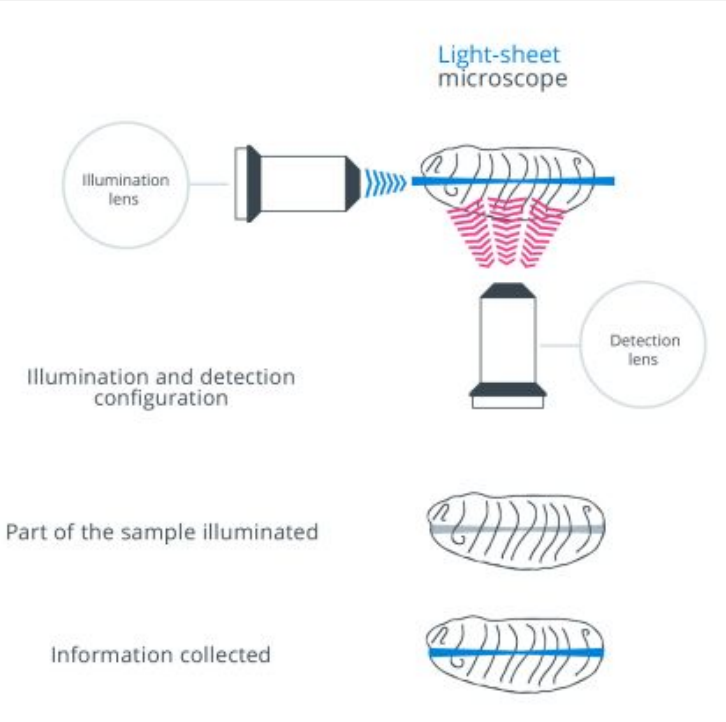
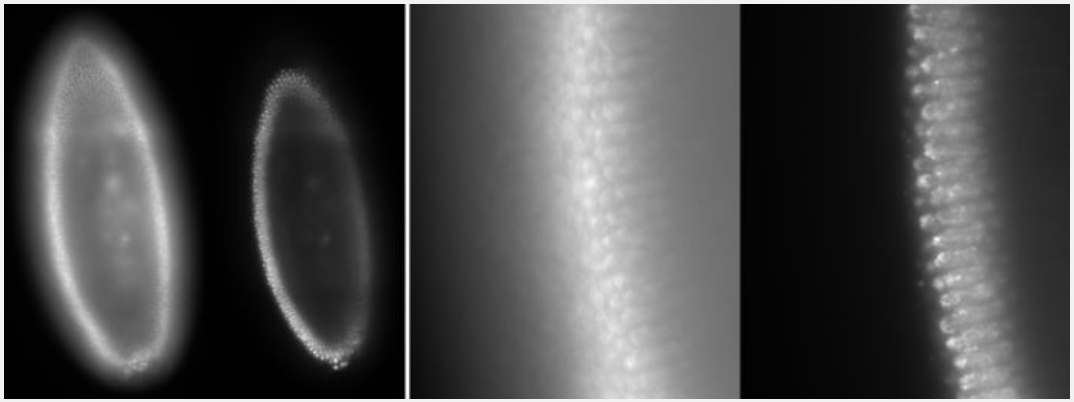
Example: light-sheet microscope



From Bruker "What is Light-Sheet Microscopy"



Fixed Drosophila embryo with Sytox Green nuclear marker, mounted in 1.5% agarose and imaged with epi-fluorescence and light sheet illumination. Epi-fluorescence illumination: 470 nm LED excitation, 488 nm LP dichroic mirror. Light sheet illumination: 491 nm excitation. BP525/50 emission filter, 50 ms exposure time. Z-stacks w/ 1 µm z-spacing. Sample by Jeehae Park, Harvard Medical School. Scale bar = 50 µm.
https://huiskenlab.com/principles/

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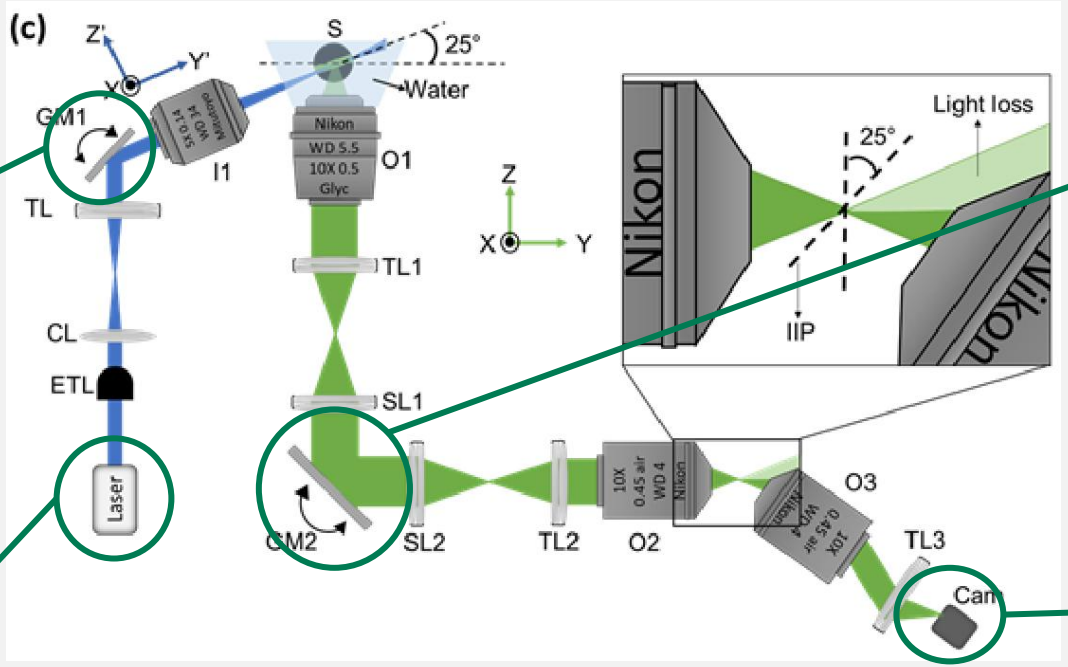
Example: light-sheet microscope

Analog galvo mirror: -5V to 5V

Intensity control: 0-5V, slow
Shutter, on/off: digital, quick



Digital galvo mirror: custom protocol

Camera trigger: TTL

Rajwinder Singh, Kaushikaram Subramanian, Rory M. Power, Alexandre Paix, Alejandro Gil, Aissam Ikmi, and Robert Prevedel, "**Oblique plane microscope for mesoscopic imaging of freely moving organisms with cellular resolution,**" Opt. Express **31**, 2292-2301 (2023)

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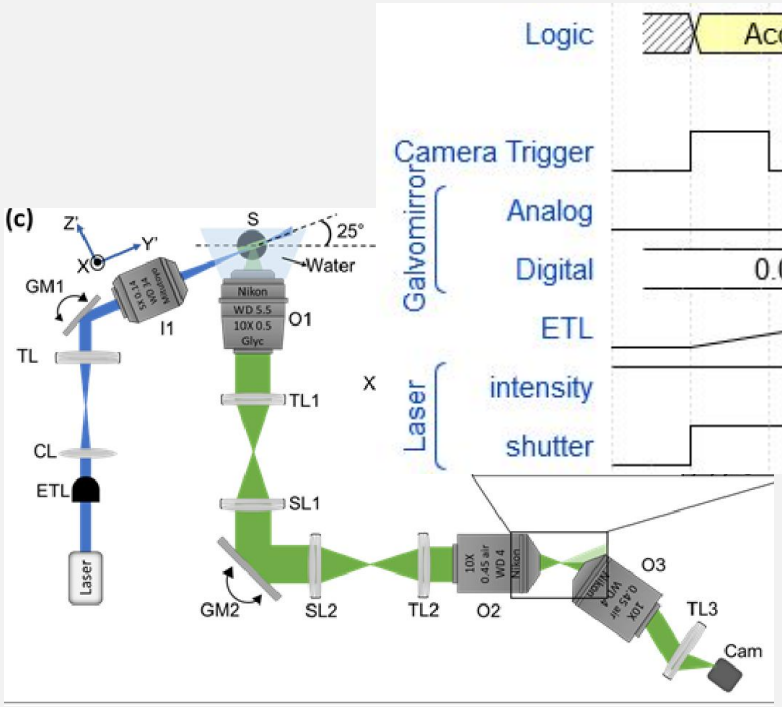
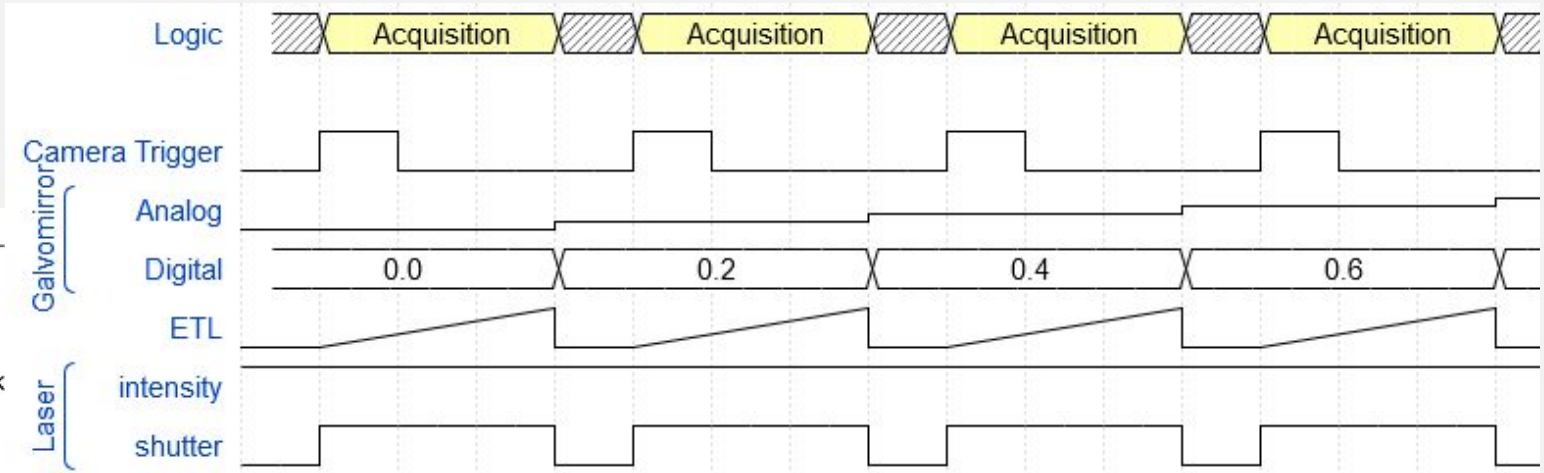
Example: light-sheet microscope





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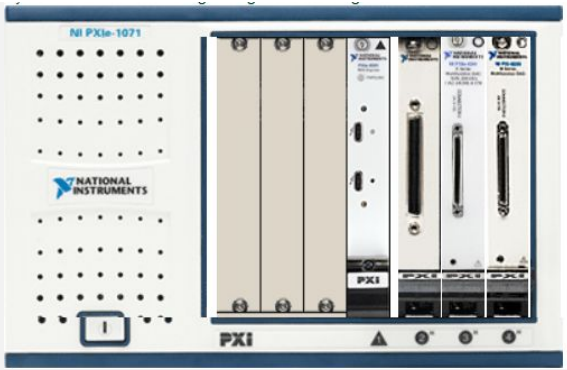
The current usual solution



Screenshot from NI website

Usual commercial solution:
National instrument and their PXI system

- Expensive
- Labview
- hard to customize
- no FPGA-level control

Companies
Can build in-house custom solution (but expensive, time-consuming and need special skills)

Research group
No real alternatives
+ scientists usually have optics or biology background

Our microscope hardware controller

Open-source, multi-modality and community driven

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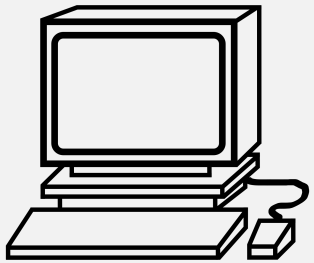
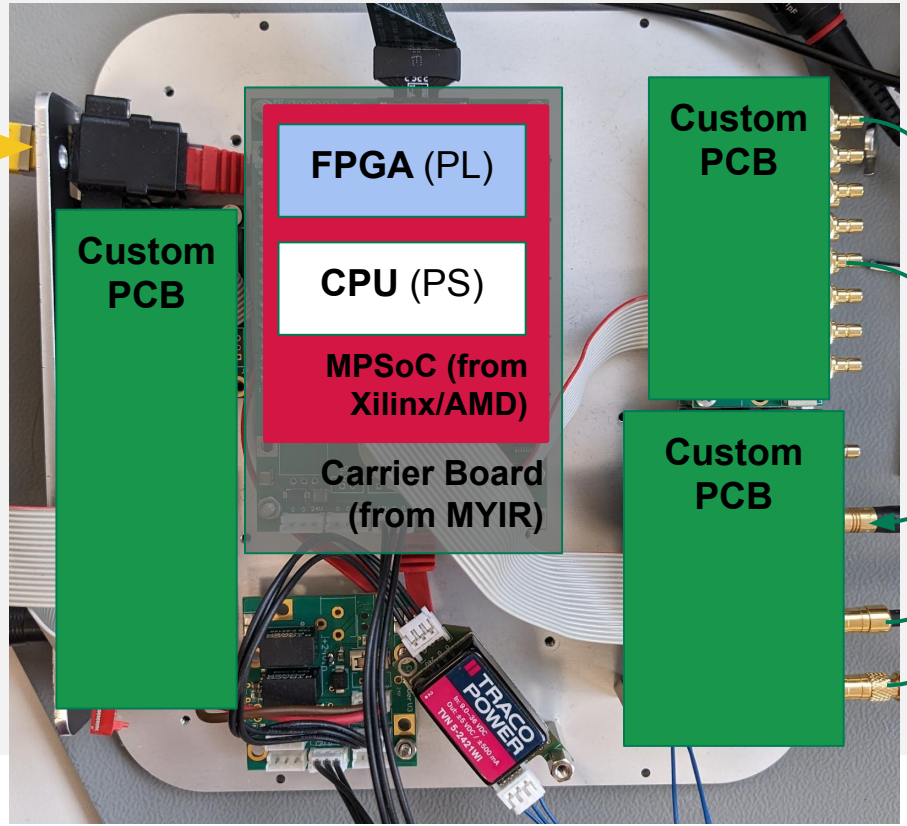
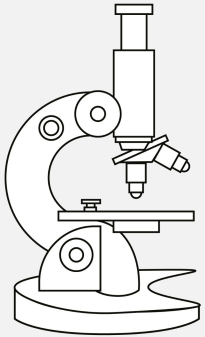
The Galaxy Cube



Workstation





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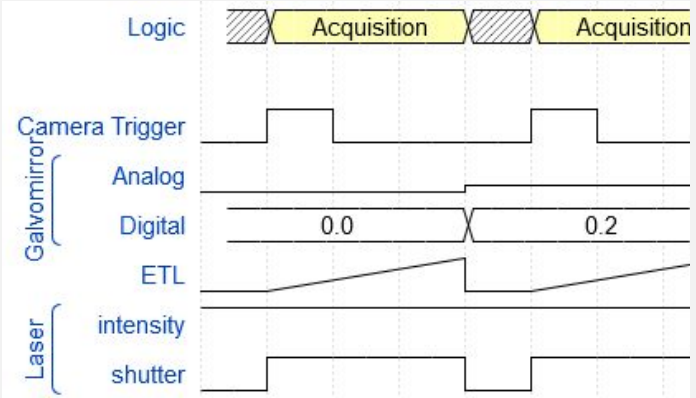
Input/output specifications

- Analog outputs
  - 4 channels: -5V to +5V, ~1 MHz
  - 8 channels: 0V to +5V, ~100kHz
- Analog inputs
  - 4 channels: -11V to +11V, 4MPS
- Digital outputs
  - 8 + 4 channels: TTL - *compatible with 3.3V and 5V logic*
- Digital inputs
  - 4 channels: TTL - *compatible with 3.3V and 5V logic*
- Serial communication
  - RS485
- Standard Ethernet port



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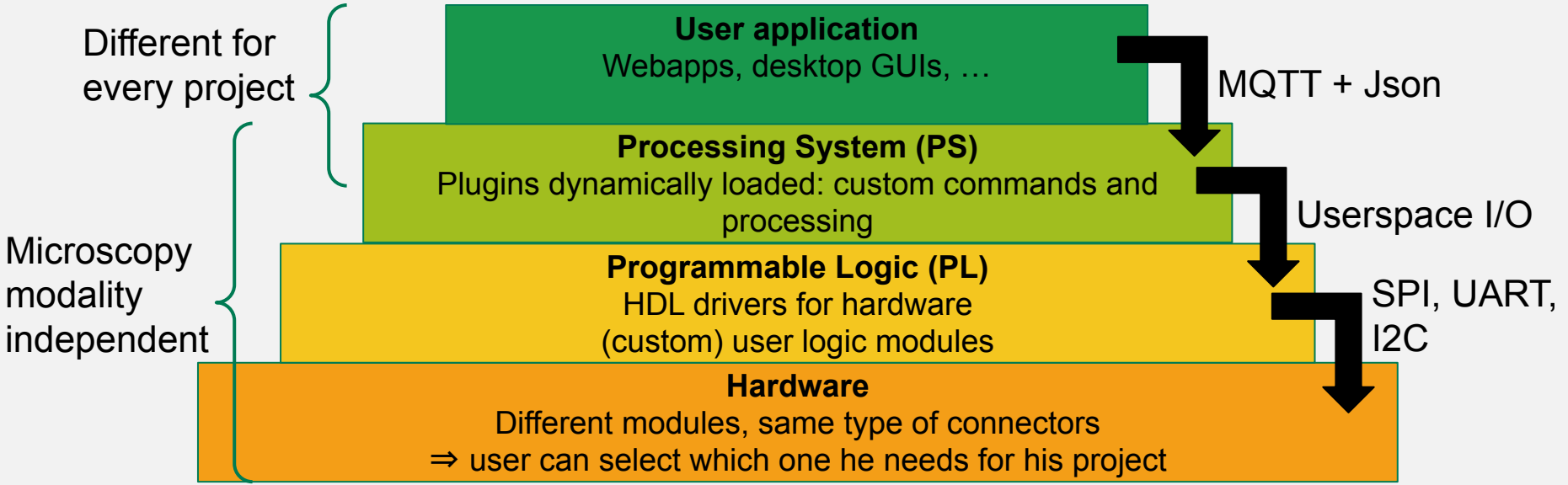
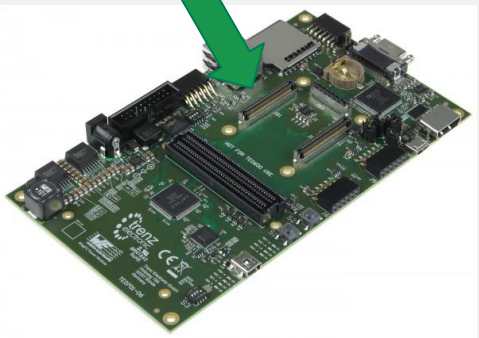
Project re-usability



Tools and Technologies

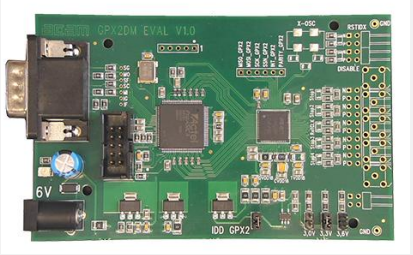
Hardware - commercial over custom built

FPGA modules (size adaptable to specific use)



Trenz Electronic carrier board (TE0701)

EVAL-ADAQ4380-4
Evaluating the ADAQ4380-4 Quad, 16-Bit, 4 MSPS, Simultaneous Sampling, µModule Data Acquisition Solution



TDC-GPX2 Evaluation Kit
Time-to-Digital Converter,
best single-shot accuracy of **10ps**
lowest pulse-to-pulse spacing 5ns

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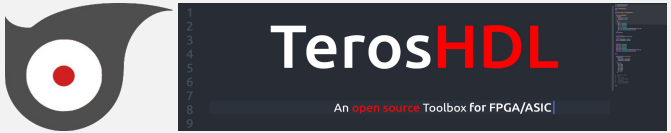
FPGA - Programming logic

Visual Studio Code

Open source toolbox for HDL developers (as a VScode plugin)



https://github.com/TerosTechnology/vscode-terosHDL

Cheby: file format to describe the HW/SW interface + tools to generate HDL, drivers, documentation...
https://gitlab.cern.ch/be-cem-edl/common/cheby

FPGA - Programming logic (Honorable mentions)

Hog: HDL on git
https://gitlab.com/hog-cern/Hog

Machine learning inference in FPGAs
https://github.com/fastmachinelearning

hls 4 ml

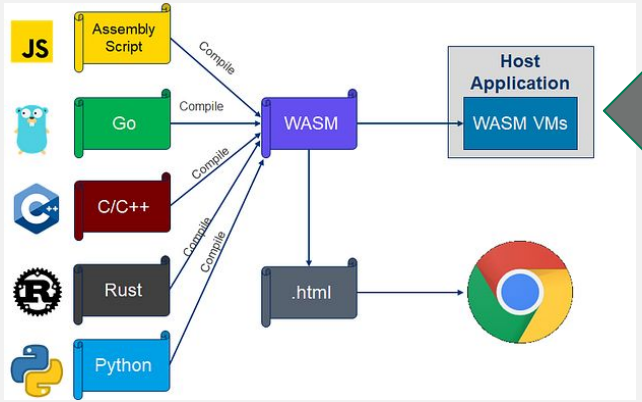
Conifer

Translates trained Boosted Decision Trees to FPGA firmware for extreme low latency inference.
https://github.com/thesps/conifer

FPGA - Programmable software

Linux

MQTT



https://medium.com/@bhavani.indukuri2/webassembly-wasm-revolutionising-web-development-with-high-performance-and-portability-e4aef76391bb

The Rust Programming Language

Firmware written in Rust

Userspace I/O

PL

VIVADO

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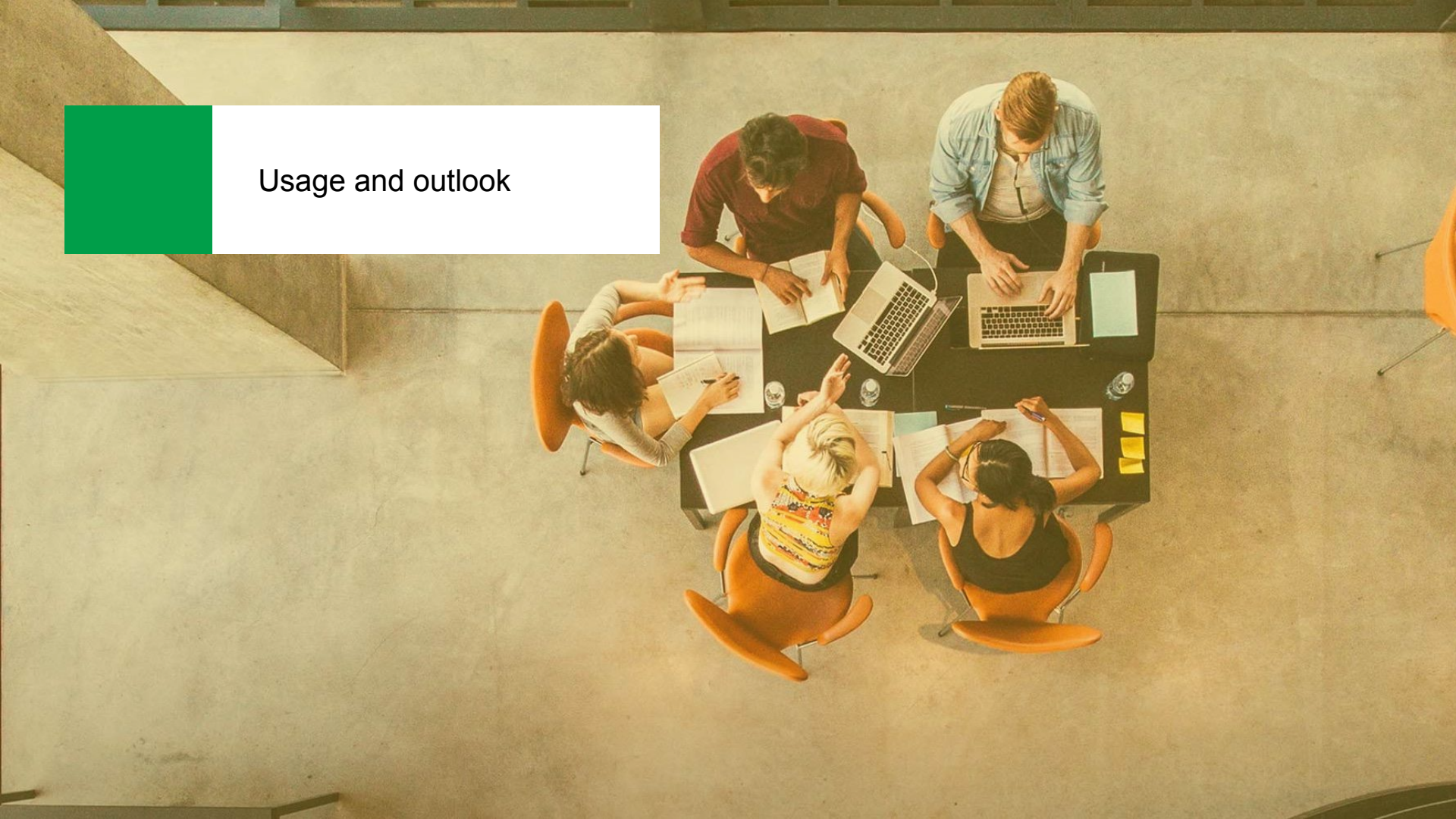
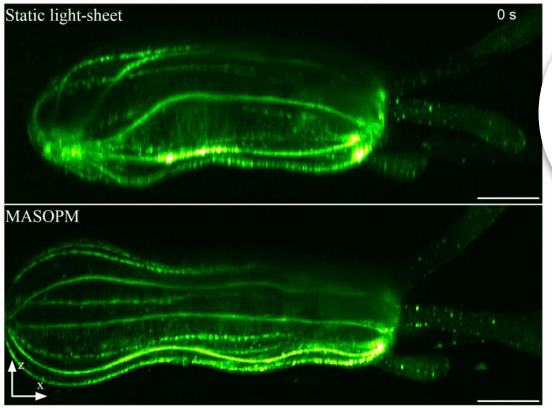
Usage and outlook



Collaborations & Plans

Oblique Plane Microscope (OPM)



Sam Davis,
Prevedel Lab
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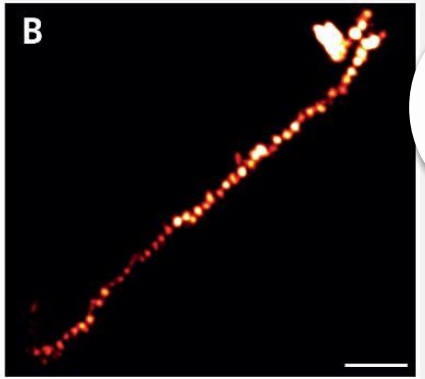
Prevedel, Robert; Davis, Samuel; Sommernes, Jon-Richard; Hambura, Sebastian; Riedel, Levin; Gil, Alejandro; et al. (2024). **A mesoscopic axially swept oblique plane microscope for imaging of freely moving organisms with near-isotropic resolution**. Optica Open. Preprint. https://doi.org/10.1364/opticaopen.26525872.v1

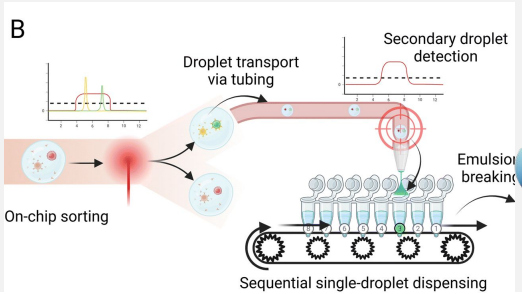
Superresolution (MINFLUX)



Ries Lab
Max Perutz Labs,
University of
Vienna

Fig. 2. MINFLUX tracking of kinesin in live cells.
Takahiro Deguchi *et al.*,
Direct observation of motor protein stepping in living cells using MINFLUX.
Science **379**,1010-1015(2023).
DOI:10.1126/science.ade2676

Fluorescence-activated droplet sorting



Weiss, M., Hasan, S., Genth, R., Mollah, M., Robert, E., Gil, A., & Hufnagel, L. (2025). A single droplet dispensing system for high-throughput screening and reliable recovery of rare events. *Lab on a Chip*. https://doi.org/10.1039/d4lc00536h

VERAXA

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Marian Weiss (Veraxa)

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