3D *ex vivo* imaging and characterization of neuromuscular junctions in rodent muscles

IMACTIV-3D

Tissues & Organs • Light Sheet • Advanced Media • Morphometric Quantification • Automatic Detection • Neurology

YOUR NEEDS

- Study of neuromuscular pathologies or atrophy models
- Preclinical evaluation of compound efficacy

OUR SOLUTIONS

- Light sheet microscopy and clearing to visualize neuromuscular junctions on whole muscle in 3D
- Automated image processing for quantification

General Procedure
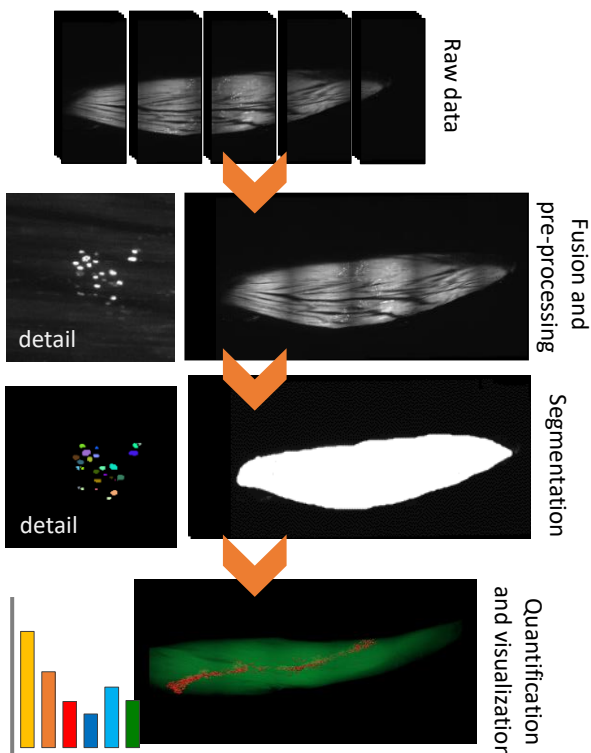
Prior to sample collection by Imactiv-3D:

- *In vivo* labeling by infusion with a fluorescent toxin
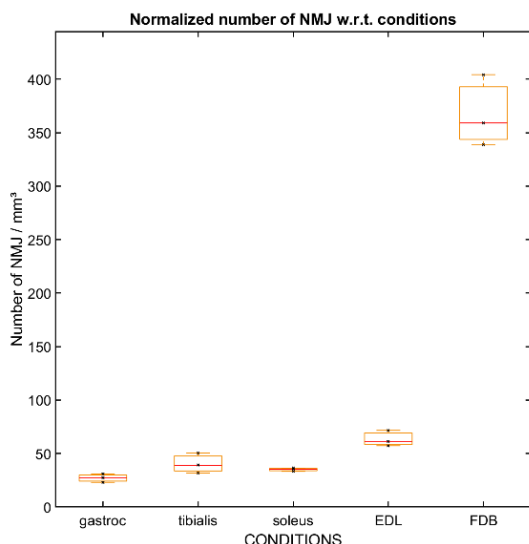- Formalin fixation of extracted sample

Image acquisition:

- Sample clearing
- 3D light sheet fluorescence microscopy
- Multi-position acquisition

Image processing and analysis:

- Quantitative characterization of all types of leg muscle:
  - Estimation of muscle volume
  - Absolute number and density of neuromuscular junctions
- 3D visualization with surface and volume rendering:
  - Reconstruction of the whole sample
  - Visualization of the spatial distribution of junctions
  - Advanced display using 3D animations

Raw data

detail — Fusion and pre-processing

detail — Segmentation

Quantification and visualization

Application example: *in collaboration with* **IPSEN** Innovation for patient care

Normalized number of NMJ w.r.t. conditions

(Y-axis: Number of NMJ / mm³; X-axis CONDITIONS: gastroc, tibialis, soleus, EDL, FDB)

- Aim: comparison of a wide range of muscle types in the rat leg in terms of neuromuscular junction density
- Technical specifications:
  - Whole sample acquisition using 3 to 10 fields of view
  - Number of junctions ranges from 3 000 to 20 000 depending on the muscle type
  - Junction average size is about 60 x 30 x 10 µm³
- Analysis of the neuromuscular junction density following this procedure on 30 muscles produced interesting results among different types of muscle