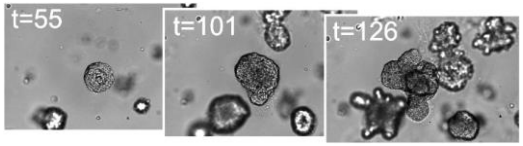
Segmentation and Tracking of Organoids in Brightfield Microscopy Image Data

Lucia Hradecká Dupkaničová | Advisor: Martin Maška
Faculty of Informatics, Masaryk University, Brno, Czech Republic

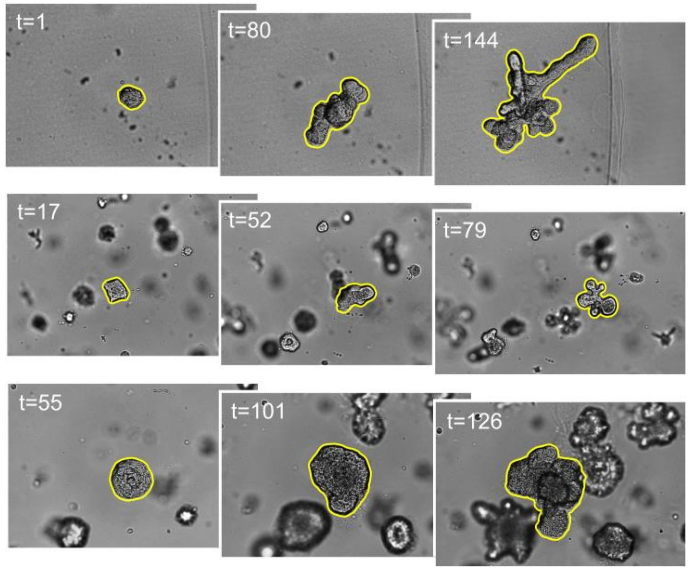
MOTIVATION

- **Organoids** are in vitro cultures that mimic living tissues and their dynamics, allowing studies of physiological processes in a controlled environment under various conditions. They are observed using **optical microscopy** that produces high-resolution time-lapse sequences. Their automated processing requires **reliable and robust algorithms** that are currently lacking in the field, thus having biologists to rely on manual processing of the acquired image data.
- **Common challenges** for automated processing of high-resolution organoid images include the presence of spurious objects, collisions, occlusions, and abrupt changes of the organoid location and texture due to manual interventions taken during long-term imaging.



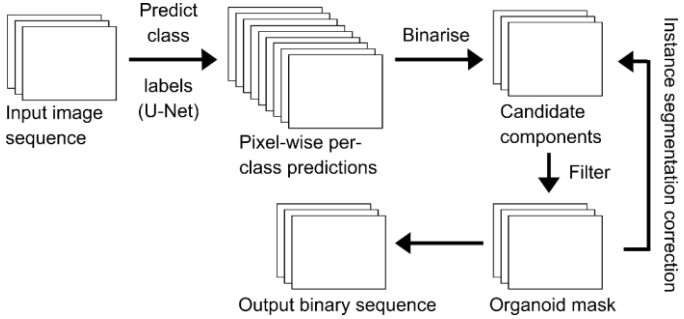
AIMS OF THE THESIS

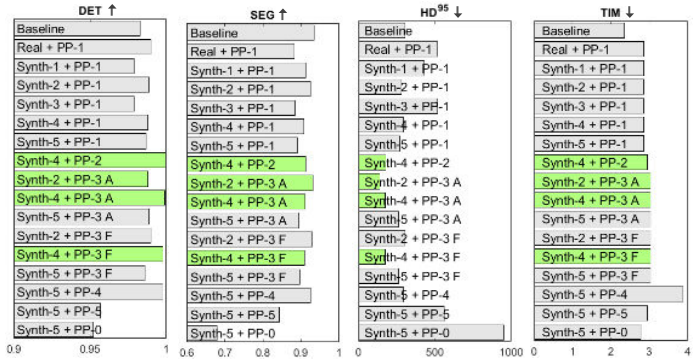
- To develop **a reliable method for automated segmentation and tracking of organoids** in high-resolution brightfield microscopy sequences
- To **validate the performance** of the method over a heterogeneous dataset of mammary epithelial organoids of five different phenotypes
- To deploy the method in **an easy-to-use form**



METHODOLOGY

- A family of **deep-learning-based** algorithms
  - Convolutional neural network (**U-Net** architecture)
  - **Tuneable** prediction **binarisation**
  - **Adaptive** morphological and component **filtering**
  - A novel **segmentation correction procedure**





RESULTS

- **Algorithm development**: real and computer-generated sequences of three distinct organoid phenotypes
  - Six semantic segmentation models *(Real, Synth-X)*
  - Five postprocessing routines *(PP-X)*
- **Algorithm validation**: real sequences of two additional organoid phenotypes with considerably different shape, size, and texture characteristics
- **Performance criteria**: single-organoid tracking accuracy (DET), segmentation accuracy (SEG), boundary localisation error (HD^{95}), and execution time (TIM)
- Recommended **four reliable and robust algorithms**
- An **easy-to-use application** with a practical guide
- One of the algorithms presented also in **a manuscript** that is under review for publication **in a top-tier journal**

CONTRIBUTION HIGHLIGHTS

- A group of **well-performing and robust algorithms** for segmentation and tracking of organoids was developed
- Their performance and robustness were **validated** using diverse image data with respect to different criteria
- **Implementation** of the developed methods is available for use in practise (https://is.muni.cz/th/hpuxk/)
- **First co-authorship of a journal article** presenting one of the recommended algorithms (https://cbia.fi.muni.cz/research/spatiotemporal/organoids.html)