# 1 Introduction

## 1.1 Motivation of current project

## 1.2 Importance of accurate cell segmentation

## 1.3 Using 3D data to increase contrast

## 1.4 Thesis outline

# 2 Cell segmentation

## 2.1 Basics of image manipulation

211 Example of digital images [211-digital\_image\_sample\_gfp-050714\_s13\_ch0\_t85\_z30]

[211-digital\_image\_sample\_bf-050714\_s13\_ch1\_t85\_z30]

212 Edge detection example [212-canny\_bf-050714\_s13\_ch1\_t85\_z30]

213 Blob example [213-blobs\_bf-050714\_s13\_ch1\_t85\_z30]

## 2.2 Basics of cell segmentation

221 Sqaussh example [231-squassh\_bf-050714\_s13\_ch1\_t85\_z30]

222 CP example [232-cp\_bf-050714\_s13\_ch1\_t85\_z30]

## 2.3 Studies that use cell segmentation

# 3 Preparing images for segmentation

## 3.1 3D Confocal imaging

311 Schematic of environment

## 3.2 Using GFP fluorescence data

321 GFP max projection image

## 3.3 Using Brightfield image data

331 Raw Brightfield example [211-digital\_image\_sample\_bf-050714\_s13\_ch1\_t85\_z30]

332 Brightfield in 3D

## 3.4 Review of study using Brightfield

341 Brightfield STD [341-bmod\_bf-050714\_s13\_ch-bmod\_t0\_z00]

# 4 Methodology

## 4.1 Definitions and assumptions

411 GFP profile [411-profile]

412 GFP profile scatter [412-scatter]

## 4.2 Generating zMod

421 zMod example [421-zmod\_example-050714\_s13\_ch-zmod\_t30\_z0]

422 zBF example [422-zbf\_example-050714\_s13\_ch-zbf\_t00\_z00]

## 4.3 Manual tracking

431 zComp [431-zcomp\_example-050714\_s13\_ch-zcomp\_t00\_z00]

432 Primary image [432-primary]

433 Primary image overlaid on Brightfield [433-primary\_with\_zcomp]

## 4.4 Generating zEdge for segmentation

441 zEdge example [441-zedge\_example-050714\_s13\_ch-zedge-8-5-5\_t0\_z00]

## 4.5 Sensitivity analysis of zMod parameters

451 Variation of zMod parameters (sigma)

452 Variation of zMod parameters (R)

# 5 Results

## 5.1 Image modification

5.1.1 zMod

5111 zMod example [5111-zmod\_example-050714\_s13\_ch-zmod\_t00\_z00]

5112 zMod 3D [5112-zmod\_3d\_1]

5113 zMod 3D [5113-zmod\_3d\_2]

5.1.2 zBF

5121 zBF example [5121-zbf\_example-050714\_s13\_ch-zbf-8-5-5\_t0\_z00]

5122 zBf bad focus [5122-zbf\_bad\_focus-260714\_s12\_ch-zbf\_t011\_z00]

5.1.3 zEdge

5131 zEdge example [5131-zedge\_example-050714\_s13\_ch-zedge-8-5-5\_t0\_z00]

## 5.2 Segmentation

5.2.1 GFP segmentation

5211 GFP segmentation example [5211-gfp\_max\_seg-tile\_050714\_s13\_ch-zbf-8-5-5-outline-zcomp-8-1-1-gmod-QBWO0G0U\_t0]

5.2.2 Brightfield variance segmentation

5221 Brightfield STD segmentation example [5221-bf\_std\_seg-tile\_050714\_s13\_ch-zbf-8-5-5-outline-zcomp-8-1-1-bmod-PLG5WADE\_t0]

5.2.3 zEdge segmentation

5231 zEdge segmentation example [5231-zedge\_seg-tile\_050714\_s13\_ch-zbf-8-5-5-outline-zcomp-8-1-1-zedge-8-5-5-TYOGE5XI\_t0]

# 6 Discussion

## 6.1 zMod sensitivity to parameters and the effects on zBF

612 zMod granular example [612-zmod\_granular\_example-050714\_s13\_ch-zmod-8-1-1\_t0\_z00]

613 Plot of zMod sensitivity to Sigma and R

## 6.2 zEdge

621 zEdge granular example [621-zedge\_granular\_example-050714\_s13\_ch-zedge-8-1-1\_t0\_z00]

## 6.3 Comparison of Brightfield variance and zEdge segmentation

611 Graph of zMod fScore vs bMod fScore

# 7 Conclusion

## 7.1 Conclusion

## 7.2 Further work

7.2.3 Edge completion