

## Contents

Preface

Acknowledgments

### 1. The Digital Cell Philosophy

Workflows and Pipelines

Using Spreadsheets for Experimental Data

Software for Digital Cell Biology

Focusing on Imaging

Golden Rules

### 2. Dealing with Data

Why Is Organization So Important?

Getting Organized

Experiment-Based Organization

Databases for Resources

Electronic Lab Notebooks (ELNs)

Databases for Imaging Data

Sharing Your Data Externally

Backing Up Your Data

Golden Rules

### 3. Imaging Data

Software Selection

Fiji

RStudio

What Is an Image?

Image Formats

Image Types

Multidimensional Image Files

Metadata

Image Transformation

Imaging Information

Trade-Offs in Imaging

Focus and Dealing with Drift

Phototoxicity and Photobleaching

Choice of Fluorophores

Dynamic Range

Golden Rules

4. Image Processing and Analysis

How to Analyze an Image

Tutorial: Quantifying Cell Protein Levels from Immunofluorescence Images

Segmentation

Further Segmentation Approaches

Image Filters

Gel Densitometry

Tutorial: Quantifying Bands on a Gel

How to Analyze a Movie

Tutorial: Counting Vesicles in Cells

Particle Tracking

Tutorial: Manual Particle Tracking

Tutorial: Automated Particle Tracking

Kymographs

Tutorial: Generating a Kymograph

Colocalization

Tutorial: Using R to Measure Colocalization over Time

Getting the Right Data Out of the Image

Validation

Back to Square One

Golden Rules

5. Statistics

Designing an experiment

What Is  $n$ ?

Golden Rules

Why Does  $n$  Matter?

Power Analysis for Cell Biologists

Basic Statistics That You Will Need

A Refresher of Summary Statistics

Always Plot Out Your Data

Descriptive Statistics

Statistical Tests

Compare One Group to a Value

Compare Two Groups

Comparing Three or More Groups

More Complicated Experimental Designs

Problems with Data

The p-Value

What It Really Means

Statistically Significant versus Biologically Significant

Effect Size

Golden Rules

6. Coding

Where to Start

Basic Principles: Workflow, Reproducibility, and Benefits

Mastering the Command Line

Getting Started with Coding

Variables and Strings

Arrays and Vectors

Loops

How to Write a Basic ImageJ Macro

Working on All Files in a Directory

Tutorial: Blinding Files for Manual Image Analysis

How to Write a Basic R Script for Analysis

What Can Go Wrong?

How to Validate and Check Your Data

Debugging

Getting Help

Getting Good

Ugly Code

Write Modular Code

Version Control and git

Sharing Your Code

Golden Rules

7. Putting It Together

Plotting Data

Best Practice

Making Figures

Best Practice

Making Figures That Look Great

Color Blindness

Contrast Adjustments

Crops and Expansion

Scale Bars

Movie Files

Unacceptable Manipulation in Figures

Golden Rules

Appendix: List of Software Featured in This Book

Bibliography

Index